During Ascending and Descending Limbs of the Blood Alcohol Concentration Curve

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Blood alcohol concentrations (BACs) as low as 0.04 mg/ml impair cognitive and visual tasks such as planning, working memory, blurred vision, and spatial awareness. Neuropsychological tests, such as the trail making test (TMT), have been shown to assess the severity of impairment. Prior research in this area has exclusively assessed these impairments via driving performance. This study aimed to investigate neurocognitive impairments associated with varying BAC levels utilizing a computerized trail making test (cTMT), which reduces practice effects via randomized stimuli location. The influences of alcohol were tested over the variables of total time, median latency, and version (Trail A, Trail B). Twenty-six participants (8 male, 18 female) with a minimum age of 21 years were recruited. Participants were randomly selected to be in the placebo (n=7) or the experimental group (n=19). The experimental group performed the cTMT at four target BAC points: baseline (0.00 mg/ml), ascending (0.06 mg/ml), peak (0.08 mg/ml), and descending (0.06 mg/ml). While participants who received alcohol tended to report themselves at a lower BAC than they were, there was no significant difference between perceived and actual BAC. Additionally, alcohol did not significantly affect performance on the cTMT for the tested levels. The cTMT detected impairments during the complex task (Trail B) but not in the simpler task (Trail A), thus indicating cognitive inflexibility and deficits in working memory. Future studies could attempt to augment BAC levels to at least 0.10 mg/ml in order to examine more distinct effects of alcohol on the specific cognitive tasks required by the cTMT.

Abbreviations: BAC – Blood Alcohol Concentration; CNS – Central Nervous System; cTMT – computerized Trail Making Test; GABA – Gamma-Aminobutyric Acid; NMDA - *N*-Methyl-*D*-Aspartate; PI – Principle Investigator; TMT – Trail Making Test

Keywords: Executive Functioning, Trail Making Test, Visuomotor, Young Adult

Introduction

Alcohol is known to have acute effects on executive functioning, such as planning and working memory, and on visuomotor performance, such as object recognition (Day et al., 2013). It has been shown to have a myriad of effects on neurological functioning at different thresholds of blood alcohol content (BAC), a measurement that indicates the percentage of alcohol per unit of blood (Starkey & Charlton, 2014). However, most research in this area focuses specifically on driving performance at differing BAC levels, rather than assessing neurocognitive functioning (Schweizer et al., 2004). This paper aims to use a computerized trail making test (cTMT) to examine the specific neurocognitive impairments brought on by alcohol at different levels of BAC.

Alcohol crosses the blood-brain barrier and penetrates the central nervous system (CNS; Zeigler et al., 2005). In other words, the alcohol in blood circulation can move to the brain. Therefore, neurotransmitter systems can be affected by

alcohol (Zeigler et al., 2005). Depressive effects on cognition and motor skills are caused by the interaction of alcohol with gamma-aminobutyric acid (GABA) receptors. Alcohol has also been shown to inhibit *N*-methyl-*D*-aspartate (NMDA) receptors (Zeigler et al., 2005). This, in turn, is responsible for the deficiency of learning and memory when drinking enough to raise BAC above approximately 0.02 mg/ml (Gilbertson et al., 2008; Sklar et al., 2014).

All lobes of the brain are affected by alcohol, and effects within the lobes are identifiable based on neurocognitive performances. For instance, the frontal lobe is responsible for motor function, problem-solving, working memory, judgment, and impulse control. Impairment to this lobe results in over-confidence or recklessness, reduced rationality, and short-term memory loss (Lyvers & Tobias-Webb, 2010). Day et al. (2013) found that the frontal lobe is also crucial in planning and response inhibition.

Additionally, the occipital lobe influences vision and the visuospatial ability (Weissenborn & Duka, 2002), and inhibition of this lobe can produce effects such as blurred vision, impaired perception of speed, and difficulty finding objects. Parietal lobe inhibition results in difficulty with voluntary movement and signs of cerebellum inhibition include staggered movement. Temporal lobe impairments include language comprehension and visual perception (Dry et al., 2012). Neuropsychological assessments can evaluate the level of impairment within these lobes.

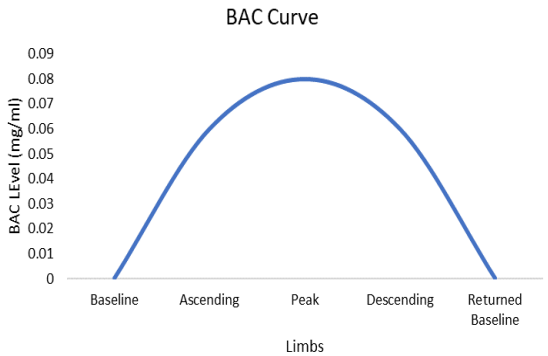

Figure 1. An example of the ascending and descending limbs on a BAC curve.

Neuropsychological tests elucidate a correlation between intoxication and neuropsychological dysfunction (Peterson et al., 1990). The TMT is considered a valid, reliable neurocognitive functioning test, and can thus be used to examine the neuropsychological impairments brought on by alcohol intoxication (Day et al., 2013). TMTs consist of two trail versions. Trail A is liable for measuring motor performance, requiring the connection of numbers in numerical order (Buck et al., 2008). Trail B necessitates a more convoluted executive process, which requires alternating between letters and numbers in ascending order. Trail B also measures cognitive set-shifting, visual scanning, and visuospatial sequencing (Buck et al., 2008). The computerized TMT (cTMT) reduces practice effects by producing random locations of stimuli, and thus a novel trail path, for each separate run. Therefore, it requires the frontal lobe to plan, process and execute the new trail continuously. Neuropsychological impairments associated with alcohol intoxication have not been studied with the cTMT.

The main features of the cTMT are visual scanning, object identification, motor planning, working memory, and executive functioning (Sanchez-Cubillo et al., 2009). Hence, the cTMT assesses specific brain functioning in the frontal, temporal, and occipital lobes. The frontal lobe's specific function in the cTMT is motor planning and execution, problem-solving, controlling working memory, and executive functioning. The temporal lobe, which is involved in visual perception, is responsible for the object identification presented in the trails. The identification is particularly important in Trail B because both numbers and letters are presented. In addition to the memory

required to recall the numbers and letters in ascending order, the stimulus has to be properly identified for the task to be completed correctly (Sanchez-Cubillo et al., 2009).

These cognitive and behavioral performances, including reaction time, divided attention, focusing on a target, attention to stimuli in the peripheral visual field, and scanning of the visual field, have been shown to be altered by the time BAC reaches 0.04 mg/ml (Gilbertson et al., 2008). Starkey & Charlton (2014) stated that higher-order attention and information processing capabilities are compromised by the time BAC reaches 0.05 mg/ml. BACs reported between 0.06 mg/ml and 0.09 mg/ml resulted in impaired memory, judgment, reaction time, and inhibition (BACtrack S80 Pro, 2017).

An individual's visuomotor and cognitive functioning does not necessarily correlate with his or her perceived intoxication. Gilbertson et al. (2009) found that the participants who received an alcohol placebo had a perceived impairment above zero. Additionally, Gilbertson et al., (2009) found that perceived impairment of participants under the age of 35 did not accurately represent real BAC values or the quality of performance on the trail making test (TMT). In other words, younger people are not capable of accurately judging their level of impairment and may believe they are more or less impaired than they are.

Furthermore, differences in visuomotor and cognitive functioning between the ascending (increasing to peak BAC levels) and descending (decreasing from peak BAC levels) limbs of the BAC curve have been inconsistent in previous research. Lyvers & Tobias-Webb (2010) found that there is no difference in performance on the ascending and descending limb with regards to prefrontal cortical functioning. However, participants in another study showed lower subjective intoxication measures on the descending limb (Starkey & Charlton, 2014). Research has uncovered that motor skills are worsened in the ascending limb compared to the descending limb (Starkey & Charlton, 2014). Curiously, the number of cognitive errors can remain the same or, in some cases, increase (Starkey & Charlton, 2014). Another study stated that impairment in motor skills increased and decreased in accordance to the rising and falling BAC (Schweizer et al., 2004). The discrepancies in the literature demonstrated above may result from specific differences within each of the studies, including age of the participants, time of their last meals, previous alcohol use, or type of tasks being administered. There is also little research about the effects of intoxication on self-evaluations of cognitive abilities with regard to increasing and decreasing BAC levels (Cromer et al., 2010). Therefore, it is important that these phenomena are analyzed in depth.

The purpose of this research was to study the effects of alcohol on executive functioning and visuomotor performance. It was intended to assess the influences of BAC on neurocognitive functioning and cTMT performance, and it sought to further understand perceived self-intoxication and self-impairment compared to BAC readings and actual neurocognitive performances. This was the first time the cTMT was used to test neurological impairment caused by alcohol intoxication. We proposed that the perceived intoxication rates would be lower than the actual intoxication rates for the experimental group. We also hypothesized that both latency and total time needed to complete the cTMT would increase with alcohol consumption. Furthermore, we postulated that these effects would be larger on the descending branch of the intoxication curve than on the ascending branch.

Material and Methods

Participants

The 26 participants (8 male, 18 female) had an average age of 21.92 years. Both genders were used in this study because the researchers were most interested in examining overall effects of alcohol intoxication, rather than on effects within a specific gender. They were recruited via word of mouth, flyers posted around campus, and using Roanoke College's SONA system (research participation link). Participants were eligible if they were current Roanoke College students, were over the age of 21, did not exceed the weight limit of 240 lbs., and were not naive to alcohol. The weight limit was set at 240 lbs. because it was the maximum weight on the dosage chart that was used to calculate drink volumes. Women who were pregnant and/or nursing were excluded from participating in the study. Participants who were taking any medication that would produce a negative reaction to alcohol were asked not to participate due to health precautions. The demographics questionnaire screened participants for these exclusion criteria. Participants that met exclusion criteria were still given credit but were discharged from the study. One participant withdrew from the study early and, thus, was excluded from data analysis. Another participant reported a perceived BAC at an impossible level, indicating that the question was misunderstood. Therefore, that participant's perceived BACs were not included in the data analysis. The sample size was n=25 following exclusions. The research was approved by the Roanoke College Institutional Review Board.

cTMT

The cTMT included two trails to be executed as efficiently and precisely as possible by the participant. For each trail, the participant completed three runs of 16 stimuli. Trail A required the participant to connect the numbered circles in ascending order by clicking on the correct numbered circle (Figure 1A). Trail A begins at 1 and ends with 16. This trail primarily evaluates visuomotor performance (Day et al., 2013). Trail B requires the participant to alternate between numbers and letters in ascending order (Figure 1B). Trail B begins with 1-A and ends with 8-H. The test assesses executive functioning through the mental flexibility demands of alternating between the numbers and letters. (Day et al., 2013). The computerized Trail Making Test was administered on a DELL laptop using MATLAB. The participants used a wired mouse to complete the tasks.

Latency or the time it took for each participant to initiate movement to the next bubble was recorded by the cTMT. This was done by measuring the time between the last click and the beginning of the next mouse movement. Additionally, the program counted the number of overshoots, which were defined by when the participant moved the cursor significantly past the target bubble. Incorrect clicks and the total time to complete each trail were also recorded.

BAC

Breath samples were collected from the participants using a handheld BAC test (BACTRACK S80 Pro; KHN Solutions Inc., San Francisco, CA). As suggested by the company, we waited twenty minutes after the last drink to administer the breathalyzer in order to produce accurate readings. This allowed enough time for the alcohol to enter the bloodstream and travel to the lungs, where the ethanol could be expelled upon respiration. The BACTRACK S80 has met all federal requirements for a breath alcohol screening device, as per testing by the Department of Transportation and National Highway Traffic Safety Administration (BACtrack S80 Pro, 2017).

Stimuli

Number of drinks was dependent upon the participant's weight and gender. The maximum number of drinks

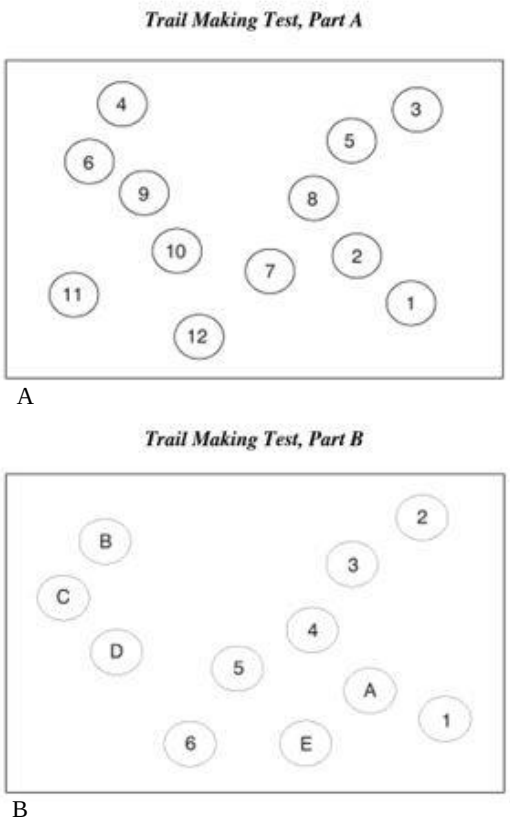

Figure 1. Examples of different possible paths produced by cTMT. Trail making test, Part A requires participants to click on each number in ascending order. Trail making test, Part B requires participants to alternate between number and letter in ascending order.

administered was four. The first drink contained a minimum of one and a half shots (2.25 ml) of vodka and 6.75 ml of Hi-C. The following drinks contained a minimum of half of a standardized shot (0.75 ml) of vodka and 1.5 ml of Hi-C. According to Virginia ABC law, an individual cannot be served more than two drinks at the same time (Retail Licensee Guide). The placebo group received 5ml (0.17 ml) of vodka which was poured on top of 8 ml of Hi-C. By mixing Smirnoff Vodka and Hi-C fruit punch, the placebo group could be deceived into believing that they were receiving a full dosage of alcohol. This was enough alcohol for the placebo participants to be able to smell and taste it, but it was not enough to intoxicate them (Sklar et al., 2014; Starkey & Charlton, 2014).

21 Jump Street (2012) and *The Hangover* (2009) were played in the waiting area between cTMT runs. Since we had to maintain supervision of all participants throughout the course of the study, these movies provided entertainment to the participants while waiting for increases and decreases in their BACs.

Surveys/Questionnaires

Once informed consent was obtained, a demographics questionnaire was completed by each participant. The questions asked for information such as gender, product allergies, current medications, race, and age.

The participants were asked to complete a survey following each cTMT run. These surveys consisted of subjective self-measures such as; “What do you think your BAC is?” and “How many total errors do you think you made on the cTMT on this trial?” It also included questions regarding perceived intoxication and perceived impairment. For example, participants were asked, “How intoxicated do you think you are?” (subjective intoxication) with “1” being “not at all” to “7” being “extremely drunk;” “How intoxicated do you feel right now compared to other times you have been intoxicated?” (comparative intoxication) where “1” indicated “least intoxicated I’ve ever felt” with “7” being “most intoxicated I’ve ever felt;” and “How well do you think you performed on the cTMT?” (perceived performance) with “1” being “very poorly” and “5” being “very well”.

Procedures

Flyers with the Principle Investigator’s (PI) email were posted around Roanoke College’s campus. Interested participants emailed the PI to schedule a day to partake in the study. Before their arrival, desks and cups were labeled with designated

numbers. The master list was stored with the rest of the data, which was kept in a locked room to preserve confidentiality. The master list contained the participant's assigned ID numbers, names, Roanoke College ID numbers, birthdate (recorded from either a valid driver's license, Passport, or Military ID), and age. The participant whose assigned ID number corresponding to the number drawn from a bag was placed into the placebo group (n=7). The others were placed in the experimental group (n=19). The researchers determined that a small placebo group was acceptable for this study because their BAC's would not be changing throughout the experiment. Therefore, only a small number of people were needed to provide a statistical baseline for comparing to the experimental group. A time-line protocol for the experimental group is provided in Table 1.

When the participants arrived, both IDs (student ID and a government-issued ID) were collected to confirm age and student status. The informed consent was reviewed, and any clarifications were made. Once the participants and researcher signed the consent form, each participant completed both the demographics questionnaire and contact form. The contact form was used in the incident that the participants' BAC did not decline to a safe level (less than 0.06 mg/ml). In this case, they were walked home, or one of their emergency contacts were called to pick them up from the study.

The participants went outside of the waiting room one at a time to step on the designated analog weight scale. This scale had the actual weights covered to eliminate any sensation of being self-conscious. The paper that covered the actual weight contained the weight range number that corresponded with the drink chart. The participant's BAC was recorded to confirm a BAC of 0.00 mg/ml.

After the BAC was recorded, the participant completed the baseline cTMT individually in a separate room. The baseline cTMT consisted of a practice demonstration of both trails along with the completion of the actual trails. The demonstration was a shorter version of the trails. The participant then returned to the waiting room and completed the first subjective survey. While each participant completed the initial measurements, a movie was playing in the waiting room to keep the other participants entertained.

The participants in both groups were given five minutes to drink each drink before their second BAC recording. This time was implemented to increase the rate at which alcohol entered the participant's body. The time was recorded when the participant received and completed their drinks. After the last drink to reach the first target point was recorded, a twenty-minute timer was set to indicate when the participant's BAC needed to be recorded. The twenty-minute timer was also used for the placebo group. This helped to remove any perceived distinction between the groups. The participants were breathalyzed for the second time to ensure the target point (0.06 mg/ml) had been reached. When the target was reached or neared, the BAC was recorded, and the participant then completed both trails on the cTMT at the ascending stage followed by the subjective survey. The placebo group proceeded to the next cTMT run and survey regardless of BAC.

The second round of drinks was administered to both groups after the completion of the second survey. The twenty-minute timer was set after the last drink had been finished. Once the timer went off to obtain the peak BAC target point (0.08 mg/ml) for all participants, the BAC was recorded. The participant completed the cTMT at the peak stage and returned to the room to complete the third subjective survey.

The duration of the descending limb varied for all participants based on their

body's metabolism. The experimental group waited another twenty-minutes to allow their BACs to drop down to 0.06 mg/ml.

The placebo group waited five minutes before their descending BAC was recorded. The placebo group completed their descending cTMT and their final subjective survey.

The experimental group was breathalyzed multiple times abiding by the twenty-minute time limit between breaths. In an attempt to decrease the descending time, snacks and water were administered to the participants. Once their BAC was around the descending target point (0.06 mg/ml), they completed their descending cTMT and their final subjective survey.

The release form was filled out by the researcher for all participants and signed by the participants, indicating that their BAC was 0.060 mg/ml or below. The time between the peak and the descending stages varied greatly, ranging from approximately 30 to 150 minutes.

Data analysis was performed using SPSS 23.0. The first 2x2x4 ANOVA was run to analyze the effects of group and stage on observed and reported BAC levels (Figure 2). A second 2x2x4 RM ANOVA was used to analyze effects of stage (baseline, ascending, peak, descending), version (Trail A, Trail B), and group (experimental, placebo) on median latency and average total time to complete the cTMT. A third ANOVA was conducted to analyze the number of incorrect clicks, version, and group. Finally, A paired samples t-test was used to determine the difference in the number of incorrect clicks between the placebo and experimental groups.

Results

For the experimental group, the reported BAC was not significantly different from the observed BAC (F (1,13) = 3.907, p = 0.070). Stage was found to have a significant main effect (F (2,26) = 12.734, p < 0.001). However, there was no interaction effect of stage and observed versus reported BAC's (F (2,26) < 0.001, p = 0.714).

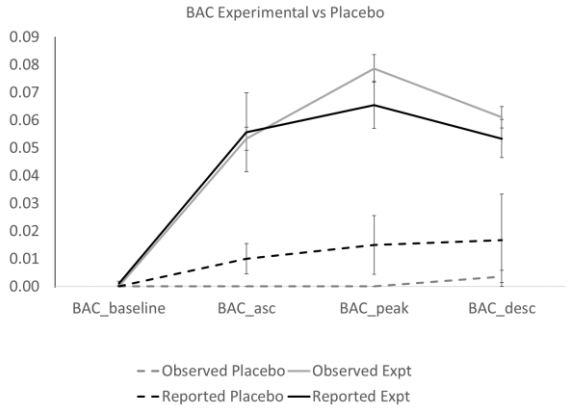

Figure 2. Experimental and placebo group observed results and their reported BAC levels.

Main effects of version on total time (F(1,17)=28.812, p<0.001) and median latency (F(1,17)=6.466, p=0.021) indicate that Trail B took longer to complete than Trail A (Table 2; Figure 3; Figure 4).

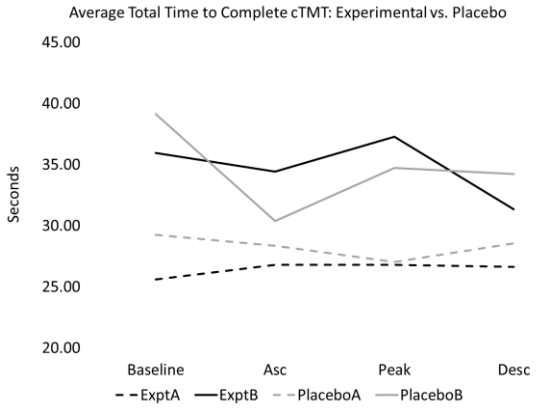

Figure 3. Average time (seconds) to complete both trails of cTMT.

An interaction effect of stage, version, and group was found on total time (F(1,17)=5.605, p=0.030) (Table 2; Figure 3). A three-way interaction effect of stage, version, and group was found on median wait time, such that stage and version changed across groups (F(3,51)=3.034, p= 0.037; Table 2; Figure 3; Figure 4).

	df	Total Time		Latency	
		F	p	F	p
Stage	3, 51	0.574	0.635	0.359	0.783
Stage*Group	3, 51	0.903	0.446	1.334	0.274
Version	1, 17	28.812	<0.001*	6.466	0.021*
Version*Group	1, 17	5.605	0.030*	0.109	0.746
Stage*Version	3, 51	0.627	0.601	1.172	0.330
Stage*Version*Group	3, 51	0.957	0.420	3.034	0.037*

*Significant at $\alpha = 0.050$

Table 2: Effects of stage (baseline, ascending, peak, descending), group (experimental, placebo) and version (Trail A, Trail B) on Total Time and Wait Time.

There was an interaction effect between incorrect clicks and group ($F(2,34)=4.130$, $p=0.025$). A marginally significant interaction effect was found between incorrect clicks and version ($F(2,34)=3.123$, $p=0.057$).

There was a significant difference between the placebo and experimental groups at both the peak ($t(17)= -2.675$, $p=0.016$) and descending ($t(16)= -2.279$, $p=0.037$) stages of intoxication.

Results indicated a significant correlation between estimated BAC levels and subjective intoxication levels for the ascending ($r(26)=0.815$, $p<0.001$) and peak ($r(24)=0.703$, $p<0.001$) stages of intoxication. A significant correlation was found between comparative intoxication and estimated BAC for the ascending ($r(26)=0.691$, $p<0.001$), peak ($r(24)=0.759$, $p<0.001$), and descending ($r(21)=0.802$, $p<0.001$) stages of intoxication.

The correlation between comparative intoxication and observed BAC levels was significant for the ascending ($r(26)=0.743$, $p<0.001$), peak ($r(24)=0.771$, $p<0.001$), and descending ($r(21)=0.470$, $p=0.032$) stages of intoxication. A correlation was also found between subjective intoxication and observed BAC levels for the ascending ($r(26)=0.694$, $p<0.001$) and peak ($r(24)=0.703$, $p<0.001$) stages of intoxication. There was not a significant correlation between these measures on the descending stage ($r(21)=0.394$, $p=0.077$).

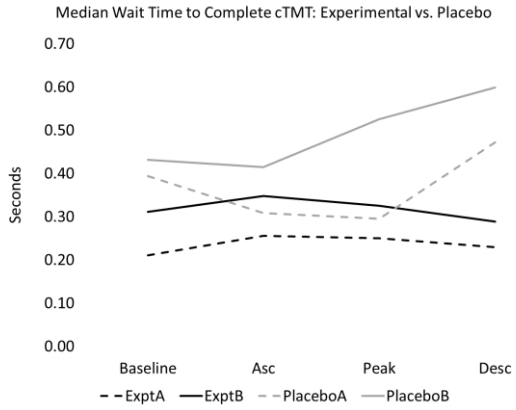

Figure 4. Median latency (seconds) across each cTMT item.

Finally, a significant correlation was found between estimated BAC and observed BAC levels in the experimental group for the ascending ($r(26)=0.460$, $p=0.018$) and peak ($r(24)=0.589$, $p=0.002$) stages of intoxication. There was no significant correlation between these measures found on the descending stage ($r(21)=0.096$, $p=0.678$).

Discussion

The experimental group's reported BAC did not support our hypothesis that the perceived BAC would generally be lower than the actual BAC. Estimated BAC, comparative intoxication, and subjective intoxication measures were all significantly

correlated at each stage. Therefore, these measures are internally consistent and are all reliable indicators of a person's perceived impairment. These three measures were also all significantly correlated with observed BAC at each stage with two exceptions. The correlation between observed BAC and subjective intoxication only approached significance on the descending stage, as did the correlation between observed BAC and estimated BAC. Hence, perceived impairment may be less accurate on the descending limb than the ascending limb, although differing BAC levels may have a confounding role in that relationship as well.

The hypotheses that latency and total time would increase as BAC increases were not supported. Additionally, latency and total time differences were not higher in the descending BAC curve as opposed to the ascending curve – a finding inconsistent with expectations. Our data does not provide evidence that alcohol has an effect on cognitive performance and visuomotor performance at these BAC levels, although it is possible that these findings were the result of a small sample size or an incomprehensive cognitive test.

Since Trail B involves alternating between numbers and letters, a more complex cognitive task, it was expected that Trail B would take longer to complete than Trail A, which also offered information about cognitive performance but on a more simplistic level. Our findings displayed that Trail B was more adept at assessing impairments than Trail A, suggesting that a major result of an increased BAC is cognitive inflexibility and deficits in more complex working memory tasks. However, more participants would be needed to support this effect statistically.

At the peak and descending stages of intoxication, the experimental group made predominantly more incorrect clicks than the placebo group. Notably, this effect was absent from the ascending stage. These findings indicate that performance is significantly impaired at a 0.08 mg/ml BAC. Additionally, performance was worse on the descending limb than the ascending limb despite a lack of differences in BAC.

When consumed, alcohol crosses the blood-brain barrier and affects the functioning of various neurotransmitters (Zeigler et al., 2005). Levels of glutamate, an excitatory neurotransmitter, are decreased by alcohol, and levels of gamma-aminobutyric acid (GABA), an inhibitory neurotransmitter, are increased. The result is decreased neural functioning in all areas of the brain. The TMT primarily measures frontal lobe functioning, but it is a less accurate measure of the functioning of other lobes.

In order to more accurately measure temporal lobe impairments, a delayed, non-cued memory task needs to be added. A study done by Peterson, Rothfleisch, Zelazo, & Pihl (1990) found that alcohol affects the function of the hippocampus, specifically, the transferring of information from short-term attention to long-term storage and retrieval (Gray & McNaughton, 2000). The septal-hippocampal system produces cue-specific states of anxiety and assists in maintaining safety. However, when under the influence of alcohol, it produces cognitive impairments and restricts the ability to adapt. A solution may be to simply administer a brief memory-retrieval test in addition to the cTMT. By doing so, both executive functioning and memory could be evaluated at different stages of intoxication.

Another limitation to this study was the required time to descend from the peak target point. The study required a minimum of three hours to complete per person. On average, the body metabolizes one ounce of alcohol every three hours (Zeigler et al., 2005). However, everyone's body metabolizes alcohol at a different rate. In addition to metabolism, there are different

forms of physiological and behavioral tolerances, each of which varies between individuals (Hart & Ksir, 2015). Thus, to reach the target BAC, some participants required more alcohol than others. Those who received more alcohol would take longer to metabolize it, which meant that they were in the lab longer. Therefore, it is possible that fatigue may have affected the cTMT results. It is also possible that the length of time during which alcohol remains within the bloodstream might confound the effects of the intoxication.

While both genders were used in the study to examine the overall effects of alcohol, there were several more females than males used, which may have created skewed results (Dufouil et al., 1997), although a previous study by Guillot and colleagues (2010) found no gender differences when examining effects of alcohol intoxication on the TMT. However, the researchers were not interested in gender differences, therefore the results within each gender were not analyzed separately. Additionally, the researchers attempted to minimize natural differences in alcohol absorption between the genders by considering gender when determining how much alcohol each participant received.

One notable result of the study was that the placebo group had significantly higher total time and latency on the descending limb. This could have been a result of disinterest since the study provided no financial compensation, possibly lowering participant interest towards the end of the study. This result may have also been caused by fatigue and diminished motivation since the study took place in the evening.

Our results also showed that there was a significant interaction effect between version and group which suggests that our placebo group was innately different from our experimental group. Such a difference may constrain the size of other effects that were found, limiting internal validity. This may be due to relative differences in size between the two groups as well as an overall small sample size. While the difference in size between the experimental and placebo groups itself should not have been a limiting factor due to the nature of the study, it is possible that randomization of participant assignments did not, by chance, adequately eliminate baseline differences between the two groups. Such an error could have been exacerbated by the substantial difference between the sample sizes and further exacerbated by the small sample size overall.

Another limitation of the study is the level of BAC the participants were allowed to reach. Other literature did not find large differences in cognitive performance until the participants reached BACs greater than 0.10 mg/ml (Starkey & Charlton, 2014). It is suggested that the cTMT does not detect this level of impairment at the BAC levels that were used in this study. Additionally, the target points that were utilized were too close together to measure any significant difference in the level of intoxication. Future studies measuring general effects of intoxication may want to exceed the legal driving limit of 0.08 mg/ml and have a greater distance between their target points. On the other hand, if studying impairments within the range of 0.00-0.08, it would be best to use a more in-depth, comprehensive cognitive test so that changes in functioning can be recorded in greater detail.

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