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RESEARCH

The effects of reward uncertainty and alcohol intoxication on human sign- and goal-tracking

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Abstract

Rational Sign- and goal-tracking (ST/GT) behaviors capture individual differences in reactivity to reward-predictive cues and are increasingly recognized as translational markers of addiction vulnerability. While animal research shows that cue-reward uncertainty (i.e. the inability to predict whether a reward will follow its predictive cue) and alcohol modulate sign-tracking, empirical evidence in humans remains scarce. This experimental study investigated how acute alcohol intoxication and cue-reward uncertainty shape attentional responses to reward cues in humans.

Methods One hundred eighteen young adult participants were assigned to one of four groups in a 2 × 2 design: Alcohol (0.5 g/kg) vs. Placebo and Uncertain (50% reinforcement) vs. Certain (100% reinforcement) reward conditions. Eye-tracking during a Pavlovian conditioned approach (PCA) task quantified ST/GT tendency, followed by a non-rewarded "omission" phase.

Results Reward uncertainty significantly increased attentional allocation to the reward-predictive cue ($\eta^2 p = 0.04$), consistent with animal evidence that uncertain outcomes amplify responses to conditioned cues. Contrary to predictions, alcohol did not produce reliable enhancements of sign-tracking ($\eta^2 p = 0.01$). Effects during reward omission were mixed: the groups that previously experienced reward certainty increased their attention towards the sign while decreasing their attention towards the goal. Exploratory analyses incorporating initial ST/GT tendencies as covariates further indicated that cue-attention during omission aligned with participants' initial ST/GT behavior.

Conclusions Reward uncertainty, but not alcohol intoxication, affected sign-tracking tendencies. By directly translating animal findings to a human paradigm with eye-tracking, these results support the idea that uncertainty affects how individuals orient toward reward-predictive stimuli, a process relevant to theories of addictive behavior.

Keywords Reward uncertainty · Alcohol · Sign-tracking · Pavlovian Conditioned Approach

Introduction

Numerous theoretical frameworks of addiction have underscored the central role of Pavlovian conditioning in the development and maintenance of substance use disorders (Robinson and Berridge 1993; Everitt et al. 2001; Wiers and Stacy 2006). Reactivity to substance-related cues has mostly been discussed in relation to physiological (Wang et al. 2019), attentional (Ramirez et al. 2015), behavioral (Acker and MacKillop 2013), subjective (Blaine et al. 2019) and even neural responses (Courtney et al. 2016). All these effects result from the learning of a Pavlovian association between a conditioned stimulus (CS) and an unconditioned stimulus (UCS).

Over the past two decades, scientific research, primarily involving animal models, has increasingly focused on the study of "sign-tracking" and "goal-tracking," two forms of Pavlovian-conditioned responses that are highly relevant to addiction (Flagel et al. 2007; Robinson and Flagel 2009). When rats are exposed to the brief presentation of a CS lever reliably followed by the delivery of a reward UCS (e.g., a

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food pellet) at a different location, they tend to adopt one of two distinct conditioned response patterns. Sign-trackers (STs) preferentially approach and engage with the CS, which not only predicts the reward but also seems to have acquired its motivational properties. In contrast, goal-trackers (GTs) direct their behavior toward the location of the expected reward, treating the cue primarily as a predictive signal only and not a motivational stimulus. Finally, some individuals, referred to as intermediates (INTs), display a mixed response pattern, alternating between sign- and goal-tracking behaviors. These phenotypic differences are thought to reflect differing propensities to attribute incentive salience to reward-predictive cues (Robinson and Flagel 2009). The ST and GT phenotypes show stable individual differences in traits such as impulsivity (Lovic et al. 2011), attentional control (Paolone et al. 2013), and dopaminergic reactivity to reward cues (Flagel et al. 2007, 2011; Robinson and Flagel 2009).

In rats, the ST phenotype has been associated with increased vulnerability to addiction compared with the GT phenotype, including a higher responsiveness to food and drug cues, enhanced drug self-administration, heightened cue-induced craving, and a greater risk of relapse (Berridge 2007; Flagel et al. 2008, 2009, 2010; Tomie et al. 2008; Saunders and Robinson 2010, 2011; Saunders et al. 2013; Robinson et al. 2014; Anselme and Robinson 2020). STs have also been shown to be more resistant to appetitive extinction and less behaviorally flexible than GTs (Ahrens et al. 2016; Fitzpatrick et al. 2019). During extinction phases, when the previously rewarded stimulus is no longer paired with reinforcement, GTs rapidly adjust their behavior by ceasing to approach the reward-associated location. In contrast, STs persist in approaching the cue-associated location for a significantly longer period, indicating a stronger cue-driven motivational bias and diminished adaptability to changes in reward contingencies. Although ST and GT phenotypes are considered relatively stable traits, with evidence pointing to a genetic basis (e.g., (Flagel et al. 2010), there is also evidence that they are not immutable. The ability to express one or the other profile notably depends on the type of CS (e.g., lever vs. tone; Meyer et al. 2014) and the consistency of the CS-UCS association (Robinson et al. 2015) (see below).

The translation of sign- and goal-tracking to human research is more recent and has only received systematic attention in the last few years (Anselme and Robinson 2020; Colaizzi et al. 2020; Heck et al. 2025a). In human studies of sign- and goal-tracking, a variety of experimental paradigms have been employed to assess individual differences (for a recent comprehensive overview, see Heck et al. 2025a). These studies include computerized Pavlovian Conditioned Approach (PCA) tasks paired with eye-tracking to assess attentional patterns (e.g. (Garofalo and di Pellegrino 2015; Schad et al. 2020; Cherkasova et al. 2024; Dinu et al. 2024; Heck et al. 2025b), as well as physical PCA tasks, which either incorporate eye-tracking (Cope et al. 2023) or rely on direct behavioral measurements of interaction with the sign (cue) and goal (reward) locations (Colaizzi et al. 2023).

Despite considerable variability in experimental methods and analytical approaches among studies (Heck et al. 2025a), human research broadly suggests that it is possible to identify individuals who exhibit sign-tracking and goal-tracking (and in some cases, intermediate) behavioral phenotypes. Importantly, these phenotypes have sometimes been linked to translationally relevant variables such as impulsivity (e.g., Garofalo and di Pellegrino 2015; Colaizzi et al. 2023; Cope et al. 2023) and alcohol use (Albertella et al. 2021; Watson et al. 2024; Heck et al. 2025b). Recent theoretical work has proposed a direct connection between incentive salience attribution and human sign-tracking, particularly through value-modulated attentional capture (VMAC) tasks, which suggest that reward-associated cues gain motivational significance and are more likely to capture attention, thereby increasing their influence on behavior (Le Pelley et al. 2024). Supporting this framework, fMRI evidence indicates differential incentive salience attribution in human sign- versus goal-trackers during Pavlovian-to-instrumental transfer (PIT) tasks (Schad et al. 2020). However, empirical studies on human sign- and goal-tracking currently still remain scarce in comparison to animal studies, and null findings have also been reported, emphasizing the need for methodological refinement and conceptual clarity in this emerging field (see Heck et al. 2025a).

Given that a CS acquires its incentive salience through its association with the UCS, one should expect that a fully consistent CS–UCS association (100% predictive of reward) enhances the CS's incentive salience and approach behavior more than an inconsistent one (only 50% predictive of reward). However, animal research has demonstrated that rats exposed to reward uncertainty increase the strength of sign-tracking, especially when it is combined with uncertainty in reward magnitude (Anselme et al. 2013; Anselme, 2015; Robinson et al. 2014), as well as the number of individuals adopting this profile (Robinson et al. 2015). Other factors that may contribute to increase reward uncertainty, such as long and variable inter-trial intervals, also increase sign-tracking (Lee et al. 2018; Kaneko et al. 2025). Importantly, dopamine signaling has been suggested being implicated in the effects of uncertainty during Pavlovian conditioning (Hart et al. 2015).

In human research, similar motivational/attentional effects have been obtained under the framework of *Uncertainty-Modulated Attentional Capture* (UMAC). UMAC refers to the tendency for reward-associated distractors to capture attention more strongly when the outcome is uncertain (Le Pelley et al. 2019; Cho and Cho 2021; Ju and Cho 2023; Massa et al. 2024; Pearson et al. 2024; Chow et al. 2025). This effect appears to be both rapid and automatic (Le Pelley et al. 2019) and it persists even when participants are explicitly informed about reward contingencies, suggesting that it does not rely on strategic or goal-directed learning (Chow et al. 2025).

In addition to uncertainty, pharmacological factors may also influence the propensity to engage in sign-tracking behavior. Among these, alcohol administration has received particular attention, as it has been shown to enhance sign-tracking tendencies in animal models. This effect may be mechanistically linked to alcohol's ability to increase dopaminergic activity in mesolimbic pathways (Boileau et al. 2003; Didone et al. 2016). For instance, Tomie and colleagues (Tomie et al. 1998a, b) demonstrated that ethanol administration significantly increased sign-tracking behavior in rats across a range of doses. These results support the broader hypothesis that sign-tracking reflects a form of impulsive responding (Tomie et al. 1998a, b), a trait frequently implicated in addictive behaviors. Evidence from both human and rodent research indicates that acute alcohol administration enhances impulsivity, likely through its effects on inhibitory control (Poulos et al. 1998; de Wit et al. 2000; Olmstead et al. 2006; Dougherty et al. 2008; Adams et al. 2012).

In humans, priming doses of alcohol have been shown to heighten attentional biases toward alcohol-related cues (Miller and Fillmore 2011; Nikolaou et al. 2013), although some studies have reported a decrease in attentional biases following consumption, possibly due to satiation effects (Weafer and Fillmore 2013; Monem and Fillmore 2019). Beyond alcohol-specific cues, cross-priming effects have been observed: alcohol administration can enhance attentional biases toward other substance-related cues, such as those associated with cocaine or nicotine (Field et al. 2005; Montgomery et al. 2010).

Collectively, these findings suggest that alcohol may not only heighten attention toward alcohol-relevant stimuli, but also amplify a more general reward sensitivity. This may occur through alcohol's capacity to increase impulsive responding, ultimately exacerbating behavioral biases toward salient cues (whether expressed as cue-driven sign-tracking behavior or attentional bias) across both human and nonhuman models.

From an attentional perspective, the Alcohol Myopia Theory[1] (Steele and Josephs 1990) offers two complementary interpretations for the effect of alcohol on cue attention (as pointed out by Watson et al. 2020). First, alcohol could narrow attentional capacity globally, reducing processing of peripheral or distracting stimuli such as a conditioned stimulus (CS) in a sign-tracking paradigm. Alternatively, alcohol may selectively focus attention on highly salient cues (Steele and Josephs 1990), which would predict an increase in sign-tracking-like behaviors. This has been examined in a VMAC study in humans. Watson et al. (2020) tested this bi-directional hypothesis and found that moderate alcohol intoxication enhanced attentional performance in a VMAC task: intoxicated individuals earned more rewards because they were less distracted by high-value predictive stimuli, suggesting that alcohol can alter attentional prioritization in complex ways. It is important to note, however, that the task structure in Watson et al.'s study differed substantially from that of the present investigation. In their paradigm, attending to the reward-predicting cue resulted in reward omission, a contingency that was not present in our design.

The aim of this study was to advance the validation of the sign-tracker/goal-tracker (ST/GT) model in humans by experimentally examining two key modulators of reward processing: reward uncertainty and acute alcohol administration. Building on robust evidence that uncertainty enhances attentional engagement and cue-driven approach behavior, we sought to determine whether it also modulates the tendency to sign-track in our previously developed PCA task (Heck et al. 2025b). Both factors have been independently linked to dopaminergic signaling, which plays a central role in incentive salience and the tendency to sign-track versus goal-track. Given these converging mechanisms, we hypothesized that alcohol and uncertainty would exert additive effects, jointly amplifying cue-directed attention (sign-tracking) relative to goal-directed attention.

Therefore, this study tested whether either factor (uncertain reward delivery or a moderate alcohol dose [0.5 g/kg]) would enhance attentional allocation to a reward-predictive cue. We hypothesized that participants exposed to both alcohol and uncertainty would exhibit the strongest bias toward the cue compared to those experiencing only one or neither factor. Additionally, we explored whether exposure to uncertainty and/or alcohol would impact behavior under reward omission. This prediction was grounded in three considerations: (1) decades of research demonstrate that partial reinforcement reliably enhances extinction resistance; (2) sign-tracking individuals typically persist longer during extinction, and both manipulations—uncertainty and alcohol—were expected to promote sign-tracking tendencies; and (3) alcohol may further augment extinction resistance through its documented effects on inhibitory control. To assess this, a non-reinforced "Omission" phase was included at the end of the PCA task. To our knowledge, this is the first experiment to examine the joint effects of reward-cue uncertainty and alcohol administration on

[1] Originally designed to explain extreme social behavior under alcohol intoxication, this theory has expanded to cognitive aspects.

human sign-tracking behavior, representing a preliminary but hypothesis-driven step in this line of research.

Methods

Participants

This study involved 118 young adult volunteers (74 females, 44 males; Table 1). Participants were first invited to complete an online screening questionnaire to assess eligibility. Inclusion criteria required: no history of severe neurological disorders; no regular use of substances (defined as use more than twice in the last two months, excluding tobacco and alcohol); no diagnosis of ADHD currently treated with medication; no color vision deficiency; and being at least occasional alcohol consumers (i.e., individuals who were fully abstinent were excluded). Additional exclusion criteria were specific to the alcohol administration component of the study and included: being pregnant, having medical conditions that contraindicated alcohol consumption, taking medications incompatible with alcohol use, and having experienced allergic or adverse reactions following alcohol consumption. All participants had normal or corrected-to-normal vision. Table 1 shows means and standard deviations for age, education level, alcohol consumption severity score (AUDIT-C), and recent drinking index in the present sample.

A priori power analysis for ANOVA, with an alpha level of 0.05, power of 0.80, and a medium effect size led to $n=128$ (32/group). We added approximately 20% to the projected sample size, due to loss of participants given the data quality criteria for our eye-tracking based ST/GT score (projected $N=152$, 38/group).

Initially, 139 participants took part in this study. Eighteen participants were removed from the analysis due to problems with one of the data quality check criteria (see "Eye-Tracking" below). Two were removed because they reported an AUDIT score = 0 at in-person testing. One participant was excluded because this individual also took part in previous studies of the same project. The final sample therefore included 118 participants for the main analysis.

Table 1 Sample Description

Variable	Median (std)	Min-Max
Age	21.00 (2.93)	18–33
Education Level	14.00 (1.99)	10–23
AUDIT-C	5.00 (2.49)	1–10
Recent Drinking Index	9.00 (55.76)	0–400

Note. Recent drinking index = number of alcoholic drinks consumed during the last 2 weeks x number of drinking occasions during the last 2 weeks

Participants were recruited via word of mouth, social media or advertising posters. The study was initially explained to the participants as investigating "individual variability in the impact of alcohol on reaction times". After filling out the online screening, participants meeting the inclusion criteria were contacted to take part in the experimental phase of the study (on-site). All participants gave informed consent to take part in the study. Recruitment and testing took place from November 2024 to June 2025, at the university's facilities. The study was conducted in accordance with institutional guidelines and the Declaration of Helsinki. It was approved by the local Ethics Committee.

General procedure

All participants were informed that they would receive varying quantities of alcohol in a mixed beverage. This approach was selected due to the inherent challenges in achieving effective blinding in completely balanced alcohol administration studies. Specifically, individuals who receive alcohol are likely to experience noticeable physiological and subjective effects, which can lead them to accurately infer their group assignment—thus undermining the credibility of an "expecting alcohol/receiving placebo" condition. Several studies have highlighted the difficulty of maintaining successful blinding in alcohol research, even with modest doses (Ross and Pihl 1989; Martin and Sayette 1993). Given these constraints, our partially informed design aimed to balance ethical transparency with methodological rigor. Prior to attending the in-person testing session, all participants were provided with the following instructions to ensure safety and data integrity: (1) refrain from consuming alcohol the evening prior to the session, (2) abstain from eating for at least five hours before the session, (3) bring a valid form of identification to allow verification of their age, and (4) bring a personal item (e.g., a book) to occupy themselves at the end of the experiment in case they experienced a waiting period due to excessive intoxication.

In-person testing was conducted in a quiet, private room and consisted of three main phases: (1) informed consent, alcohol/placebo administration, and pre-experimental questionnaires (approximately 15–20 min); (2) completion of the PCA task (~20 min); and (3) post-experimental questionnaires, a full debriefing, and a second informed consent (~15 min). The third phase was followed by a potential washout period, if required. Short breaks were allowed between phases to minimize participant fatigue. At the end of the session, all participants were fully debriefed about the true purpose of the study and their actual group assignment. In total, four groups were tested: Alcohol + Uncertainty, Alcohol + Certainty, Placebo + Uncertainty, and Placebo + Certainty (see below for details). Group comparability regarding alcohol consumption variables was verified, confirming that no confounding factors influenced the outcomes despite randomization (see Supplementals S1).

Alcohol/placebo administration

The beverage administration procedure was identical for participants in the Alcohol and Placebo groups, the only difference being the presence or absence of alcohol. To begin with, all participants completed a breathalyzer test to ensure a zero baseline blood alcohol concentration. Participants' IDs were then checked to verify that they were over 18 years of age. Each participant was subsequently weighed—calibrated for light or heavy clothing when necessary—to calculate the beverage volume required to reach a target blood alcohol concentration (BAC) of 0.5 g/kg. For practical reasons, experimenters were not blinded, as they were responsible for preparing the beverages. However, given the minimal researcher involvement in the experimental protocol, which was largely computerized, this was deemed acceptable.

The alcoholic beverage consisted of Captain Morgan Spiced Gold Rum (35% alcohol by volume), mixed with cola and lime juice. The placebo beverage was prepared identically, using Captain Morgan Spiced Gold 0.0% (non-alcoholic) instead. Following established procedures in alcohol administration research (Rohsenow and Marlatt 1981), all drinks were prepared in the participant's presence using branded bottles, to enhance the credibility of the placebo. Participants were instructed to consume the entire beverage within five minutes to standardize the timing of alcohol absorption. This was followed by a 10-minute absorption period during which participants completed questionnaires (see Questionnaire section for details).

A second breathalyzer test was conducted after this waiting period, with participants blinded to the results. At the end of the experimental session, a final breath alcohol measurement was taken to ensure that any participant intending to drive was within the legal limit. Mean breath alcohol concentration (BrAC) increased significantly from before the experimental tasks (M = 0.18, SD = 0.06) to after the PCA tasks and questionnaires (M = 0.22, SD = 0.06; $t_{(56)} = 4.81$, $p < 0.001$).

PCA task

Before starting the ST/GT task, participants were instructed that they could collect a certain number of points in the next tasks, and that more points collected would unlock more glorious expertise badges.

Points corresponded to virtual coins collected on each trial (1- or 10-point coins). Performance was defined as the total number of coins collected and was converted into badge levels using predefined percentage intervals: Bronze (0–25%), Silver (25–40%), Gold (40–55%), Platinum (55–70%), Diamond (70–85%), and Expert (85–100%). The variation of coin magnitude (1 vs. 10 points) was implemented to maintain curiosity and reduce task monotony, rather than induce differential performance.

The stimuli were presented on a computer screen (1920 × 1080 pixels) with a grey background. A battery of two computer tasks was implemented: the simple reward task and the PCA task. The tasks were run on a homemade custom Matlab software (version R2019b).

Briefly, the simple reward task involved a fixed blue rectangle filled with blue squares (goal location) on the screen. Goal location was counterbalanced across participants (bottom or upper part of the screen). The coin appeared for 1.5 s on the goal shape, and the sound of a falling coin could immediately be heard when the participant pressed the key to collect the reward. There was a 20% chance that the coin was a "10-point coin" to maintain a level of curiosity and avoid monotony during the task. The aim of this first task was for the participants to learn where to collect the rewards and was designed to mimic the magazine training phase in animal models, where rewards are regularly delivered without any CS presentation. The simple reward task consisted of 10 trials, with a variable inter-trial interval (ITI) from 4 to 8 s. Prior to starting the task, participants were instructed as follows: "*During this task*, *objects will be displayed in a blue rectangle. These will allow you to earn points. Your objective is to respond as quickly as possible to their presentation by pressing the "enter" key*."

The PCA task was an adapted version of the autoshaping procedure used in animal ST/GT studies. It consisted of two blocks of 20 trials with a variable ITI ranging from 4 to 8 s. Like in the previous task, there was a 20% chance that a "10-point coin" was obtained. The goal shape was permanently present and could be located at the bottom or upper part of the screen (the location being the same as in the simple reward task). The sign shape was displayed on the opposite part of the screen (upper or bottom) for 3 s and disappeared just before a coin was presented on the goal shape for 3 s. Therefore, the sign shape was defined as a CS announcing reward delivery with a given probability. Two experimental groups were tested: one experienced a 50% CS–reward probability (uncertainty), the other a 100% probability (certainty). Participants collected rewards by pressing "enter," triggering a coin sound. Prior to starting the PCA task, participants were instructed as follows: "*This task is similar to the first one*, *except that we are no longer measuring your reaction time here. You can also score points. Use the "enter" key as before. Please keep your attention on the computer screen and don't look at the keyboard*."

In the PCA task, task parameters ensured that all participants reached the highest (Expert) badge level, irrespective of point magnitude condition, such that no meaningful differences in total badge value were observed between groups. Badges therefore served as a symbolic incentive and were not used as outcome measures. The primary focus of the task was the assessment of attentional allocation between the coin (goal-related) location and the sign (cue) location.

Figure 1 (reproduced from Heck et al. 2025b) depicts the layout of the newly developed computerized PCA task alongside the traditional animal PCA paradigm, illustrating design elements that were intended to approximate key features relevant to cross-species comparisons of sign-tracking behavior.

Following the PCA task, participants completed a so-called reward "omission" block that closely mirrored the original design but consisted of 15 trials in which the CS was presented for 3 s without the opportunity to earn any reward or coin. Participants were informed only that this task resembled the previous one. No explicit instructions regarding reward omission were provided, as in animal studies, the sudden transition to the omission phase being likely to be surprising.

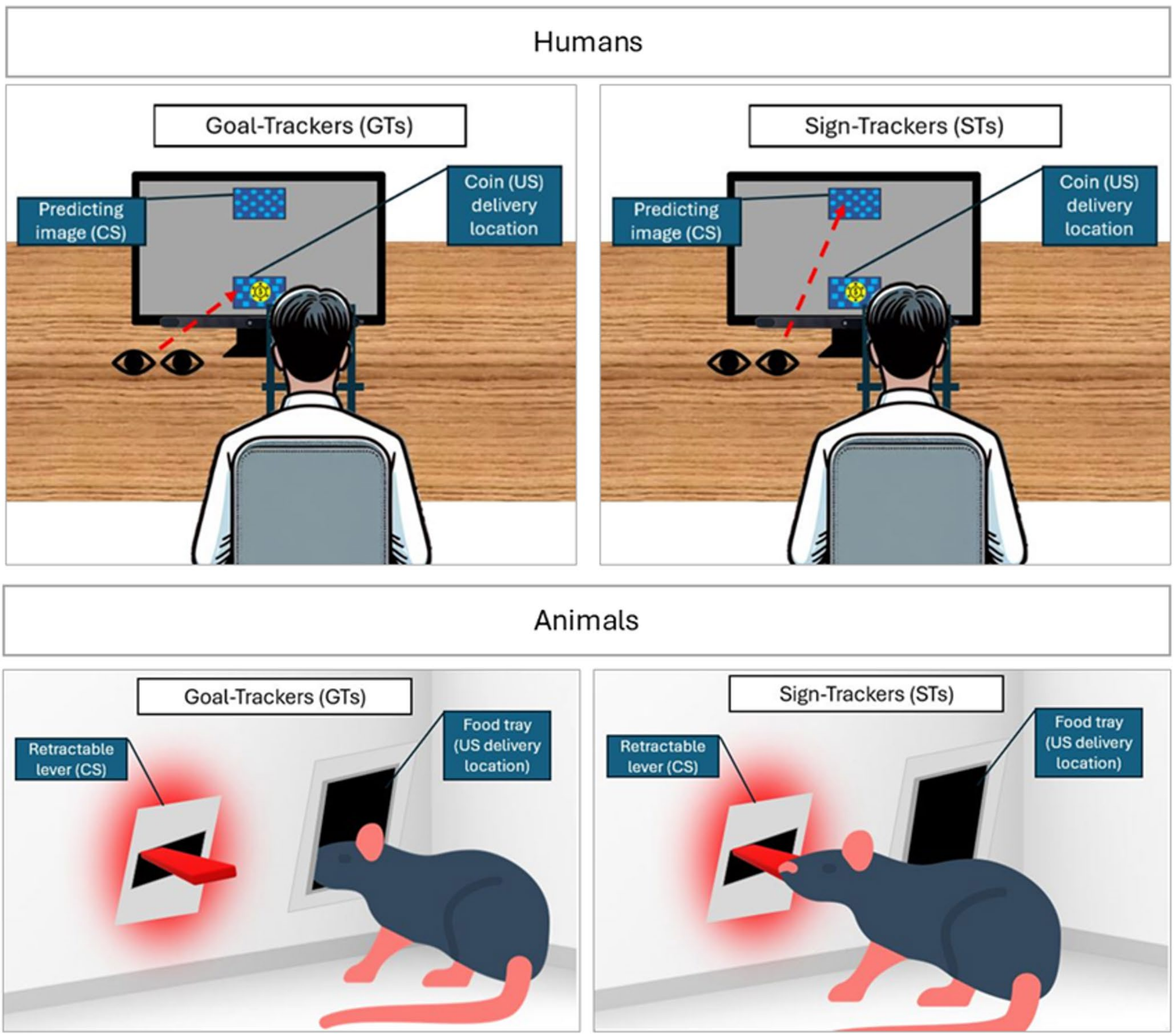

Fig. 1 Layout of the Newly Developed Computerized Pavlovian Conditioning Approach (PCA) Task in Comparison to the Original Animal Task. Note. The lower part of the figure shows the traditional animal PCA paradigm (adapted from Kuhn et al. 2018), while the upper part represents our newly developed task in humans. The person is seated in front of a computer screen, with their head in a chinrest. The task consists of multiple presentations of a predicting cue and a reward cue (coin). Approach behavior is inferred from eye-gaze behavior. Reproduced with permission from American Psychological Association. No further reproduction or distribution is permitted

Questionnaires

In the screening phase, all participants completed sociodemographic online questionnaires including the history of neurological problems that could affect cognitive functioning, and regular use of tranquilizers or illicit drugs. They also completed the short version of the alcohol use disorder identification test (AUDIT-C, Bush et al. 1998).

In the pre-experimental phase, all participants completed electronic versions of the short version of the French Impulsive Behavior Scale (UPPS-P, Billieux et al. 2012). Additional questionnaires were included but not relevant to the present analyses: The Edinburgh handedness inventory (Oldfield 1971), which assesses handedness through 22 items. The 11-item version was used. The Karolinska Sleepiness Scale (KSS, Kaida et al. 2006), which measures the subjective level of fatigue at a particular time of day based on a nine-point Likert-type scale, was also administered.

Between the PCA task and the Omission block, participants completed electronic versions of alcohol subscale of the Drug Effects Questionnaire (DEQ, Morean et al. 2013). A 100 points visual analog scale was used to assess four dimensions of alcohol's subjective effects: "Feel", ("Do you FEEL any drug effects right now?"), "High" ("Are you HIGH right now?"), "Like" ("Do you LIKE the effects you are feeling right now?"), "More" ("Would you like MORE of what you consumed, right now?").

In the post-experimental phase, participants completed electronic versions of the following questionnaires:

- *The Brief Assessment Tool for Compulsivity Associated Problems* (BATCAP, Albertella et al. 2019), which assesses a range of compulsive symptom types. It covers alcohol use, gambling, binge eating and excessive Internet use (as well as contamination, checking, and ordering compulsions). In this study, we only used the addiction-related aspects.
- *Questions about current alcohol consumption*. To gain insight into participants' recent drinking behavior, we included the following two questions into the protocol: "How many drinks have you had in the last 14 days?" and "On how many occasions have you drunk alcohol in the last 14 days?". A recent drinking index was computed by multiplying these two numbers. We also measured the following item: "How many times have you engaged in binge drinking in the past month? (Defined as consuming > 5 drinks within 2 hours for men and > 4 drinks within 2 hours for women.)".
- *Alcohol Consumption*. The French version (Gache et al. 2005) of the Alcohol Use Disorders Identification Test (AUDIT; Saunders et al. 1993) consists of 10 multiple-choice items measuring alcohol consumption, alcohol dependence, and alcohol-related problems. Higher scores reflect a high probability of harmful alcohol use.

Eye-tracking

A Tobii 5 eye-tracking system (with an image sampling rate of 133 Hz) was mounted on a 60.5 cm diagonal computer screen. The eye tracker was calibrated using a six-point procedure at the beginning of the experiment. A chinrest was used to standardize the head-screen distance at 70 cm. Tobii Experience software was used to calibrate the eye-tracking system.

To compute eye gaze-based ST/GT scores, the screen was virtually separated into 9 regions of interest (ROI, 3 equal parts on each axis). Eye gaze position was collected throughout the PCA task. The two main ROIs used to compute the scores were the upper center and the bottom center of the screen, where the goal and the sign stimuli were located. We used a custom implementation of the Velocity Threshold Algorithm for fixation identification. First, eye position in pixels was converted to visual degrees. Then velocity was computed in degrees/sec for axes X and Y. To distinguish fixations from saccades, we used individualized thresholds for each subject based on their gaze velocity during mandatory fixations in an additional training task. Each threshold was computed by adding 1.95*standard deviation to their mean gaze velocity during fixations (= confidence interval of 95%). Eye movements were considered fixations when they were strictly below the personalized threshold. Too short fixations (< 100 ms) were removed to smoothen eye-gaze data. For each identified fixation, duration and ROI were collected.

As in our previous study (Heck et al. 2025b), analyses were conducted separately for the first and second blocks of the ST/GT task (each consisting of 20 trials). To focus on the most sensitive and reliable measure, only scores based on Dwell Time (total time spent looking at the ROI) are reported in the present manuscript. However, the full dataset (including measures such as number of fixations) is openly available and can be consulted by interested readers via our public repository.

Three main criteria were used for data quality check and poor quality data exclusions: A participant was excluded (1) if the gaze stagnated in the middle zone throughout the whole task, without variation (a possible indicator of poor calibration or other technical problem: $n = 0$), (2) if there were more than 30% of gazes at the outer edge of the screen (5% of the screen) compared to the rest of the screen (poor calibration or unfocused participant: $n = 0$ for the main PCA task and $n = 1$ for the Omission Block), and (3) if on average the individual gazed less than 1 s per CS presentation at either of the ROI (goal and sign) or at the center of the screen (due to too few data to compute a meaningful sign-tracking score: $n = 18$ for the main PCA task and $n = 4$ for the Omission Block).

Eye-tracking based ST/GT scores were computed similarly to the response bias scores classically used in animal research (e.g., Meyer et al. 2012). Dwell time on the “goal” was subtracted from dwell time on the “sign” and divided by the total dwell time on both ROIs (during the 3 s CS presentation). Thus, the ST/GT score ranged from −1 to 1, with negative scores theoretically suggesting a “goal-tracking” behavior, positive scores pointing towards a “sign-tracking” behavior, and scores around 0 meaning a rather intermediate tendency. This score was computed for the first and second blocks of the PCA task using Dwell Times.

Data analysis

Statistical analyses were performed using R 4.4.1 (R Core Team, 2024). The following packages were used: basic data import, processing, and visualization were performed using readxl (Wickham et al., 2025), tidyverse (Wickham et al. 2019), and ggpubr (Kassambara 2025a). ANOVAs and related analyses were conducted with rstatix (Kassambara 2025b) and car (Fox and Weisberg 2019). Effect sizes were computed using effectsize (Ben-Shachar et al. 2020), and visualizations were assembled with patchwork (Pedersen 2025). Correlation analyses were carried out with psych (Revelle 2025). Data transformations were performed using rnomi (McCaw 2023). Mixed-effects models and pairwise comparisons were conducted using lme4 (Bates et al. 2015), lmerTest (Kuznetsova et al. 2017), and emmeans (Lenth et al. 2025). Alpha level was set at 0.05, but corrections for multiple correlations were applied when needed. Given that the questionnaires were administered electronically, participants were required to answer all questions to proceed—there was no missing data.

As a preliminary step, we compared the Alcohol and Placebo groups on subjective responses to the beverage using the subscales of the Drug Effects Questionnaire (DEQ). Because the DEQ scores were not normally distributed, we conducted non-parametric Mann–Whitney U tests to verify the effectiveness of the experimental manipulations and ensure that the placebo beverage elicited convincing subjective effects.

The first main hypothesis was that both alcohol intoxication and reward uncertainty would significantly influence sign-tracking tendencies, with each factor expected to increase the ST/GT score, and their combination leading to the highest sign-tracking scores. To test this, we conducted a two-way ANOVA including the interaction between alcohol and uncertainty. Since the assumptions of normality and homoscedasticity were not initially met, the ST/GT scores were transformed using an Inverse Normal Transformation (Blom’s method). This transformation successfully corrected the distribution, resulting in both normality and homogeneity of variance being satisfied.

The second hypothesis was exploratory and tested whether alcohol intoxication and reward uncertainty would significantly affect cue-attention during a subsequent non-reward (so-called reward omission). During the “reward omission” phase, the goal rectangle (the location where coins were previously delivered) remained visible, analogous to the food magazine in rodent studies. We anticipated that “rational actors” might continue to monitor the goal location to some extent in expectation of coins, given the absence of explicit instructions about reward omission. For this reason, we focused on dwell time towards the sign as the primary variable of interest: continued attention to the sign despite the absence of reinforcement was interpreted as “resistance to reward omission”. Thus, during reward omission, we expected a more pronounced reduction in sign-directed attention in the 100%–placebo group, with weaker decreases in the other groups. Additionally, we also examined goal-directed attention, although we did not hold specific a priori predictions regarding the direction of these effects.

1. To investigate whether behavioral extinction took place in the non-rewarded phase compared to the acquisition block (Block 1 was chosen here to have the purest effect of uncertainty), we conducted linear mixed-effects analyses examining sign- and goal-directed attention across the three experimental blocks (Block 1, Block 2, and Omission), including Group (Alcohol vs. Placebo), Certainty (50 vs. 100), and their interactions. Pairwise comparisons between blocks were then performed in each group, with p-values adjusted for multiple testing using the Tukey correction.
2. We further investigated the relationship between individual differences in sign-tracking tendency and attentional allocation to the sign cue during the reward omission phase. Specifically, we tested whether higher ST/GT scores from Block 1 were associated with increased dwell time on the sign cue during omission, using a Spearman correlation.
3. To complement this analysis, we conducted a multiple linear regression including initial ST/GT tendencies (Block 1 scores) as a continuous predictor. The model tested the main and interactive effects of Group (Alcohol vs. Placebo), Probability (50% vs. 100%), and mean-centered ST/GT scores on Sign Dwell Time during reward omission. Mean-centering the continuous predictor reduced multicollinearity and facilitated

interpretation of interaction terms. All resulting variance inflation factors were below 5, indicating an acceptable level of multicollinearity. Some degree of multicollinearity was expected by design, as both alcohol intoxication and reward uncertainty were hypothesized to influence directly the third predictor, i.e. the ST/GT score. Thus, the predictors in the model were not fully orthogonal, which reflects the theoretical and experimental rationale behind the manipulation of these variables. Centering ST/GT scores allowed us to isolate more clearly and interpret more carefully the unique and interactive contributions of each factor despite their anticipated overlap.

In line with common practices in the sign-tracking/goal-tracking literature and to better capture potential interactions between ST/GT behavior and contextual manipulations, participants were also categorized into three discrete phenotypic groups (sign-trackers (ST), goal-trackers (GT), and intermediates (INT)) based on a tertile split of their Block 1 ST/GT scores (entire sample). These analyses are reported for exploratory and comparative purposes in the supplemental information (S2). S2 also presents a linear mixed-effects model assessing trial-by-trial changes in attention allocated to the goal location, included to confirm that participants showed comparable learning of the CS–UCS contingency.

Additional exploratory analyses

In a previous study (Heck et al. 2025b) using the same PCA task, we observed a significant positive association between AUDIT-C scores and ST/GT scores (r = 0.29), as well as a trend toward significance between ST/GT scores and the positive urgency dimension of impulsivity (r = 0.25). In the present study, replicating these findings is not straightforward due to methodological constraints: the four experimental groups differed not only in terms of beverage content (alcohol vs. placebo), but also in task contingencies (50% vs. 100% reward probability), which are known to impact cue-driven behavior. This experimental heterogeneity limits the interpretability of pooled correlations across the entire sample. However, we conducted exploratory correlation analyses within each subgroup (i.e., 2 × 2 crossing of beverage and certainty conditions) to provide preliminary insights into whether the relationship between alcohol use severity and ST/GT performance is preserved across task variants. These subgroup-level findings should be interpreted with caution due to limited power and lack of formal hypothesis, as reported in the Supplemental Information (S4).

Finally, for completeness and informational purposes, exploratory correlational analyses were conducted separately within the Alcohol and Placebo groups to examine associations between alcohol consumption levels, self-reported subjective effects of intoxication, and measured breath alcohol concentration (in the Alcohol Group). The results of these analyses are presented in the Supplemental Information (S5).

Results

Manipulation check

To confirm that the placebo beverage was effective in eliciting subjective effects, we first assessed whether DEQ subscale scores in the Placebo group were significantly greater than zero. All DEQ subscales showed significant deviations from zero in the expected direction, indicating that participants in the Placebo group did report subjective effects despite the absence of alcohol: “Feel” (W = 1128, $p < 0.001$, r = 0.76), “High” (W = 703, $p < 0.001$, r = 0.68), “Like” (W = 1891, $p < 0.001$, r = 0.87), and “More” (W = 1275, $p <$ 0.001, r = 0.79).

To evaluate whether the experimental manipulations produced the expected differences between groups, a series of Mann–Whitney U tests were conducted on the DEQ subscales. As predicted, the Alcohol group reported significantly higher ratings than the Placebo group on the “Feel” (W = 2793, $p < 0.001$, r = 0.52; Alcohol: M = 58.93, SD = 28.94; Placebo: M = 26.80, SD = 26.20) and “High” (W = 2679, $p < 0.001$, r = 0.47; Alcohol: M = 44.47, SD = 30.39; Placebo: M = 15.33, SD = 21.04) subscales. In contrast, no significant group differences were found for the “Like” (W = 1681, p = 0.76, r = 0.03; Alcohol: M = 55.09, SD = 21.75; Placebo: M = 56.39, SD = 12.64) or “More” subscales (W = 1448, p = 0.15, r = 0.13; Alcohol: M = 38.49, SD = 28.98; Placebo: M = 47.53, SD = 33.72), suggesting that these aspects of the subjective responses were not reliably influenced by the presence of alcohol. Violin and box plots with individual data points are available in Supplemental Information (S6) for each subscale.

Is sign-tracking tendency affected by alcohol intoxication and reward probability?

Block 1

A two-way ANOVA was conducted to examine the effects of Group (Alcohol vs. Placebo) and Reward Probability (50% vs. 100%) on the ST/GT scores. The analysis revealed a statistically significant main effect of Probability ($F_{(1, 114)}$ = 4.53, p = 0.03, partial η^2 = 0.04), indicating that reward uncertainty significantly influenced participants' sign-tracking responses, in the sense that ST/GT scores were higher in the 50% group. However, the main effect of Group was not statistically significant ($F_{(1, 114)}=1.61$, $p=0.20$, partial $\eta^2 = 0.01$) suggesting no overall difference between the Alcohol and Placebo conditions. The Group × Probability interaction was also not statistically significant ($F_{(1, 114)}=0.34$, $p=0.56$, partial $\eta^2 = 0.003$), indicating that the effect of probability did not differ as a function of group. Figure 2 shows the distribution of the ST/GT score across experimental conditions.

Block 2

A two-way ANOVA was also conducted to examine the same effects in the second block of the task. The analysis did not reveal any statistically significant main effect: nor for Probability ($F_{(1, 114)}=1.80$, $p=0.18$, partial $\eta^2 = 0.02$), neither for Group ($F_{(1, 114)}=0.74$, $p=0.39$, partial $\eta^2 = 0.006$). The Group × Probability interaction was also not statistically significant ($F_{(1, 114)}=0.02$, $p=0.88$, partial $\eta^2 < 0.001$).

To further examine what happened during the first block, specifically regarding participants' attention toward the Sign, we conducted the same analysis using Dwell Time on the Sign only. This analysis is available in Supplemental Information (S7).

Do alcohol intoxication and reward uncertainty increase resistance to reward omission?

For **sign-directed attention**, the mixed effects model (fit by REML) revealed a significant interaction between Block and Certainty on fixation duration. Specifically, the effect of Omission differed depending on certainty level, as indicated by a significant Omission × Certainty (100%) interaction (Estimate=0.38, SE=0.13, t(109)=2.81, $p=0.005$). No significant main effect of Omission was observed ($p=0.110$), nor were the main effects of group ($p=0.136$) or certainty ($p=0.164$) significant. The main effect of Block 2 (compared to 1) was however significant ($p=0.007$). Similarly, the interaction between Omission and group ($p=0.291$), as well as the three-way interaction among Omission, group, and certainty ($p=0.994$), were not significant. Post-hoc pairwise comparisons (adjusted using Tukey's method) of estimated marginal means revealed that the effect of Block on sign Dwell Time varied across certainty levels. For participants in both 50% certainty groups, the difference between acquisition (Block 1) and reward omission was not significant (estimate = −0.08, SE=0.07, z = −1.28, $p=0.407$), nor was the difference between Block 2 and omission. However, between block 1 and 2 there was a statistically significant difference with the second block being below the first (estimate=0.17, SE=0.05, z=3.26, $p=0.003$. In contrast, for participants with 100% certainty, Dwell Time to the sign significantly increased during reward omission in both groups when compared to the first acquisition Block : estimate=0.29, SE=0.06, z=4.58, $p<0.001$). The difference between Block 2 and Omission was also statistically significant in these groups (estimate=0.33, SE=0.05, z=6.19, $p<0.001$). Figure 3 shows means and boostrapped confidence intervals of Sign Dwell Time in Blocks 1 and 2 and "Omission", without considering the alcohol group variable (non-significant) for clarity. For completeness, the complete figure, including also alcohol group is available in the supplementals (S3).

For **goal-directed attention**, the mixed effects model (fit by REML) revealed a significant interaction between Block and Certainty on Goal fixation duration. Specifically, the effect of Omission differed again depending on certainty level, as indicated by a significant Omission × Certainty (100%) interaction (Estimate =-0.64, SE = 0.20, t(109) = -3.29, p = 0.001). No significant main effect of Omission was observed (p = 0.07), nor were the main effects of group (p = 0.73) or certainty (p = 0.44) significant. Similarly, the

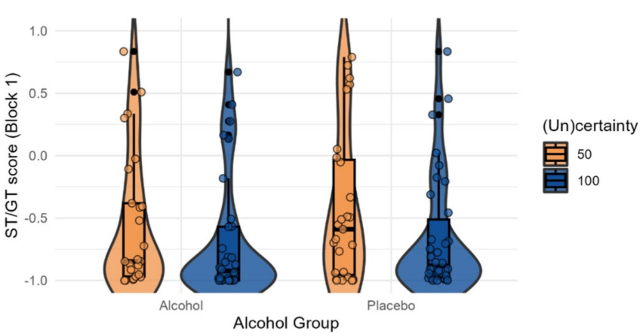

Fig. 2 Distribution of ST/GT Scores by Group and (Un)Certainty Level. Note. Horizontal lines are the medians of the ST/GT scores during the first block of the PCA task. The lower and upper limits of the boxes are lower and upper quartiles, the ends of the whiskers represent the lower and upper extremes. The lines around the boxes represent the distribution of the data in each group

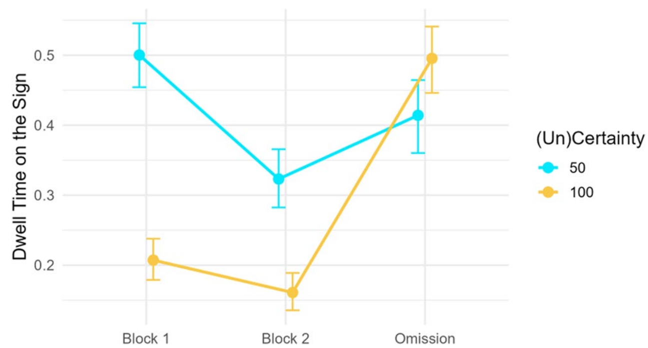

Fig. 3 Sign-Directed Dwell Time Across Blocks and Omission by (Un)Certainty. Note. Points represent the means, and vertical error bars indicate the 95% bootstrap-based confidence intervals. Alcohol Group was not included here for clarity in data display, see Supplementals S3 for the comprehensive Figure

interactions between Omission and group (p = 0.42) and between group and certainty (p = 0.86), as well as the three-way interaction among Omission, group, and certainty (p = 0.60), were not significant.

Post-hoc pairwise comparisons (corrected using Tukey's method) of estimated marginal means revealed that the effect of Omission on goal Dwell Time varied across certainty levels. For participants in the 50% certainty groups, the difference between acquisition (Block 1) and Omission was not significant (estimate = 0.18, SE = 0.10, z = 1.78, $p = 0.175$). The differences between Block 1 and 2 was statistically significant (alcohol and placebo groups pooled, estimate = 0.23, SE = 0.08, z = 2.84, $p = 0.012$) while between block 2 and Omission it was not (estimate = 0.05, SE = 0.09, z = 0.58, $p = 0.83$). In contrast, for participants with 100% certainty, Dwell Time to the goal significantly decreased between acquisition (Block 1) and reward omission (both alcohol and placebo groups pooled) : estimate = −0.39, SE = 0.09, z = −4.28, $p < 0.001$). Both groups also displayed statistically significant differences between Block 2 and Omission ($p < 0.001$), as well as between Block 1 and 2 ($p = 0.015$). Figure 4 shows means and boostrapped confidence intervals of Goal Dwell Time in Blocks 1, 2 and Omission.

Taking initial ST/GT tendency as a covariate

First, the relationship between initial ST/GT scores (Block 1) and dwell time for the sign during reward omission was tested using Spearman's Rho correlation. A positive and statistically significant correlation of almost large size was found ($\rho_S = 0.47$, $p < 0.001$).

Sign and goal dwell time during omission

A multiple linear regression was conducted to examine the effects of Group (Alcohol vs. Placebo), Probability (50% vs. 100%), initial ST/GT score (Block 1, centered), and their interactions on Sign-directed dwell time during the omission phase. The model accounted for a significant portion of variance ($R^2 = 0.31$, *adj.* $R^2 = 0.27$, $F_{(7, 105)} = 6.86$, $p < 0.001$).

There was a statistically significant interaction between Group and ST/GT score ($b = 11.00$, $SE = 3.55$, $p = 0.002$, partial $\eta^2 = 0.03$), suggesting that higher ST/GT scores predicted longer Dwell Times in the Placebo group, but not in the Alcohol group. Additionally, the three-way interaction between Group, Probability, and ST/GT score was also significant ($b = -13.21$, $SE = 5.15$, $p = 0.012$, partial $\eta^2 = 0.06$), suggesting modulation of the effect by probability. Specifically, the positive association between the ST/GT score and Dwell Time for the Sign cue in the Placebo group was stronger when certainty was minimal (50%), while it was stronger in the Alcohol group when certainty was maximal (100%). Figure 5 (upper panel) illustrates the predicted Sign-directed Dwell Times based on the fitted model, allowing visualization of the interaction between Group, Probability, and ST/GT scores. No significant main effects were observed for Group ($p = 0.116$, partial $\eta^2 = 0.05$) and ST/GT score ($p = 0.478$, partial $\eta^2 = 0.22$), with a non-significant trend for Probability ($p = 0.056$, partial $\eta^2 = 0.008$), when controlling for the other factors. There were also no significant two-way interactions found between Group and Probability ($p = 0.484$, partial $\eta^2 = 0.01$) or between Probability and ST/GT score ($p = 0.239$, partial $\eta^2 = 0.009$). These results suggest that the relationship between initial sign-tracking behavior and Dwell Time to the Sign (during reward omission) is context-dependent, emerging primarily under conditions of uncertainty, in the absence of alcohol.

The same analysis was conducted to examine the effects of Group (Alcohol vs. Placebo), Probability (50% vs. 100%), ST/GT score (Block 1, centered), and their interactions on Goal-directed dwell time during the omission phase. The model accounted for a significant portion of variance ($R^2 = 0.29$, *adj.* $R^2 = 0.25$; $F_{(7, 105)} = 6.23$, $p < 0.001$).

A significant main effect of Probability emerged, with higher predicted Goal-directed Dwell Times in the 50% condition compared to the 100% condition ($p = 0.011$, partial $\eta^2 = 0.07$; Condition 50% (Uncertainty): $M = 30.28$, SD = 14.71, Min-Max = 4.20–52.15.20.15; Condition 100% (Certainty): $M = 19.98$, SD = 11.11, Min-Max = 1.65–43.08). Additionally, the interaction between Group and ST/GT score was statistically significant ($p = 0.026$, partial $\eta^2 = 0.03$), suggesting that the relationship between initial ST/GT score and Goal Dwell Time varied between Alcohol and Placebo conditions. Figure 5 (bottom panel) illustrates predicted Goal Dwell Times during the reward omission block as a function of initial ST/GT scores (Block 1), Group (Alcohol vs. Placebo), and Probability (50% vs. 100%), based on the fitted linear model. Specifically, in the Placebo

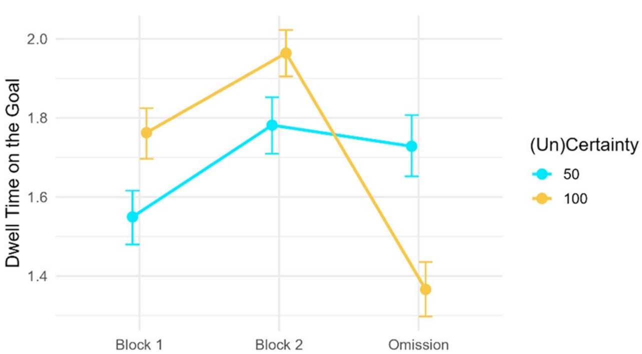

Fig. 4 Goal-Directed Dwell Time Across Blocks and Omission by (Un)Certainty. Note. Points represent the means, and vertical error bars indicate the 95% bootstrap-based confidence intervals. Alcohol Group was not included here for clarity in data display, see Supplementals S3 for the comprehensive Figure

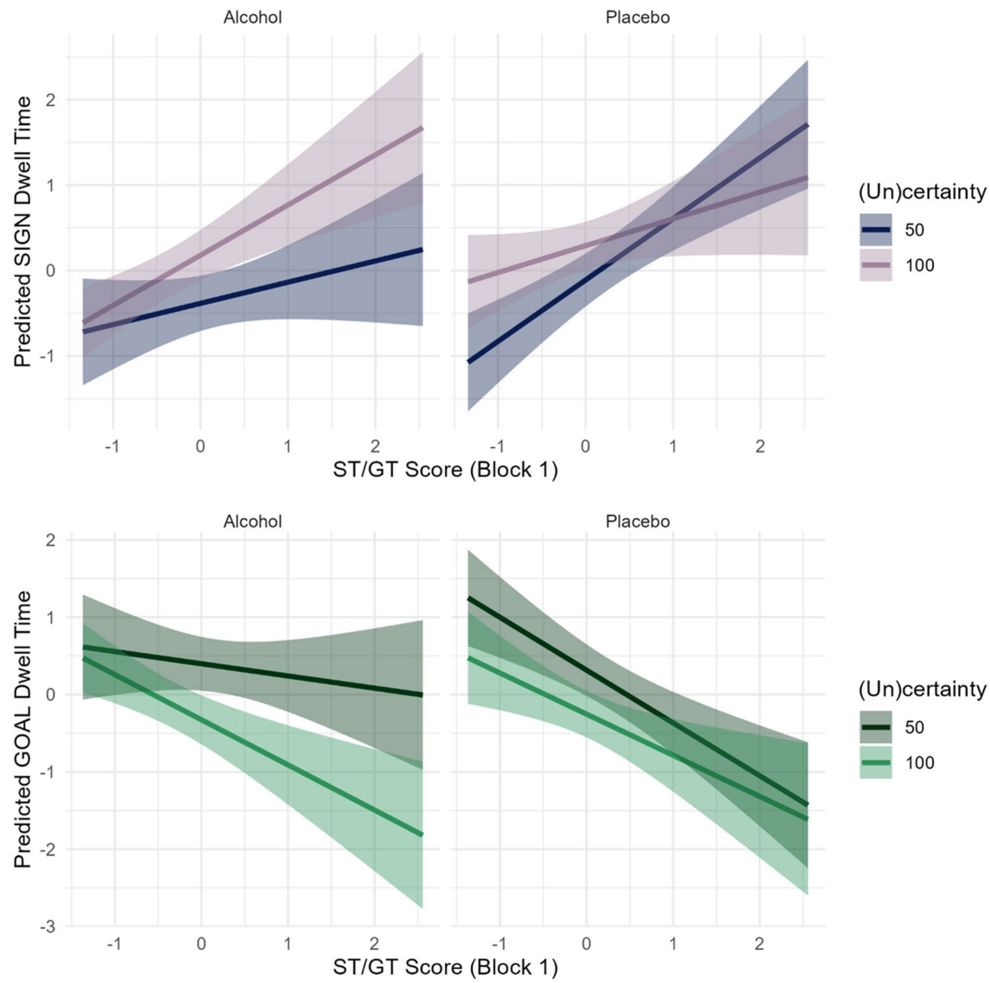

Fig. 5 Predicted Dwell Times for Sign and Goal by Group, Probability, and ST/GT Scores during Reward Omission. Note. The upper panel shows dwell times toward the Sign during reward omission, while the bottom panel shows dwell times towards the Goal during reward omission. Both dwell time scores were Inverse Normal Transformed (Blom's Method). Shaded areas represent 95% Confidence Intervals. Lines are split by Probability (dark grey for 50% in printed version vs. light grey for 100% in printed version) and panels by Group (Alcohol vs. Placebo)

group, higher sign-tracking scores were associated with decreased Goal-oriented dwell time, while this relationship was weaker in the Alcohol group. No other main effects or interactions reached statistical significance (all $ps < 0.18$).

Discussion

The present study aimed to extend the translational validity of the sign-tracker/goal-tracker (ST/GT) model in humans by examining the impact of two modulators of reward processing—reward uncertainty and acute alcohol administration—on cue-driven behavior. Building on prior animal and human research suggesting that both factors can amplify impulsive responding and enhance attention to reward-predictive cues (e.g., (Anselme et al. 2013; Duckworth 2017), we hypothesized that each of them would increase sign-tracking behavior as measured in a PCA task. Moreover, given that sign-tracking is known to increase resistance to extinction (Ahrens et al. 2016), we expected these effects to persist during a non-rewarded phase.

Consistent with our first hypothesis, we found that reward uncertainty significantly increased the ST/GT score (Block 1), supporting the notion that uncertainty enhances motivation and attention towards reward-predictive cues. This effect was even stronger when examining raw Sign-oriented dwell times, with a medium effect size (partial $\eta^2 = 0.07$).

The motivating effect of uncertainty can be conceptually understood through several complementary explanatory frameworks. From a learning theory perspective, unpredictable rewards generate larger prediction errors (i.e., discrepancies between expected and actual outcomes) which enhance the associability of predictive cues and increase attentional exploration to optimize learning (Pearce and Hall 1980). From cognitive and evolutionary viewpoints, exposure to reward uncertainty promotes sign-tracking because the lack of cognitive or predictive control over outcomes encourages organisms to attend to any stimulus that might signal the possibility of reward. This strategy may represent an adaptive mechanism to increase survival in environments where food resources are scarce and unpredictable (Anselme et al. 2013; Anselme and Güntürkün 2018).

Contrary to a consistent cue-reward association, an inconsistent one is not attractive (low incentive salience) but it is assumed to increase effort mobilization to seek—rather than approach—potential rewards in challenging contexts (Anselme 2025).

Surprisingly, results from the second block of the PCA task suggested that the stimulating effect of uncertainty on ST/GT tendencies was significant during the first block only. A two-block analysis has already been carried out in the study of human sign-tracking (Dinu et al. 2024; Garofalo and di Pellegrino 2015), however, it is quite common to consider only the second block of conditioning trials for the learning to be in place and stable (Garofalo and di Pellegrino 2015; Colaizzi et al. 2023). However, a similar pattern of transient modulation has been reported by Cho and Cho (2021), who found that uncertainty effects on cue-directed attention in the UMAC task also diminished after the initial block. A plausible explanation for this pattern is that participants' knowledge of what happens during the task is substantially different from Block 1 to Block 2. By the start of Block 2, they had already experienced the CS–UCS contingency once and may have learned that the UCS followed the CS approximately 50% of the time. This prior exposure likely reduced the novelty and unpredictability of the contingency, meaning that the sense of uncertainty was less salient in Block 2 than during the initial learning phase in Block 1. At least in humans, this suggests that uncertainty may exert a short-lived but potent influence on initial cue engagement, possibly through heightened orienting responses that taper as the task progresses and participants adapt to contingencies. These findings were not anticipated in the original design and hypotheses, and focusing on Block 1 departs from typical practice in human ST/GT research. Therefore, caution is warranted. Replications are clearly needed to support our interpretation.

Contrary to our expectations, alcohol administration did not increase ST/GT scores, suggesting that a moderate dose (0.5 g/kg) was perhaps not sufficient to modulate sign-tracking tendencies in our task or that its effects were more subtle or indirect. This finding contrasts with earlier rodent studies—for instance, Tomie and colleagues (1998a, b) reported a significant increase in sign-tracking behavior in rats following ethanol administration. As outlined in the introduction, impulsivity and attentional bias (both traits often linked to sign-tracking tendencies) are frequently shown in the literature to increase following alcohol administration, which further reinforced our expectation that alcohol would enhance sign-tracking. However, other studies have yielded opposite results. Versaggi et al. (2016) for instance, observed that ethanol administration reduced sign-tracking and increased goal-tracking behavior across several days, concluding that ethanol decreased the incentive salience attributed to the CS. Similarly, Fiorenza et al. (Fiorenza et al. 2018) found that while chronic intermittent ethanol (CIE) exposure and Pavlovian conditioning with alcohol rewards enhanced dopamine release, this effect was not accompanied by greater sign-tracking; instead, CIE shifted behavior from sign-tracking toward goal-tracking. These inconsistencies highlight the complexity of alcohol's influence on cue-motivated behavior. Notably, the present study used a single acute ethanol dose, which may partly explain the divergent findings. From an attentional perspective based on alcohol myopia theory, it has also been suggested that due to attentional capacity reduction, acute alcohol could decrease cue-related attention, which has been shown experimentally in a VMAC study (Watson et al. 2020). It is worth noting that the present study is among the very few to examine alcohol's influence on sign-tracking behavior in humans. Our findings suggest that the relationship between alcohol and sign-tracking may be more complex and context-dependent than previously assumed. Dose-dependent effects may also be a key factor: an experiment from Duckworth's thesis (2017) found that sign-tracking was increased following a low dose of alcohol (0.3 g/kg) but remained unaffected at a higher dose (0.6 g/kg), indicating that alcohol's influence on sign- versus goal-tracking may vary non-linearly with dosage. Our intermediate dosage (0.5 g/kg) was possibly too elevated for optimal effects.

Regarding our second, exploratory hypothesis on reward omission, we observed a pattern that diverged from our initial predictions. Specifically, a significant interaction between Omission block and certainty revealed that sign-directed attention *increased* during extinction in the 100% (certainty) conditions (both Alcohol and Placebo). Rather than the anticipated attenuation of cue-directed attention under non-reinforcement in this group, reward omission was associated with a paradoxical increase in attention toward the sign when prior reward contingencies were fully predictable. Additionally, a significant reduction in goal dwell time was observed across all high-certainty (100%) conditions (Fig. 4), which is consistent with a straightforward extinction-like decline in attention to the goal location once reward delivery ceased.

One plausible explanation for this unexpected pattern of results is related to the limited number of omission trials included in the present study ($n=15$). In animal conditioning preparations, early extinction is often characterized by an initial transient increase in the vigor of conditioned responding (referred to as an extinction burst, Shahan 2022) before a gradual decline occurs. Because the extinction phase in our study was intentionally brief to preserve participant engagement and avoid excessive task duration, it is possible that the increase in sign-directed attention we observed reflects this early, transient burst rather than a genuine failure of extinction learning. Similar increases in responding have also been linked to greater behavioral variability during early extinction (Donoso et al. 2021).

From the perspective of reward prediction error frameworks, early extinction represents a moment of unexpected ambiguity: the organism's prediction is violated, and this mismatch can redirect attention toward the broader environment, promoting exploratory behaviors that help reduce uncertainty (Rosas et al. 2006; Gottlieb et al. 2013; Beesley et al. 2015). A familiar example is a dog repeatedly retrieving a thrown object—attention remains tightly focused on the throw. However, when the owner pretends to throw the object and withholds it, the dog rapidly shifts to scanning and sniffing the surroundings. This attentional reorientation occurs precisely because the previously reliable cue no longer predicts its expected outcome In the context of the present study, participants in the 100% group had experienced fully predictable CS–UCS pairings during acquisition. For them, the transition to omission introduced a novel source of ambiguity: the CS no longer guaranteed reward delivery. This shift may have encouraged exploratory behavior, such as increased attention to the alternative stimulus on the screen and shifting attention away from the goal, as participants attempted to update their beliefs about the task structure. In other words, participants may simply react to the unexpected change in the task by 'trying to figure out what is going on.' By contrast, participants who had already encountered uncertain cue–reward contingencies during acquisition may have been less affected by this change, as ambiguity was already a familiar feature of their learning environment.

To further explore the factors contributing to reward omission-phase behavior, we conducted two multiple linear regression analyses: one predicting sign-oriented dwell time, and the other goal-oriented dwell time, during reward omission. In each model, we included initial ST/GT scores, reward uncertainty, alcohol administration, and their interactions as predictors. The results indicate that in the placebo groups (regardless of probability during the PCA task), individuals having higher initial ST/GT scores (indicative of sign-tracking behavior), were more focused on the Sign cue during the non-rewarded omission phase. This result is consistent with the literature on sign-tracking during traditional extinction (Ahrens et al. 2016). Additionally, the three-way interaction, indicated that participants most likely to persist in sign-oriented attention during reward omission were those who had not consumed alcohol and who exhibited higher initial ST/GT scores. Interestingly, participants who had consumed alcohol also demonstrated greater persistence in sign-oriented attention when they combined strong sign-tracking tendencies with prior reward certainty (100%), highlighting a more conditional role of alcohol effects (Fig. 5).

In the model predicting goal-oriented dwell time during reward omission, we observed a significant main effect of uncertainty (medium effect size), with participants in the 50% condition exhibiting greater goal-directed attention than those in the 100% condition, regardless of alcohol administration. Additionally, a significant interaction between alcohol administration and the ST/GT score (small effect size), revealed that the negative association between ST/GT score and goal-oriented attention was stronger in the placebo group than in the alcohol group (Fig. 5, bottom panel). In other words, in the absence of alcohol, baseline ST/GT tendencies more strongly predicted resistance to disengaging from the goal location during reward omission: individuals with more goal-tracking-like behaviors (i.e., lower ST/GT scores during initial PCA task) were more likely to maintain attention to the reward location despite non-reinforcement. This pattern contrasts with findings from the animal literature, where goal-trackers typically show rapid reductions in goal-directed behavior during Pavlovian extinction (Ahrens et al. 2016). One possible explanation is that alcohol introduced additional variability or noise in attentional deployment, thereby attenuating the relationship between baseline tracking tendencies and extinction behavior.

It is noteworthy that we observed a significant positive correlation between initial sign- and goal-tracking tendencies and sign-oriented attentional behavior during reward omission ($\rho = 0.47$, $p < 0.001$). The regression analyses further indicated that this association was more pronounced in placebo conditions, whereas alcohol appeared to blunt the relationship between baseline ST vs. GT-like behavior and reward omission-phase attention.

Regarding the exploratory findings of this study, several observations merit attention. First, although not statistically significant, we observed a positive correlation between the ST/GT score and alcohol consumption variables (Supplemental S4) in the 100% placebo group—the condition that resembles the most the conditions tested in other works. Notably, the magnitude of these correlations was similar to those reported in our earlier study using the same task (Heck et al. 2025b). In contrast, results concerning impulsivity dimensions were less consistent. Statistically significant positive correlations between ST/GT scores and impulsivity traits emerged only in groups that received alcohol. However, all of these correlations should be interpreted with caution due to the limited statistical power inherent in performing correlational analyses at the group level.

Although not directly related to the primary objectives of the present study, a noteworthy secondary finding emerged. In the alcohol group (but not the placebo group), AUDIT scores were significantly negatively correlated with subjective ratings of alcohol intoxication, as measured by the DEQ "feel" and "high" dimensions (see Supplemental S5), and positively correlated with the "more" dimension. This exact pattern was previously observed (Morean et al. 2013) during validation of the DEQ. Morean and collaborators proposed it may reflect tolerance (i.e., a reduced subjective response to alcohol associated with heavier use). This finding aligns with broader evidence suggesting that heavy drinkers exhibit blunted subjective responses to alcohol, consistent with a low-response phenotype (Schuckit et al. 2011). Furthermore, BrAC levels measured at the end of the experiment were positively correlated with DEQ "feel" and "high" ratings, with medium to large effect sizes, supporting the construct validity of these DEQ subscales as reliable subjective indices of alcohol intoxication.

Limitations and perspectives

The main strength of this study lies in its contribution to the growing body of evidence demonstrating the stimulating effect of uncertainty on approach behavior toward reward-predictive stimuli. As outlined in the introduction, converging results in humans are beginning to mirror findings from decades of research in animal models (see Anselme et al. 2013; Crawford et al. 1985; Glueck et al. 2018; Pearce et al. 1985), reinforcing the translational relevance of this phenomenon.

In contrast, the effects of alcohol on sign-tracking tendency in this study were less conclusive. Future research should examine the impact of varying alcohol doses within this PCA paradigm, as dose-dependent effects have been observed both in humans (Duckworth 2017) and animals (Tomie et al. 1998a, b) for sign-tracking behavior.

An important aspect to emphasize is that human PCA tasks inherently differ from rodent models in terms of the spatial and attentional demands of sign- and goal-tracking behavior. Because the CS and reward location were spatially proximate in our setup, and shifting gaze between them was uncostly, human participants may have adopted a more fluid strategy (moving their eyes up and down regularly) or an overlapping strategy (maintaining gaze in the middle of the screen). This may sharply contrast with rodent paradigms, where the physical separation of the lever CS and reward magazine imposes clear behavioral choices and hence exclusive approach patterns. Moreover, whereas human sign-tracking scores are typically derived from a relatively small number of CS–UCS pairings (e.g., two blocks of 20 trials), sign- and goal-tracking behaviors in animal studies are often established across multiple sessions, usually containing 30 + trials per training day. Additionally, our design did not allow for direct comparisons between individuals formally classified as sign-trackers versus goal-trackers, since we did not assess baseline ST/GT behavior prior to experimental manipulation, which may have contributed to variability in the results. Thus, future research should address this limitation by conducting within-subject (repeated measures) designs with an initial task using more conventional parameters (e.g., CS–US contingencies around 80%, 30 trials) to enable a clearer phenotypic classification before examining how uncertainty and alcohol interact with these individual differences. Incorporating a reinstatement phase following extinction would also be valuable, as it could reveal whether sign- versus goal-tracking–prone individuals differ in reacquiring Pavlovian attentional responses to both sign and goal cues, and whether reinstatement is modulated by reward probability or alcohol administration.

As already mentioned, current knowledge of the factors that modulate human sign- and goal-tracking remains limited. Consequently, we cannot yet assert with confidence that simplified, computerized adaptations of PCA tasks fully capture the same underlying processes as observed in animal models. For example, our task does not allow us to conclude with certainty that incentive salience has been attributed to the CS, because our procedure did not include a control cue (not associated with reward delivery). Nevertheless, recent theoretical and empirical work has begun to establish explicit links between incentive salience attribution and human sign-tracking. In particular, VMAC paradigms suggest that reward-associated cues acquire motivational significance, increasing their attentional salience and influencing behavior accordingly (Le Pelley et al. 2024). Complementing this, fMRI-based evidence from PIT tasks indicates differential incentive salience attribution in human sign- versus goal-trackers (Schad et al. 2020). Importantly, this remains the only study to date that has addressed the question directly, underscoring the need for further research. Future work should clarify the role of incentive salience attribution in human sign- and goal-tracking and develop robust, standardized methods for assessing it in tasks such as computerized PCA.

Additional limitations may also account for the inconclusive effects of alcohol, including individual differences in response to intoxication. Although we attempted to standardize intoxication levels across participants by adjusting doses according to body weight and requiring participants to fast for five hours before testing, there was still considerable variability in subjective and observable intoxication. Some participants exhibited only mild signs of intoxication, while others showed more pronounced effects. This variability is, however, common in alcohol research and is likely attributable to individual differences in alcohol metabolism (Gentry 2000). A possible improvement would be to calculate alcohol dosage based on body mass index (BMI) rather than body weight alone, as this may better account for individual differences in alcohol distribution volume (Jones 2007).

Beyond its main hypotheses, the study also yielded theoretically important secondary findings concerning placebo effects in single-blind alcohol administration paradigms and subjective intoxication ratings. Specifically, while the experimental groups differed significantly in their ratings of "feeling high" and "feeling alcohol effects," they did not differ in ratings of "liking the effects" or "wanting more". Notably, participants in the placebo group scored significantly above zero on all subscales, indicating that even without consuming actual alcohol, the experience of drinking a non-alcoholic beverage presented as rum elicited appreciable subjective effects. These participants reported liking and wanting these effects to the same extent as those who consumed real alcohol. From an applied perspective, these findings support the idea that non-alcoholic beverages (or "mocktails") may hold promise in reducing alcohol consumption while still providing positive experiential outcomes, with potential implications for harm-reduction strategies and the development of appealing alcohol-free alternatives. However, from an experimental perspective, the strength of the placebo effect observed in the mocktail condition may plausibly contribute to the absence of a detectable alcohol effect in the present study. Specifically, the placebo group may have exhibited an alcohol-like conditioned response, thereby diminishing observable differences between conditions. However, our hypotheses pertained to the pharmacological effects of alcohol rather than expectancy-based mechanisms. Future research should address this limitation by explicitly separating these components—for example, by including a control group that knowingly consumes a non-alcoholic beverage—to better isolate pharmacological effects from expectancy and conditioning influences.

In summary, while the study has some methodological limitations, it provides valuable experimental insights into human ST/GT behavior and highlights, most notably, the role of uncertainty in modulating responses to reward-predictive cues. Still, given the novelty of this experimental investigation, caution is warranted, and further research is essential before drawing firm conclusions or generalizing beyond the present context.

Conclusion

This study provides novel insights into how environmental factors (such as reward uncertainty) and pharmacological influences (such as alcohol intoxication) shape reward cue-reactivity. These findings mark a step forward in the translational implementation of sign- and goal-tracking behaviors, a field that has long been established in animal research but remains in its early stages with respect to humans. By advancing our understanding of the mechanisms underlying these behaviors, this work contributes to the broader effort to uncover both individual and environmental vulnerability factors relevant to addiction and other cue-driven psychopathologies. In particular, it highlights how uncertainty can intensify attentional focus on reward-predictive cues—a process that, while adaptive in some contexts, may become maladaptive by promoting compulsive approach behavior and increasing susceptibility to addiction.

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Data availability The experimental data are available on OSF, at the following link [https://osf.io/9zfkb].

Declarations

Human ethics and consent to participate The study was conducted in accordance with institutional guidelines and the Declaration of Helsinki. It was approved by the local Ethics Committee, and informed consent was given by all participants.

Use of IA Artificial intelligence (ChatGPT, OpenAI, 2024) was used exclusively to enhance the readability and stylistic clarity of the manuscript. No AI tools were employed in the generation of scientific ideas, data analyses, or interpretations. The authors take full responsibility for the content and accuracy of the information presented in this article.

Clinical trial number Not applicable.

Competing interests The authors declare no competing interests.

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