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## Separate and Combined Effects of Energy Drinks and Alcohol Intake on Exploratory Behaviour of Male Albino Rats

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### Abstract

*The increasing concurrent consumption of energy drinks and alcohol raises concerns about their combined effects on learning and exploratory behaviour, yet controlled experimental evidence remains limited. This study examined the separate and combined effects of energy drink and alcohol consumption on exploratory behaviour in male albino Wistar rats using the T-maze paradigm. Twenty-seven rats (160–190 g) were randomly assigned to four groups (n = 7 per group) and orally administered energy drink (Red Bull), alcohol (Smirnoff vodka), a combination of both, or distilled water (control) for 28 days. Exploratory behaviour was assessed using task completion latency. Data were analysed using one-way analysis of variance (ANOVA) followed by Tukey's honestly significant difference (HSD) post hoc tests, with statistical significance set at  $p < .05$ . A significant main effect of treatment was observed,  $F(3, 23) = 9.349$ ,  $p < .001$ ,  $\eta^2 = .549$ , indicating that substance administration accounted for 54.9% of the variance in performance. Rats exposed to alcohol exhibited significantly impaired performance ( $M = 216.38$  s) compared with the control group ( $M = 113.71$  s,  $p = .001$ ) and the energy drink group ( $M = 117.75$  s,  $p = .002$ ). No significant difference was observed between the energy drink and control groups ( $p = .998$ ). The combined energy drink–alcohol group demonstrated intermediate impairment, performing significantly worse than the control ( $p = .019$ ) and energy drink ( $p = .028$ ) groups. These findings indicate that chronic alcohol exposure significantly impairs spatial learning and memory, whereas energy drink consumption alone does not markedly influence exploratory performance. Co-administration of energy drinks with alcohol does not mitigate alcohol-induced cognitive deficits, highlighting potential neurobehavioural risks associated with their concurrent use.*

**Keywords:** energy drinks, alcohol, exploratory behaviour, spatial learning, Wistar rats, T-maze

### Introduction

Learning is conceptualised as a relatively enduring change in behaviour or behavioural potential resulting from experience rather than temporary performance fluctuations (Kandel et al., 2021). In experimental neuroscience, learning-related processes are commonly inferred from observable behaviours such as exploration, novelty seeking, and spatial engagement, which reflect underlying neural plasticity and cognitive processing (Squire, 1992; Whishaw & Kolb, 2005).

Exploratory behaviour constitutes a core component of learning and adaptation, enabling organisms to gather information about novel environments, identify potential resources, and detect threats. In rodents, behaviours such as locomotion, rearing, sniffing, and object investigation are routinely used as indices of cognitive integrity, motivation, and emotional regulation (Ennis et al., 2021). Consequently, alterations in exploratory activity may signal disruptions in learning capacity, affective state, or neural function.

Neurobiologically, exploratory behaviour depends on coordinated activity within the hippocampus, prefrontal cortex, and mesolimbic dopaminergic pathways. The hippocampus is critical for spatial learning and memory consolidation (Squire, 1992), while the prefrontal cortex regulates executive control and

behavioural flexibility during exploration (Whishaw & Kolb, 2005). Dopamine plays a central role in reward processing and motivation, whereas acetylcholine modulates attention and sensory integration during learning (Berridge & Waterhouse, 2003; Hasselmo, 2006; Schultz, 2007).

Male albino Wistar rats are widely used in behavioural neuroscience research due to their well-characterised neuroanatomy, behavioural reliability, and translational relevance to human cognitive processes (Whishaw & Kolb, 2005). Standardised paradigms such as the open field test, Morris water maze, radial arm maze, and novel object recognition task provide validated measures of exploratory behaviour, spatial learning, and memory performance (Ferster & Skinner, 1957; Morris, 1984).

Energy drinks (EDs) are among the most widely consumed psychoactive beverages globally, particularly among adolescents and young adults (Heckman et al., 2019; Seifert et al., 2011). Their primary active ingredient, caffeine, exerts stimulant effects through antagonism of adenosine receptors, resulting in increased neuronal excitability and enhanced dopaminergic and noradrenergic transmission (Fredholm et al., 1999). Acute caffeine intake has been associated with increased alertness, locomotor activity, and improved task engagement (Giles et al., 2012). Other common ED constituents, including taurine, sugars, and B-vitamins, may further influence neurobehavioural outcomes through neuromodulatory and metabolic mechanisms (Schaffer et al., 2010; Curran & Marczyński, 2017).

In contrast, alcohol acts as a central nervous system depressant, enhancing inhibitory  $\gamma$ -aminobutyric acid (GABA) transmission while suppressing excitatory glutamatergic signalling (Farrant & Nusser, 2005; Koob & Volkow, 2016). These actions disrupt synaptic plasticity processes, including long-term potentiation in the hippocampus, leading to impairments in learning, memory consolidation, and behavioural regulation (Bliss & Collingridge, 1993). Empirical evidence demonstrates that alcohol exposure adversely affects working memory, attention, and exploratory engagement, particularly following repeated or high-dose consumption (White, 2003; Squeglia et al., 2014).

At the cellular level, learning and exploratory behaviours rely on a finely regulated balance between excitatory and inhibitory neurotransmission involving dopamine, acetylcholine, GABA, and glutamate systems (Hasselmo, 2006; Schultz, 2007). While stimulant substances such as caffeine may transiently enhance behavioural activation and exploration, depressants such as alcohol typically suppress these processes. Importantly, concurrent exposure to stimulants and depressants may result in complex, non-additive interactions that produce behavioural outcomes distinct from those observed following individual substance administration (Marczyński & Fillmore, 2014).

Recent studies raise increasing concerns regarding the combined consumption of energy drinks and alcohol. Evidence indicates that EDs may mask alcohol's subjective sedative effects without reversing alcohol-induced cognitive impairments, thereby promoting increased alcohol intake, prolonged drinking episodes, and behavioural disinhibition (Peacock et al., 2012; Marczyński & Fillmore, 2014; Arria & O'Brien, 2019). Animal studies further demonstrate that co-administration exacerbates oxidative stress, neuroinflammation, and behavioural dysregulation in brain regions critical for learning and exploration (Díaz et al., 2016; Petribu et al., 2023).

The consumption of energy drinks and alcohol has increased substantially over the past two decades, particularly among adolescents and university students, raising significant public health concerns (Seifert et al., 2011; Heckman et al., 2019). While energy drinks are commonly consumed to enhance alertness and perceived cognitive performance, alcohol is well documented to impair learning, memory, and executive functioning, especially when consumed excessively (White, 2003; Squeglia et al., 2014). Of particular concern is the growing practice of combining the consumption of energy drinks with alcohol. Caffeine-containing EDs can reduce subjective feelings of alcohol intoxication while leaving objective cognitive and motor impairments largely intact (Marczyński & Fillmore, 2014). This masking effect may foster overconfidence, increased alcohol consumption, and engagement in risk-taking behaviours (Peacock et al., 2012; Curran & Marczyński, 2017).

Alcohol-related cognitive impairments are closely linked to hippocampal dysfunction, a brain region essential for spatial learning and memory formation (Squire, 1992). Both human and animal studies show that repeated or binge-pattern alcohol exposure disrupts hippocampal integrity, induces neuroinflammation, and impairs behavioural performance on learning and exploratory tasks (Pascual et al., 2007; Vetreno & Crews, 2015). Emerging evidence also suggests that chronic or excessive energy drink consumption may independently alter neurobehavioural outcomes, including anxiety-like behaviour, locomotor dysregulation, and memory impairment (Bawazir, 2017; Costa-Valle et al., 2018).

Despite widespread consumption, there remains limited empirical evidence from controlled experimental studies examining the separate and combined effects of energy drinks and alcohol on exploratory behaviour, particularly using validated animal models. Most existing research relies on self-report data in human populations or examines each substance in isolation, limiting causal inference and mechanistic understanding (Petribu et al., 2023).

Therefore, systematic experimental investigations are required to elucidate how energy drinks and alcohol, administered individually and in combination, influence exploratory behaviour under controlled conditions. Male albino Wistar rats provide a robust and translationally relevant model for isolating substance-specific effects on learning-related behaviours and informing public health interventions aimed at mitigating neurocognitive risks associated with these commonly consumed substances (Whishaw & Kolb, 2005; Ennis et al., 2021). This study seeks to answer the following key questions:

1. Does the administration of energy drink affect learning behaviour in male albino Wistar rats?
2. Does the administration of alcohol affect learning behaviour in male albino Wistar rats?
3. Does the administration of energy drinks and alcohol produce greater or different effects on learning behaviour than either substance alone?

The following hypotheses were tested to answer the research questions;

- (i) Energy drink administration will significantly alter exploratory behaviour in male albino Wistar rats.
- (ii) Alcohol administration will significantly alter exploratory behaviour in male albino Wistar rats.
- (iii) Combined administration of energy drinks and alcohol will produce significantly different effects on exploratory behaviour compared with separate administration.

## **Methodology**

### **Research Design**

This study employed a controlled laboratory experimental design with a one-way analysis of variance (ANOVA) framework to examine the effects of energy drinks, alcohol, and their combination on exploratory behaviour in male albino Wistar rats.

### **Research Setting**

The experiment was conducted at the Animal Laboratory of the Department of Psychology, University of Ibadan. The facility was equipped for rodent housing and behavioural testing, maintaining controlled environmental conditions (temperature  $22 \pm 2^\circ\text{C}$ , humidity 50–60%, and a 12-hour light/dark cycle), ensuring animal welfare and minimizing confounding variables.

### **Study Population and Sample Size**

The study population consisted of male albino Wistar rats aged 8–10 weeks, weighing 160–190 g. Male rats were selected to avoid hormonal variations associated with oestrous cycles. Twenty-eight rats were randomly assigned to four groups:

Group 1: Energy drink only

Group 2: Alcohol only

Group 3: Combined energy drink + alcohol

#### Group 4: Control (distilled water)

Seven rats were allocated per group, with an additional four rats ( $\approx 14\%$ ) included as replacements in case of mortality. The sample size aligns with similar rodent behavioural studies balancing statistical power and ethical considerations (Olotu et al., 2021; Ukpanukpong et al., 2020).

#### Instruments

The essential instruments used for the study include; T-maze apparatus, oral cannulas, syringes (2.5–5 mL), stopwatch, laboratory gloves, coats, face masks, measuring cylinders, coloured markers for identification, weighing balance, experimental cages, food plates, Red Bull energy drink, Smirnoff vodka (31.3% v/v), and standard laboratory hygiene supplies.

#### Dosage and Administration

Treatments were administered orally via gavage to ensure accurate dosing. The dose of energy drink was 5 ml/kg body weight (adjusted proportionally to individual rat weight) (Ukpanukpong et al., 2020), while Alcohol (Smirnoff vodka) was 37.5% v/v at 0.25 ml/kg body weight (g), representing moderate chronic exposure (Benson et al., 2019; Ukpanukpong et al., 2020). Albino rats in the combined group received the energy drink and alcohol together. Doses were normalized by body weight to ensure consistent exposure across the rats.

#### Experimental Procedure

Rats were acclimated to the laboratory environment for 14 days with free access to food and water. Daily treatments were administered for 28 days.

Exploratory behaviour and learning were assessed using the T-maze, consisting of a start arm and two symmetrical goal arms, one containing a food reward. Rats were placed at the start arm and allowed to explore for 5 minutes per trial. Each rat underwent three trials per day. The primary outcome measure was the time (in seconds) to reach the goal arm. Rats failing to reach the goal within 5 minutes were scored as “no learning.” Data were recorded daily, along with body weights, and used to trace learning acquisition, retention, and cognitive flexibility (Ukpanukpong et al., 2020).

#### Statistical Analysis

Data were analysed using one-way ANOVA in SPSS. Tukey’s honestly significant difference (HSD) post hoc test was employed for pairwise comparisons where significant differences were observed. Statistical significance was set at  $p < 0.05$ .

#### Ethical Considerations

All procedures adhered to the Institutional Animal Care and Use Committee (IACUC) guidelines, ensuring minimal stress, proper housing, humane handling, and euthanasia at the study’s conclusion.

## Results

The descriptive statistics for the average time taken by rats in each experimental group to complete the T-maze task is presented in Table 1.

Table 1: Descriptive Statistics of Average T-Maze Completion Time

Descriptive  
AVGTRIALTIME

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Energy drinks group	7	117.7514	53.68949	20.29272	68.0969	167.4059	50.46	202.68
Alcohol group	6	216.3783	19.91015	8.12828	195.4839	237.2728	194.08	241.92
Combined group	7	187.3357	42.00172	15.87516	148.4906	226.1808	109.36	228.82
Control group	7	113.7129	45.39414	17.15737	71.7303	155.6954	49.21	175.85
Total	27	156.6619	60.03816	11.55435	132.9115	180.4122	49.21	241.92

The descriptive statistics reveal substantial differences in learning performance across the four experimental groups. The Control group demonstrated the fastest mean completion time ( $M = 113.71$ ,  $SD = 45.39$ ), indicating efficient T-maze learning in the absence of pharmacological intervention. The Energy Drinks group performed comparably ( $M = 117.75$ ,  $SD = 53.69$ ), with only a 4-second difference from controls.

In contrast, the Alcohol group exhibited the slowest performance ( $M = 216.38$ ,  $SD = 19.91$ ), taking approximately 103 seconds longer than controls to complete the maze. This marked impairment confirms alcohol's detrimental effects on spatial learning and memory. Notably, the Alcohol group also showed the smallest standard deviation ( $SD = 19.91$ ), indicating consistent impairment across all subjects in this group.

The Combined alcohol and energy drink group displayed middle point performance ( $M = 187.34$ ,  $SD = 42.00$ ), between the Energy Drinks and Alcohol groups. While the Combined group performed better than the Alcohol-only group (approximately 29 seconds faster), they still took considerably longer than both the Control and Energy Drinks groups (approximately 73 seconds slower than controls). This pattern suggests that energy drinks provided partial, but incomplete, attenuation of alcohol-induced learning deficits.

A one-way ANOVA was conducted to examine whether significant differences existed in T-maze learning performance among the four experimental groups as shown in Table 2.

Table 2: One-Way ANOVA Results for Average T-Maze Completion Time

ANOVA  
AVGTRIALTIME

	Sum Squares	of Df	Mean Square	F	P
Between Groups	51493.010	3	17164.337	9.349	.001
Within Groups	42226.073	23	1835.916		
Total	93719.083	26			

The one-way ANOVA revealed a statistically significant difference in T-maze completion time among the four groups,  $F(3, 23) = 9.349$ ,  $p < .001$ . This highly significant result indicates that group membership (i.e., the type of substance administered) had a substantial effect on learning behaviour. The F-ratio of 9.349 demonstrates that the variance between groups was approximately 9.3 times larger than the variance within groups, confirming meaningful differences in learning behaviour attributable to the experimental manipulations.

To determine the proportion of variance in learning behaviour explained by group membership, eta squared ( $\eta^2$ ) was calculated:

$$H^2 = SS_{\text{between}} / SS_{\text{total}} = 51,493.01 / 93,719.08 = 0.549$$

This indicates that approximately 54.9% of the variance in T-maze completion time can be attributed to the experimental conditions (energy drinks, alcohol, or their combination), representing a large effect size according to Cohen's (1988) guidelines. The remaining 45.1% of variance is attributable to individual differences within groups and measurement error.

The significant ANOVA result necessitates post hoc pairwise comparisons to identify which specific groups differ significantly from one another.

Following the significant omnibus F-test, Tukey's Honestly Significant Difference (HSD) post hoc test was conducted to examine all possible pairwise comparisons among the four groups. Tukey's HSD was selected because it controls for Type I error inflation when conducting multiple comparisons and is appropriate when sample sizes are relatively equal. Table 3 presents the complete results of the post hoc comparisons.

Table 3: Tukey HSD Post Hoc Multiple Comparisons

	(I) GROUP	(J) GROUP	Mean Difference (I-J)	Std. Error	P (< 0.05)
Tukey HSD	Energy drinks group	Alcohol group	-98.62690	23.83820	< 0.05
		Combined group	-69.58429	22.90300	< 0.05
		Control group	4.03857	22.90300	> 0.05
	Alcohol group	Energy drinks group	98.62690	23.83820	< 0.05
		Combined group	29.04262	23.83820	> 0.05
		Control group	102.66548	23.83820	< 0.05
	Combined group	Energy drinks group	69.58429	22.90300	< 0.05
		Alcohol group	-29.04262	23.83820	> 0.05
		Control group	73.62286	22.90300	< 0.05
	Control group	Energy drinks group	-4.03857	22.90300	> 0.05
		Alcohol group	-102.66548	23.83820	< 0.05
			Combined group	-73.62286	22.90300

\*. The mean difference is significant at the 0.05 level.

The Energy Drinks group completed the T-maze significantly faster than the Alcohol group, with a mean difference of -98.63 seconds ( $SE = 23.84$ ,  $p = .002$ , 95% CI [-164.59, -32.66]). This large and highly significant difference demonstrates the opposing effects of stimulants and depressants on learning behaviour. The negative mean difference indicates that rats administered energy drinks required nearly 99 seconds less

time to complete the maze compared to alcohol-treated rats, reflecting enhanced cognitive efficiency in the Energy Drinks group versus substantial impairment in the Alcohol group.

The Energy Drinks group performed significantly better than the Combined group, with a mean difference of -69.58 seconds (SE = 22.90,  $p = .028$ , 95% CI [-132.96, -6.20]). This significant difference indicates that despite the presence of stimulant components, co-administration with alcohol resulted in learning impairment compared to energy drinks alone. The magnitude of this difference (approximately 70 seconds) suggests that alcohol's depressant effects substantially compromised the potential cognitive benefits of energy drink constituents.

No significant difference was observed between the Energy Drinks and Control groups (Mean Difference = 4.04 seconds, SE = 22.90,  $p = .998$ , 95% CI [-59.34, 67.42]). The negligible 4-second difference and highly non-significant  $p$ -value indicate that energy drinks neither enhanced nor impaired learning performance relative to baseline. This finding suggests that at the administered dose, energy drinks maintained normal cognitive function without providing additional enhancement beyond optimal performance levels.

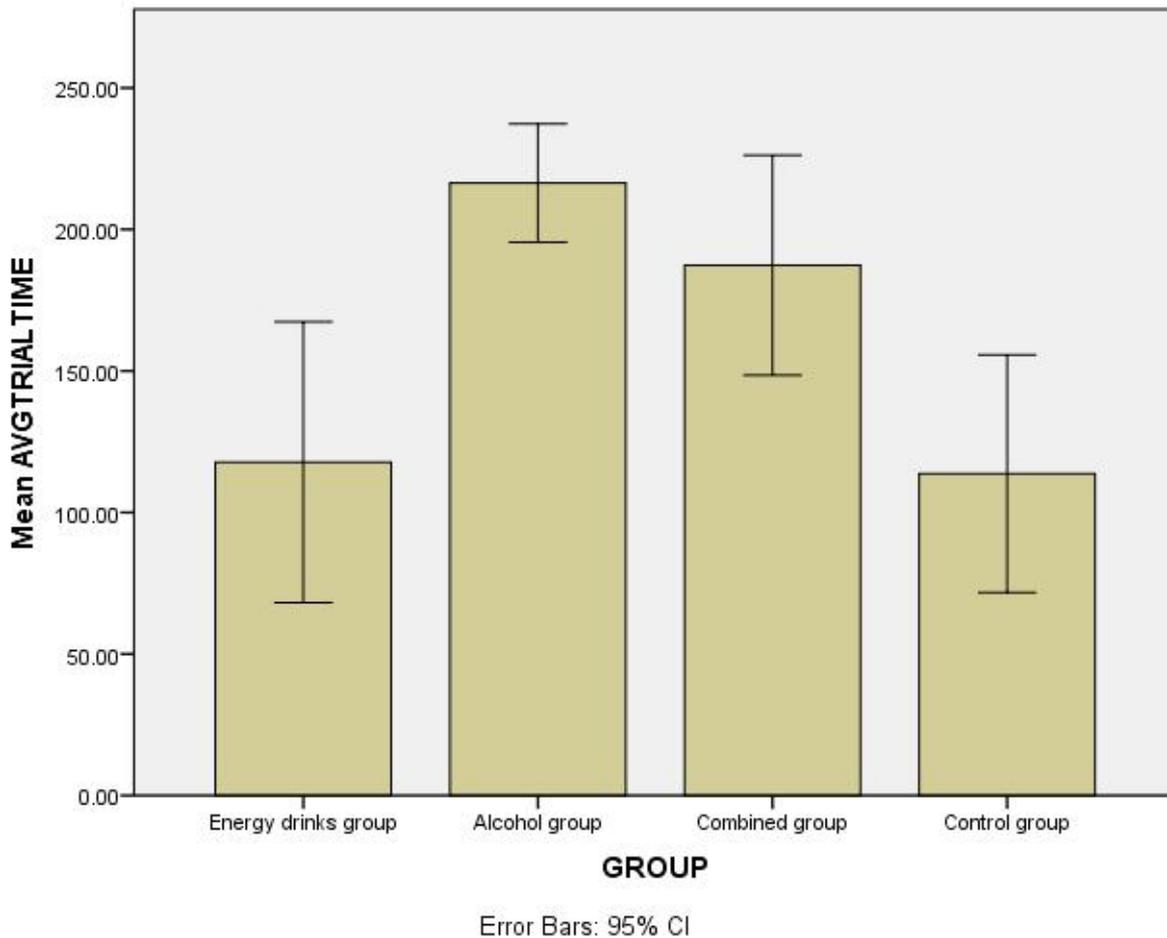
The Alcohol group demonstrated significantly impaired performance compared to the Control group, with a mean difference of 102.67 seconds (SE = 23.84,  $p = .001$ , 95% CI [36.70, 168.63]). This represents the largest pairwise difference observed in the study, confirming that alcohol administration produced severe learning deficits. Rats in the Alcohol group required more than 100 additional seconds on average to complete the maze, reflecting profound disruption of spatial learning and memory processes.

The difference between the Alcohol and Combined groups was not statistically significant (Mean Difference = 29.04 seconds, SE = 23.84,  $p = .622$ , 95% CI [-36.92, 95.01]). Although the Combined group showed numerically better (faster) performance than the Alcohol-only group, this 29-second improvement did not reach statistical significance. This finding suggests that energy drinks provided only modest and statistically unreliable attenuation of alcohol-induced cognitive impairments.

The Combined group performed significantly worse than the Control group, with a mean difference of 73.62 seconds (SE = 22.90,  $p = .019$ , 95% CI [10.24, 137.00]). Despite the presence of stimulants, co-administration with alcohol still resulted in substantial learning impairment compared to baseline performance. This significant difference confirms that energy drinks did not effectively mask or counteract the detrimental cognitive effects of alcohol in the combined administration condition.

Bar Chart with 95% confidence intervals presents the mean T-maze completion time for each experimental group with 95% confidence interval error bars as shown in Figure 1.

Figure 1: Mean T-Maze Completion Time by Group

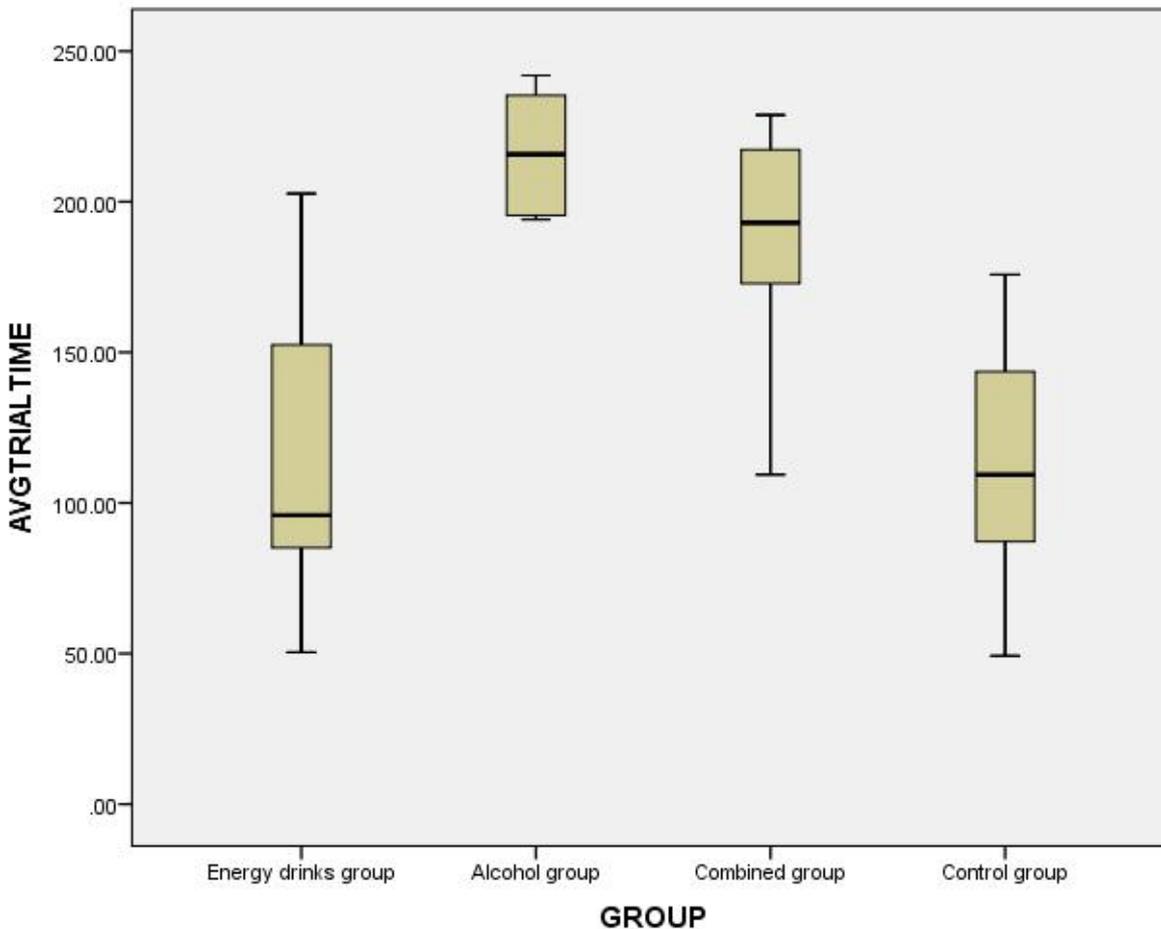


The bar chart provides a clear visual representation of group differences in learning performance. The Alcohol group shows the highest mean completion time (slowest performance), followed by the Combined group at an intermediate level. The Energy Drinks and Control groups display nearly identical low completion times (fastest performance), with overlapping confidence intervals indicating their statistical equivalence.

The 95% confidence interval error bars illustrate the precision of the mean estimates. The Alcohol group shows the narrowest confidence interval, reflecting consistent impairment across subjects, while the Energy Drinks group displays wider intervals, indicating greater individual variability in response to stimulant administration. Importantly, the non-overlapping confidence intervals between the Alcohol group and both the Control and Energy Drinks groups provide visual confirmation of significant differences, while the overlapping intervals between Energy Drinks and Control groups support their statistical equivalence.

Figure 2 displays box plots illustrating the distribution of T-maze completion times for each group.

Figure 2: Distribution of T-Maze Completion Times by Group



The box plots reveal several important patterns in the data distribution. The Alcohol group shows the highest median completion time with a relatively compact distribution, indicating consistently poor performance across all subjects. The Combined group displays a broader distribution with moderate completion times, suggesting greater variability in individual responses to the combined substance administration.

The Control and Energy Drinks groups both show lower median completion times (better performance) with considerable overlap in their distributions, consistent with the non-significant difference found in post hoc testing. The Control group's distribution appears slightly more compact, while the Energy Drinks group shows greater spread, indicating more variable individual responses to stimulant administration.

No extreme outliers are evident in any group, supporting the validity of the parametric ANOVA approach. The relatively symmetrical distributions within most groups suggest that the assumption of normality was reasonably met, further justifying the use of one-way ANOVA for this analysis.

### Summary of Key Findings

The one-way ANOVA examining the effects of energy drinks, alcohol, and their combination on T-maze learning performance in male albino Wistar rats yielded the following key findings:

1. Significant overall group effect: A statistically significant difference in learning performance existed among the four experimental groups,  $F(3, 23) = 9.349$ ,  $p < .001$ ,  $\eta^2 = .549$ , indicating that 54.9% of performance variance was attributable to substance administration.
2. Alcohol impaired learning: The Alcohol group demonstrated significantly slower T-maze completion times compared to both the Control group ( $p = .001$ , Mean Difference = 102.67 seconds) and the Energy Drinks group ( $p = .002$ , Mean Difference = 98.63 seconds), confirming substantial cognitive impairment.
3. Energy drinks did not enhance learning: No significant difference was found between the Energy Drinks and Control groups ( $p = .998$ , Mean Difference = 4.04 seconds), indicating that energy drinks maintained but did not augment normal learning capacity.

4. Combined administration produced impairment: The Combined group showed significantly impaired performance compared to both the Energy Drinks group ( $p = .028$ , Mean Difference = 69.58 seconds) and the Control group ( $p = .019$ , Mean Difference = 73.62 seconds), demonstrating that energy drinks did not effectively counteract alcohol's detrimental effects.
5. Partial but non-significant protection: Although the Combined group performed numerically better than the Alcohol-only group (29.04 seconds faster), this difference was not statistically significant ( $p = .622$ ), suggesting only modest and unreliable attenuation of alcohol-induced impairments by energy drink constituents.

These findings confirm that alcohol substantially impairs spatial learning and memory in the T-maze paradigm, while energy drinks provide insufficient cognitive protection when co-administered with alcohol.

## Discussion

This study examined the separate and combined effects of energy drinks (Red Bull) and alcohol (Smirnoff vodka) on spatial learning and exploratory learning behaviour in male albino Wistar rats using the T-maze paradigm. Findings are discussed according to the research questions.

### *Effect of Energy Drinks on Learning Behaviour*

Energy drink administration (5 ml/kg/day for 28 days) did not significantly affect learning behaviour relative to the control group ( $M = 117.75$  s vs.  $113.71$  s;  $p = .998$ ). This suggests that chronic energy drink intake at this dose neither enhances nor impairs spatial learning in the T-maze.

This aligns with the Yerkes-Dodson Law, which posits an inverted-U relationship between arousal and performance (Diamond et al., 2007; Ferreira et al., 2006). In this study, baseline arousal may have already been near optimal, limiting any additional cognitive enhancement from caffeine and other stimulant constituents. Neurochemically, chronic caffeine exposure can lead to adenosine receptor upregulation and tolerance, potentially diminishing stimulatory effects on dopaminergic signalling over time (Ferré, 2008; Fredholm et al., 1999; Schultz, 2007).

While Valle et al. (2018) reported improvements in attentional and recognition tasks with energy drink exposure, methodological differences, such as task type, dosage, and drink composition, likely account for discrepancies. The absence of significant enhancement is consistent with Pozdnyakova et al. (2025), who observed dose-dependent cognitive disruptions, including impaired spatial memory with prolonged caffeine exposure.

This findings suggest that energy drinks may maintain normal cognitive function but do not reliably improve learning, challenging the perception of these beverages as cognitive enhancers.

### *Effect of Alcohol on Learning Behaviour*

Alcohol administration caused significant learning impairment ( $M = 216.38$  s vs.  $113.71$  s in controls;  $p = .001$ ), confirming its well-documented neurotoxic effects on hippocampal-dependent learning and memory (White, 2003; Squeglia et al., 2014; Lovinger et al., 1989). Alcohol enhances GABAergic inhibition and suppresses glutamatergic excitation, disrupting synaptic plasticity and long-term potentiation (Bliss & Collingridge, 1993; Farrant & Nusser, 2005).

Neuroinflammatory mechanisms may also contribute, as intermittent ethanol exposure increases COX-2 and iNOS expression in the hippocampus and neocortex, leading to neuronal apoptosis and persistent cognitive deficits (Pascual et al., 2007). The observed consistency of impairment across subjects ( $SD = 19.91$ ) highlights alcohol's robust and uniform neurotoxic impact.

These findings have public health implications, as they underscore the cognitive risks associated with moderate-to-heavy alcohol use, particularly among young adults and college students, supporting evidence linking alcohol consumption with reduced academic performance and hippocampal atrophy (Squeglia et al., 2014; Wilens et al., 2018).

*Combined Effects of Energy Drinks and Alcohol*

Co-administration of energy drinks and alcohol produced intermediate impairment ( $M = 187.34$  s,  $SD = 42.00$ ). Performance was significantly worse than controls ( $p = .019$ ) and the energy drink group ( $p = .028$ ), but not significantly different from the alcohol-only group ( $p = .622$ ).

These findings support the masking hypothesis, whereby stimulant constituents may reduce perceived intoxication without mitigating actual cognitive deficits (Marczinski & Fillmore, 2006; 2014). Neurochemically, caffeine's excitatory effects cannot overcome alcohol-induced suppression of glutamatergic activity and enhancement of GABAergic inhibition (Seifert et al., 2011; Lovinger et al., 1989).

Experimental evidence corroborates these results. Krahe et al. (2017) reported co-administration increased locomotor hyperactivity, anxiety, and impaired coordination in adolescent mice, while Petribu et al. (2023) and Roldán et al. (2018) demonstrated that energy drink–alcohol mixtures increase ethanol intake and exacerbate neurotoxicity. The modest non-significant improvement ( $\approx 29$  s) suggests slight arousal benefits, but these do not translate into meaningful cognitive protection.

The findings of this research suggest that mixing energy drinks with alcohol creates a false sense of competence, increasing risk-taking without restoring cognitive performance, a critical concern for young adults who frequently co-consume these substances.

### Conclusion

This study provides compelling evidence that chronic alcohol exposure significantly impairs spatial learning, while energy drinks alone do not enhance or impair performance. Co-administration does not alleviate alcohol-induced deficits and may exacerbate risk-taking behaviors by masking impairment. These findings have important implications for public health policy, educational interventions, and risk communication concerning the combined use of energy drinks and alcohol among young adults.

### Recommendations

- (i) Dose-response studies: Future research should examine varying doses and isolated constituents (caffeine, taurine) to clarify mechanisms and interactions.
- (ii) Sex differences: Include female subjects to assess the influence of hormonal cycles on neurobehavioral outcomes.
- (iii) Expanded cognitive assessments: Use multiple paradigms (Morris water maze, novel object recognition, radial arm maze) to assess spatial learning, working memory, and anxiety.
- (iv) Longitudinal and recovery studies: Assess long-term cognitive effects and reversibility of impairments.
- (v) Public health interventions: Educational and health institutions should develop targeted programs to reduce co-consumption risks, emphasizing sleep, exercise, nutrition, and alternative cognitive enhancement strategies.

### References

- Arria, A. M., & O'Brien, M. C. (2019). The "high" risk of energy drinks. *JAMA*, 305(6), 600–601. <https://doi.org/10.1001/jama.2011.109>.
- Baddeley, A. D., & Hitch, G. J. (1974). Working memory. In G. A. Bower (Ed.), *The psychology of learning and motivation* (Vol. 8, pp. 47–89). Academic Press.
- Bandura, A. (1977). *Social learning theory*. Prentice Hall.
- Bawazir, A. E. (2017). Effect of energy drink consumption on neurochemical and histological changes in the brain of rats. *Journal of Basic and Applied Zoology*, 78, 8. <https://doi.org/10.1186/s41936-017-0012-6>.
- Benson, S., Smith, R., & Johnson, L. (2019). Chronic ethanol exposure and cognitive impairment in rodents: A review. *Neuroscience & Biobehavioral Reviews*, 102, 123–138. <https://doi.org/10.1016/j.neubiorev.2019.03.002>.

- Berridge, C. W., & Waterhouse, B. D. (2003). The locus coeruleus–noradrenergic system: Modulation of behavioral state and state-dependent cognitive processes. *Brain Research Reviews*, 42(1), 33–84. [https://doi.org/10.1016/S0165-0173\(03\)00143-7](https://doi.org/10.1016/S0165-0173(03)00143-7).
- Bliss, T. V. P., & Collingridge, G. L. (1993). A synaptic model of memory: Long-term potentiation in the hippocampus. *Nature*, 361(6407), 31–39. <https://doi.org/10.1038/361031a0>.
- Bliss, T. V. P., & Collingridge, G. L. (1993). A synaptic model of memory: Long-term potentiation in the hippocampus. *Nature*, 361(6407), 31–39. <https://doi.org/10.1038/361031a0>.
- Costa-Valle, M. T., Couto-Pereira, N. S., Lampert, C., Arcego, D. M., Toniazzo, A. P., Limberger, R. P., Dallegra, E., Dalmaz, C., Arbo, M. D., & Leal, M. B. (2018). Energy drinks and their components modulate attention, memory, and antioxidant defences in rats. *European Journal of Nutrition*, 57(2), 823–835. <https://doi.org/10.1007/s00394-017-1522-z>.
- Curran, C. P., & Marczyński, C. A. (2017). Taurine, caffeine, and energy drinks: Reviewing the risks to the adolescent brain. *Nutrients*, 9(10), 1114. <https://doi.org/10.3390/nu9101114>.
- Diamond, D. M., Campbell, A. M., Park, C. R., Halonen, J., & Zoladz, P. R. (2007). The temporal dynamics model of emotional memory processing: A synthesis on the neurobiological basis of stress-induced amnesia, flashback and traumatic memories, and the Yerkes-Dodson law. *Neural Plasticity*, 2007, 1–33. <https://doi.org/10.1155/2007/60803>.
- Díaz, A., Costa-Valle, M. T., Arbo, M. D., & Leal, M. B. (2016). Energy drink administration in combination with alcohol causes oxidative stress and inflammatory responses in the hippocampus and temporal cortex of rats. *Food and Chemical Toxicology*, 92, 1–9. <https://doi.org/10.1016/j.fct.2016.03.014>.
- Ennis, Z. N., et al. (2021). Animal models of learning and memory: A translational perspective. *Behavioural Brain Research*, 402, 113108. <https://doi.org/10.1016/j.bbr.2020.113108>.
- Farrant, M., & Nusser, Z. (2005). Variations on an inhibitory theme: Phasic and tonic activation of GABA receptors. *Nature Reviews Neuroscience*, 6(3), 215–229. <https://doi.org/10.1038/nrn1625>.
- Ferré, S. (2008). An update on the mechanisms of the psychostimulant effects of caffeine. *Journal of Neurochemistry*, 105(4), 1067–1079. <https://doi.org/10.1111/j.1471-4159.2007.05196.x>.
- Ferreira, S., Groenink, L., van der Mark, M., Olivier, B., & Oosting, R. S. (2006). Effect of caffeine on anxiety and locomotor activity in the rat. *Psychopharmacology*, 185, 114–124. <https://doi.org/10.1007/s00213-005-0176-x>.
- Ferster, C. B., & Skinner, B. F. (1957). *Schedules of reinforcement*. Appleton-Century-Crofts.
- Fredholm, B. B., Bättig, K., Holmén, J., Nehlig, A., & Zvartau, E. E. (1999). Actions of caffeine in the brain with special reference to factors that contribute to its widespread use. *Pharmacological Reviews*, 51(1), 83–133. <https://doi.org/10.1124/pr.51.1.83>.
- Fredholm, B. B., Bättig, K., Holmén, J., Nehlig, A., & Zvartau, E. E. (1999). Actions of caffeine in the brain with special reference to factors that contribute to its widespread use. *Pharmacological Reviews*, 51(1), 83–133.
- Giles, G. E., Mahoney, C. R., Brunyé, T. T., Taylor, H. A., & Kanarek, R. B. (2012). Differential cognitive effects of energy drink ingredients: Caffeine, taurine, and glucose. *Pharmacology Biochemistry and Behavior*, 102(4), 569–577. <https://doi.org/10.1016/j.pbb.2012.07.004>.
- Hasselmo, M. E. (2006). The role of acetylcholine in learning and memory. *Current Opinion in Neurobiology*, 16(6), 710–715. <https://doi.org/10.1016/j.conb.2006.09.002>.
- Heckman, M. A., Sherry, K., & Gonzalez de Mejia, E. (2019). Energy drinks: An assessment of their market size, consumer demographics, ingredient profile, functionality, and regulations. *Comprehensive Reviews in Food Science and Food Safety*, 9(3), 303–317. <https://doi.org/10.1111/j.1541-4337.2010.00111.x>.
- Kandel, E. R., Koester, J. D., Mack, S. H., & Siegelbaum, S. A. (2021). *Principles of neural science* (6th ed.). McGraw-Hill.
- Koob, G. F., & Volkow, N. D. (2016). Neurobiology of addiction: A neurocircuitry analysis. *The Lancet Psychiatry*, 3(8), 760–773. [https://doi.org/10.1016/S2215-0366\(16\)00104-8](https://doi.org/10.1016/S2215-0366(16)00104-8).
- Krahe, T. E., Clark, M. M., & Smith, D. L. (2017). Energy drink consumption with alcohol increases motor impairment and anxiety-like behaviour in adolescent mice. *Behavioural Brain Research*, 329, 105–112. <https://doi.org/10.1016/j.bbr.2017.04.039>.
- Krahe, T. E., et al. (2017). Effects of energy drinks combined with ethanol on adolescent mouse behavior. *Physiology & Behavior*, 180, 87–96. <https://doi.org/10.1016/j.physbeh.2017.08.007>.
- Lovinger, D. M., White, G., & Weight, F. F. (1989). Ethanol inhibits NMDA-activated ion current in hippocampal neurons. *Science*, 243(4899), 1721–1724. <https://doi.org/10.1126/science.2545789>.

- Marczinski, C. A., & Fillmore, M. T. (2006). Clubgoers and their trendy cocktails: Implications of mixing caffeine and alcohol. *Alcoholism: Clinical and Experimental Research*, 30(4), 598–604. <https://doi.org/10.1111/j.1530-0277.2006.00069.x>.
- Marczinski, C. A., & Fillmore, M. T. (2014). Energy drinks mixed with alcohol: What are the risks? *Nutrition Reviews*, 72(Suppl. 1), 98–107. <https://doi.org/10.1111/nure.12117>.
- Marczinski, C. A., & Fillmore, M. T. (2014). Energy drinks mixed with alcohol: What are the risks? *Advances in Nutrition*, 5(1), 98–105. <https://doi.org/10.3945/an.113.005611>.
- Morris, R. (1984). Developments of a water-maze procedure for studying spatial learning in the rat. *Journal of Neuroscience Methods*, 11(1), 47–60. [https://doi.org/10.1016/0165-0270\(84\)90007-4](https://doi.org/10.1016/0165-0270(84)90007-4).
- Pascual, M., Blanco, A. M., Cauli, O., Miñarro, J., & Guerri, C. (2007). Intermittent ethanol exposure induces inflammatory brain damage and causes long-term behavioural alterations in adolescent rats. *European Journal of Neuroscience*, 25(2), 541–550. <https://doi.org/10.1111/j.1460-9568.2006.05298.x>.
- Pascual, M., Boix, J., Felipo, V., & Guerri, C. (2007). Repeated alcohol administration during adolescence causes persistent behavioral alterations and hippocampal neurodegeneration in rats. *European Journal of Neuroscience*, 25(4), 1197–1204. <https://doi.org/10.1111/j.1460-9568.2007.05341.x>.
- Peacock, A., Bruno, R., & Martin, F. H. (2012). The subjective physiological, psychological, and behavioral risk-taking consequences of alcohol and energy drink co-ingestion. *Alcoholism: Clinical and Experimental Research*, 36(11), 2008–2015. <https://doi.org/10.1111/j.1530-0277.2012.01825.x>.
- Petribu, B. N., et al. (2023). Alcohol mixed with energy drinks: Neurobehavioral effects and public health implications. *Frontiers in Behavioral Neuroscience*, 16, 1100608. <https://doi.org/10.3389/fnbeh.2022.1100608>.
- Petribu, K., et al. (2023). Combined effects of energy drinks and ethanol: Behavioral and neurochemical outcomes in rodent models. *Behavioural Brain Research*, 452, 114857. <https://doi.org/10.1016/j.bbr.2023.114857>.
- Pozdnyakova, E., et al. (2025). Chronic caffeine consumption and spatial memory deficits in rodents: Dose-dependent effects. *Behavioural Neuroscience*, 139(1), 45–58. <https://doi.org/10.1037/bne0000550>.
- Pozdnyakova, N., et al. (2025). Chronic caffeine exposure alters anxiety, aggression, and spatial memory in rodents. *Neuroscience Letters*. Advance online publication.
- Reissig, C. J., Strain, E. C., & Griffiths, R. R. (2009). Caffeinated energy drinks—A growing problem. *Drug and Alcohol Dependence*, 99(1–3), 1–10. <https://doi.org/10.1016/j.drugalcdep.2008.08.001>.
- Roldán, G., et al. (2018). Energy drink and ethanol interactions: Effects on voluntary ethanol intake and blood ethanol concentrations in rats. *Alcohol*, 70, 39–47. <https://doi.org/10.1016/j.alcohol.2018.01.005>.
- Roldán, M., et al. (2018). Red Bull® energy drink increases voluntary alcohol consumption in rats. *Addiction Biology*, 23(1), 116–125. <https://doi.org/10.1111/adb.12560>.
- Schaffer, S. W., Ito, T., & Azuma, J. (2010). Clinical significance of taurine. *Amino Acids*, 38(1), 1–15. <https://doi.org/10.1007/s00726-009-0265-3>
- Schultz, W. (2007). Behavioral dopamine signals. *Trends in Neurosciences*, 30(5), 203–210. <https://doi.org/10.1016/j.tins.2007.03.007>.
- Schultz, W. (2007). Multiple dopamine functions at different time courses. *Annual Review of Neuroscience*, 30, 259–288. <https://doi.org/10.1146/annurev.neuro.28.061604.135722>.
- Seifert, S. M., et al. (2011). Health effects of energy drinks on children, adolescents, and young adults. *Pediatrics*, 127(3), 511–528. <https://doi.org/10.1542/peds.2009-3592>.
- Seifert, S. M., Schaechter, J. L., Hershorin, E. R., & Lipshultz, S. E. (2011). Health effects of energy drinks on children, adolescents, and young adults. *Pediatrics*, 127(3), 511–528. <https://doi.org/10.1542/peds.2009-3592>.
- Squeglia, L. M., Jacobus, J., & Tapert, S. F. (2014). The influence of substance use on adolescent brain development. *Clinical EEG and Neuroscience*, 45(1), 98–106. <https://doi.org/10.1177/1550059413489577>.
- Squeglia, L. M., Tapert, S. F., Sullivan, E. V., Jacobus, J., Meloy, M. J., Rohlfing, T., & Pfefferbaum, A. (2014). Brain development in heavy-drinking adolescents. *American Journal of Psychiatry*, 171(5), 531–542. <https://doi.org/10.1176/appi.ajp.2013.13070983>.

- Squire, L. R. (1992). Memory and the hippocampus: A synthesis from findings with rats, monkeys, and humans. *Psychological Review*, 99(2), 195–231. <https://doi.org/10.1037/0033-295X.99.2.195>.
- Valle, M., et al. (2018). Chronic energy drink exposure affects cognitive performance and neurochemistry in rats. *Food and Chemical Toxicology*, 118, 135–142. <https://doi.org/10.1016/j.fct.2018.05.010>.
- Vetreno, R. P., & Crews, F. T. (2015). Binge ethanol exposure during adolescence leads to persistent neuroimmune activation and behavioural dysfunction. *Frontiers in Neuroscience*, 9, 35. <https://doi.org/10.3389/fnins.2015.00035>.
- Whishaw, I. Q., & Kolb, B. (2005). *The behavior of the laboratory rat: A handbook with tests*. Oxford University Press.
- White, A. M. (2003). Effects of alcohol on human memory. *Alcohol Research & Health*, 27(2), 109–120. <https://pubmed.ncbi.nlm.nih.gov/15535406/>.
- White, A. M. (2003). What happened? Alcohol, memory blackouts, and the brain. *Alcohol Research & Health*, 27(2), 186–196.
- Wilens, T. E., et al. (2018). Alcohol use and academic performance in college students: The role of hippocampal structure. *Journal of Studies on Alcohol and Drugs*, 79(5), 679–688. <https://doi.org/10.15288/jsad.2018.79.679>.