fMRI response to spatial working memory in adolescents with comorbid marijuana and alcohol use disorders[*]

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Abstract

Alcohol and marijuana use are prevalent in adolescence, yet the neural impact of concomitant use remains unclear. We previously demonstrated functional magnetic resonance imaging (fMRI) response to spatial working memory (SWM) among teens with alcohol use disorders (AUD) compared to controls, and predicted that adolescents with marijuana and alcohol use disorders would show additional abnormalities.

Participants were three groups of 15−17-year-olds: 19 non-abusing controls, 15 AUD teens with limited exposure to drugs, and 15 teens with comorbid marijuana and alcohol use disorders (MAUD) and minimal other drug experience. After >2 days' abstinence, participants performed a SWM task during fMRI acquisition.

fMRI brain response patterns differed between groups, despite similar performance on the task. MAUD youths showed less activation in inferior frontal and temporal regions than controls, and more response in other prefrontal regions. Compared to AUD teens, MAUD youths also showed less inferior frontal and temporal activation, but more medial frontal response.

Overall, MAUD youths showed different brain response abnormalities than teens with AUD alone, despite relatively short histories of substance involvement. This pattern could suggest compensation for marijuana-related attention and working memory deficits. However, relatively recent use and premorbid features may influence results, and should be examined in future studies.

Keywords

Alcohol abuse; Marijuana abuse; fMRI; Adolescents

1. Introduction

Alcohol and marijuana use are common in adolescence. In 2003, 31% of 12th graders reported getting drunk in the past month, 21% of 12th graders revealed using marijuana in the past month, and 6% of 12th graders disclosed daily marijuana use (Johnston et al., 2004). Further, 40% of high school students who used marijuana in the past year met criteria for marijuana

abuse or dependence (Chen et al., 2004). Moreover, 58% of adolescent drinkers also report marijuana use (Martin et al., 1996), and alcohol and marijuana use disorders are highly comorbid (Agosti et al., 2002). Despite the prevalence of heavy alcohol and marijuana use in teenagers, it is unclear how such protracted use may affect brain functioning during youth, particularly as adolescent neuromaturation continues.

Neuropsychological studies of teens with alcohol use disorders (AUD) have reported decrements in language skills, problem solving, verbal and non-verbal retention, working memory, and visuospatial performance (Brown et al., 2000; Moss et al., 1994; Tapert et al., 2002). In addition, we previously examined functional magnetic resonance imaging (fMRI) brain response during a spatial working memory (SWM) task among teens with AUD and demographically similar non-abusing controls (Tapert et al., 2004). Groups performed comparably on the task, but AUD teens demonstrated less brain response than controls in the midline precuneus/posterior cingulate, and more activation in bilateral posterior parietal cortex, suggesting subtle alcohol-related neural reorganization and compensation. These neuropsychological and imaging findings suggest that heavy alcohol use during youth adversely affects frontal and parietal circuitry, but the additional impact of marijuana use is less well understood.

Few studies have examined neurocognition in youths who use cannabis heavily. Neuropsychological assessments of substance use disordered teens have described marijuana use related deficits in learning and memory (Millsaps et al., 1994) and attention (Tapert et al., 2002). A longitudinal study of marijuana dependent adolescents demonstrated further short-term memory decrements that persisted after 6 weeks of monitored abstinence (Schwartz et al., 1989). In addition, compared to individuals with adult-onset cannabis use disorder and non-abusing controls, adolescent-onset cannabis use disordered adults showed attenuated electrophysiological response during selective attention (Kempel et al., 2003), as well as smaller frontal and parietal volumes and increased cerebral blood flow (Wilson et al., 2000). These studies indicate that heavy marijuana use during youth may adversely affect cognition and brain functioning, particularly short-term memory and attention, and raise questions about the integrity of frontal and parietal brain regions in adolescents with marijuana use disorders.

In order to understand the neural correlates of concomitant heavy marijuana and alcohol use during youth, we assessed blood oxygen level dependent (BOLD) fMRI response among short-term abstinent teens with comorbid marijuana and alcohol use disorders (MAUD) compared to AUD-only and non-abusing control teens reported in a previous study (Tapert et al., 2004). We measured BOLD response during an SWM task that typically activates bilateral prefrontal and posterior parietal networks among adults and youths (Thomas et al., 1999). Based on our earlier findings among AUD and control adolescents, we predicted that MAUD teens would show greater fMRI response than controls in regions subserving SWM, including prefrontal and bilateral posterior parietal cortices. We hypothesized further that MAUD teens would show more prefrontal and parietal activation than AUD youths, since we predicted that concurrent heavy marijuana and alcohol use would influence functioning more than protracted alcohol use alone.

2. Method

2.1. Participants

Flyers were distributed at local high schools to recruit adolescents, as described previously (Tapert et al., 2003). We obtained written informed consent and assent from interested teens and their guardians, approved by the University of California San Diego Human Research Protections Program.

Adolescents were administered a 90-min telephone screening interview to ascertain family history of substance use and psychiatric diagnoses using the Family History Assessment Module screener (Rice et al., 1995), lifetime substance use and abuse/dependence criteria using the Customary Drinking and Drug Use Record (CDDR) (Brown et al., 1998), and history of psychiatric disorders using the Diagnostic Interview Schedule for Children (Shaffer et al., 2000). Collateral interviews were administered to a guardian, usually a parent. Exclusion criteria included history of head injury with loss of consciousness >2 min, neurological or medical problems, learning disabilities, DSM-IV psychiatric disorder other than conduct disorder, current psychotropic medication use, significant maternal drinking or drug use during pregnancy, family history of bipolar I or psychotic disorder, and left handedness. Teens meeting criteria for conduct disorder (six cases DSM-IV mild and four cases moderate severity) were not excluded due to high comorbidity with substance use disorders (Brown et al., 1996).

Eligible participants were ages 15−17, and groups were demographically similar (see Table 1). Controls ($n = 19$) had little experience with alcohol or other drugs. AUD adolescents ($n = 15$) met DSM-IV criteria for current alcohol abuse or dependence, but had limited experience with marijuana (<40 times in life). Only two AUD teens disclosed marijuana use in the month before scanning (2 and 20 days prior). MAUD adolescents ($n = 15$) met DSM-IV criteria for both current marijuana and alcohol abuse or dependence, had ≥100 lifetime experiences with marijuana and had used ≥10 days/month in the three months before scanning. One MAUD participant reported stopping marijuana use 4 months prior to scanning; however, the urine toxicology screen indicated recent use. Twelve other MAUD teens reported marijuana use in the week before the scan, with last use 3.3 ± 1.7 days prior to scanning. Participants in each group had little experience with drugs other than alcohol and marijuana (<30 times in life total and <10 times in life for any other drug type), had not used other drugs for 30 days prior to imaging, and had not used marijuana or alcohol for at least 48 h before scanning. Importantly, AUD and MAUD youths demonstrated similar alcohol use disorder characteristics (see Table 2). Both AUD and MAUD teens were primarily weekend heavy drinkers, as evidenced by an overall average 15.13 days since last drink and typical blood alcohol concentration reaching 0.107. Two AUD teens and one MAUD teen reported abstinence from alcohol in the month before scanning. AUD and MAUD teens displayed similar cigarette smoking patterns, but more MAUD teens had experiences with other drugs than AUD and control teens, although such use was limited (8.38 ± 8.52 lifetime episodes among MAUD teens who had used; 5.50 ± 6.37 lifetime episodes among AUD teens who had used). Although MAUD and AUD teens had higher rates of conduct disorder than control teens, severity was mild to moderate reflected by the normal range Child Behavior Checklist (CBCL) (Achenbach, 1991) externalizing scores (see Table 1).

2.2. Measures

2.2.1. Substance involvement assessment—Substance involvement and abuse/dependence diagnoses were assessed using the CDDR (Brown et al., 1998). The CDDR collects lifetime and past 3-month information on alcohol, nicotine, and other drug use, and assesses DSM-IV abuse and dependence criteria, withdrawal symptomatology, and other negative consequences associated with substance use. The CDDR also obtains information necessary to estimate typical blood alcohol concentrations (BAC) reached using the Widmark method, i.e. amount consumed, duration of drinking, height, weight, and gender (Fitzgerald, 1995). Strong internal consistency, test–retest, and inter-rater reliability have been demonstrated with adolescent CDDR assessments (Brown et al., 1998; Stewart and Brown, 1995). The Timeline Followback (Sobell and Sobell, 1992) obtained detailed substance use patterns for the 30 days prior to scanning. On the day of the scanning session, all participants submitted samples for Breathalyzer and urine drug toxicology analyses.

2.2.2. Neuropsychological and behavioral measures—On the scan day, a neuropsychological battery assessed multiple cognitive domains, including attention, working memory, learning and memory, executive, visuospatial, and language functioning. The Beck Depression Inventory (Beck, 1978) and state scale of the Spielberger State Trait Anxiety Inventory (Spielberger et al., 1970) assessed mood at the time of scanning. The Stanford Sleepiness Scale measured alertness immediately before and after scanning with self-report ratings (Glenville and Broughton, 1978). Parents completed the CBCL (Achenbach, 1991).

2.2.3. Spatial working memory task—The SWM task (Kindermann et al., 2004; Tapert et al., 2001) consisted of eighteen 20-s blocks alternating between SWM and simple attention baseline conditions, and three rest blocks (total time = 7:48 minutes). During the SWM condition, abstract line drawings were presented sequentially in varied locations. Participants made a button press when a design appeared in a location that had already been occupied during that block. Three of the 10 stimuli in each block were targets, which matched the location of a figure presented two trials previously. In the simple attention condition, the stimuli were presented in a similar manner, but a dot appeared above figures on 30% of trials. Participants made a button press when they saw a design with a dot. The goal of this active baseline condition was to control for the motor, sensory, and attention processes involved in SWM.

2.3. Procedures

Participants were asked to abstain from substance use for at least 48 h before imaging to avoid intoxication and acute withdrawal during scanning. Imaging sessions were held Thursday evenings between 8 and 10 p.m. to maximize recovery from weekend binge drinking and maintain consistent circadian influence across subjects. According to self-report on the Timeline Followback (Sobell and Sobell, 1992), the most recent alcohol use was 72 h and marijuana use was 48 h before scanning. No withdrawal symptoms were disclosed or evident in any participant on the day of scanning. Upon arrival for the imaging session, all participants submitted samples for Breathalyzer and urine drug toxicology for THC, ethanol, amphetamines, methamphetamines, barbiturates, benzodiazepines, cocaine, codeine, morphine, and PCP. No participant had a positive breath alcohol concentration. Due to experimenter error, toxicology screens were unavailable for one control teen, one AUD teen, and five MAUD teens. Based on available data, only MAUD participants ($n = 5$) produced toxicology screens positive for cannabinoids, and no toxicology screens were positive for any drug other than cannabinoids. Although it is possible that MAUD teens were over-reporting marijuana use, self-reported marijuana use has been an accurate predictor of verified use (Martin et al., 1988).

After lying in the scanner, the participant's head was comfortably secured to minimize motion. Task stimuli were back-projected onto a screen at the foot of the MRI bed and viewed from a mirror attached to the head coil. A magnet-safe button box collected task responses during scanning. A high-resolution structural image was collected in the sagittal plane using an inversion recovery prepared T1-weighted 3D spiral fast spin echo sequence (TR = 2000 ms, TE = 16 ms, FOV = 240 mm, resolution = 0.9375 mm × 0.9375 mm × 1.328 mm) (Wong et al., 2000 Functional imaging was collected in the axial plane using T2*-weighted spiral gradient recall echo imaging (TR = 3000 ms, TE = 40 ms, flip angle = 90°, FOV = 240 mm, 20 continuous slices, slice thickness = 7 mm, in-plane resolution = 1.875 mm × 1.875 mm, 156 repetitions).

2.4. Data analysis

Neuropsychological test scores were converted to standard scores based on published norms. SWM task accuracy and reaction time were calculated for SWM and simple attention conditions. Group differences in neuropsychological test scores and SWM task performance

were examined with one-way ANOVAs. We followed up significant ANOVAs ($p < .05$) with Tukey's all pairwise t-tests between the three groups.

Imaging data were processed and analyzed using the Analysis of Functional NeuroImages (AFNI) package (Cox, 1996). We first applied a motion-correction algorithm to the time series data (Cox and Jesmanowicz, 1999). Second, we correlated the time series data with a set of reference vectors that represented the block design of the task and accounted for delays in hemodynamic response (Bandettini et al., 1993), while covarying for estimated motion and linear trends. Next, we transformed imaging data to standard coordinates (Lancaster et al., 2000; Talairach and Tournoux, 1988) then resampled the functional data into 3.5 mm^3 voxels. Finally, we applied a spatial smoothing Gaussian filter (full width half maximum = 3.5 mm) to account for anatomic variability.

After processing functional data, we examined average BOLD response to the SWM task in each group using one sample t-tests, and determined regions that showed greater response to SWM relative to simple attention (SWM activation), reduced response during SWM relative to rest (SWM deactivation), and greater simple attention response than SWM response. We next compared response during SWM relative to simple attention between groups with ANOVAs, and performed pairwise comparisons between groups. We performed group comparisons on the whole brain, rather than discrete regions thought to be activated by the task, because previous studies by our group (Tapert et al., 2001, 2004) and others (Desmond et al., 2003; Pfefferbaum et al., 2001) have suggested neural reorganization and use of alternate brain systems during working memory among individuals with AUD. To control for Type I error in group analyses, we required significant voxels ($\alpha < .05$) to form clusters $\geq$1072 μl (25 contiguous 3.5 mm^3 voxels), yielding a cluster-wise $\alpha < .0167$ (Forman et al., 1995; Ward, 1997). We utilized the Talairach Daemon (Lancaster et al., 2000; Ward, 1997) and AFNI (Ward, 1997) to confirm gyral labels for clusters.

Previous research has suggested that neuropsychological deficits among adult marijuana users are associated with lingering effects of recent use, and that these impairments dissipate with extended abstinence (Pope et al., 2001). To understand whether group differences in the current study relate to recent marijuana use, we performed post-hoc regressions within the MAUD group. First, we extracted the average fit coefficient for each MAUD participant from each cluster where we observed a difference between MAUD and control or AUD teens. Next, we used regression analyses to examine whether days since last marijuana use predicted brain response within each group difference cluster.

3. Results

Groups did not significantly differ on any neuropsychological performance measure (all p's > .4). SWM accuracy was 86 ± 9% in the control group, 91 ± 5% in the AUD group, and 92 ± 5% in the MAUD group, revealing a trend for MAUD to be more accurate than controls ($p = .056$). However, one control performed at 60% accuracy, which was >2.5 standard deviations below the mean for that group, and exclusion of this participant removed the group difference in SWM accuracy. This raised the concern that this individual impacted the fMRI group analyses. Upon further examination, we determined that this participant's brain response was within the normal range (within one standard deviation of the control group mean) for each significant cluster described below. Groups did not differ on simple attention accuracy or reaction time to either condition.

The overall pattern of BOLD response to the SWM condition relative to simple attention was similar in all three groups. Participants showed SWM activation (more response during SWM than during simple attention blocks) in several regions, including bilateral prefrontal, premotor,

cingulate, and posterior parietal areas ($p < .0167$). Groups showed SWM deactivation (less response during SWM relative to rest blocks) in medial prefrontal cortex, a large posterior midline region including posterior cingulate and cuneus, and several temporal regions ($p < .0167$). Although groups demonstrated similar patterns of response localization, several significant group differences emerged.

The response differences between AUD and control teens are detailed elsewhere (Tapert et al., 2004). Briefly, AUD teens showed less SWM response than controls in the left precentral gyrus and midline precuneus/posterior cingulate, but more SWM activation than controls in bilateral posterior parietal cortex ($p < .0167$).

MAUD participants evidenced altered BOLD response compared to controls in several regions: bilateral inferior frontal gyri, right superior temporal/supramarginal gyri, right middle and superior frontal gyri (dorsolateral prefrontal cortex), and anterior cingulate ($p < .0167$) (see Table 3 and Fig. 1; areas not highlighted are statistically similar between groups). In both right inferior frontal and superior temporal regions, MAUD teens demonstrated less SWM response than controls. Moreover, while controls showed SWM activation in the right superior temporal gyrus, MAUD teens showed greater simple attention response than SWM response. In right dorsolateral prefrontal cortex, MAUD youths showed more SWM activation than controls. Both controls and MAUD evidenced SWM deactivation in the anterior cingulate; however, MAUD showed a greater intensity of deactivation than controls. MAUD also demonstrated deactivation in the left inferior frontal gyrus, where controls showed no significant activation or deactivation.

MAUD teens showed different response intensity relative to AUD teens in the right inferior frontal gyrus/insula, left precuneus, right middle temporal/supramarginal gyri, left superior temporal gyrus, and a large cluster spanning anterior cingulate and bilateral inferior frontal gyri ($p < .0167$) (see Table 4 and Fig. 1). In the precuneus, groups showed SWM activation, yet AUD teens showed greater response than MAUD teens. Similar to controls, AUD teens showed SWM activation in right inferior frontal and middle temporal areas, while MAUD teens evidenced greater simple attention response than SWM response. In the left superior temporal gyrus, AUD showed SWM deactivation, while MAUD demonstrated no significant activation or deactivation. Finally, a group difference was observed in a large cluster spanning anterior cingulate and bilateral inferior frontal gyri. In this cluster, both AUD and MAUD showed deactivation, but MAUD showed greater intensity and spatial extent of deactivation.

Days since last marijuana use did not significantly predict brain response among MAUD teens in any cluster where MAUD teens had significantly different SWM response than controls or AUD teens. A trend was found for more recent use to be associated with reduced brain response in the right middle temporal gyrus ($p = .06$), where MAUD teens showed less SWM response than AUD teens.

4. Discussion

This study investigated the neural correlates of SWM in adolescents with comorbid marijuana and alcohol use disorders, teens with alcohol use disorders alone, and demographically similar non-abusing adolescents. The groups showed similar neuropsychological abilities, SWM task performance, and general BOLD response localization patterns. However, MAUD teens demonstrated significantly more dorsolateral prefrontal SWM activation and anterior cingulate deactivation, and significantly less right inferior frontal and superior temporal response compared to control teens. Similarly, MAUD youths also showed significantly more medial frontal deactivation as well as less right inferior frontal and bilateral temporal activation compared to AUD teens.

As noted above, MAUD teens showed more SWM activation than control teens in the right dorsolateral prefrontal cortex, a brain region consistently active during working memory (Wager and Smith, 2003). A recent fMRI study of heavy cannabis using adults also demonstrated greater dorsolateral prefrontal recruitment relative to controls during SWM 6 to 36 h after last marijuana use, despite similar task performance (Kanayama et al., 2004). More intense and widespread fMRI response despite intact behavioral performance has also been observed among adult alcoholics, suggesting that while some task-related areas demonstrate deficient processing, other ancillary regions may become active to compensate, resulting in an altered functional network among alcoholics (Pfefferbaum et al., 2001). Similarly, the MAUD teens in this study may compensate for subtle neuronal disruption with increased task-related neural recruitment in frontal regions, observed in fMRI as heightened activation. However, MAUD teens did not show the aberrant parietal response we expected given the role of parietal cortex in SWM tasks (Wager and Smith, 2003). While greater SWM task difficulty is associated with increased activity in both frontal and parietal cortices (Diwadkar et al., 2000; Jansma et al., 2000), increased dorsolateral prefrontal activation may be associated with general task difficulty, whereas greater parietal response relates to visuospatial demands (Diwadkar et al., 2000). Therefore, the increase in response among MAUD teens in frontal regions, but not parietal cortex, may suggest a greater difficulty with general task demands, despite similar task performance. However, given a more difficult task, frontal regions may no longer be able to compensate, and activation may decrease in parallel with decreasing task performance.

Although all three groups demonstrated SWM deactivation in the anterior cingulate, MAUD teens showed significantly more deactivation than controls and AUD-only adolescents. The anterior cingulate is highly active at "rest" (i.e., during fixation blocks), during which it is thought to monitor various environmental and internal processes (McKiernan et al., 2003). During a cognitive task, these ongoing processes are suspended as attention is shifted to the task demands, resulting in reduced cingulate activation. Such anterior cingulate deactivation has been observed across a variety of tasks, and increases with greater working memory demands, suggesting reallocation of attentional and working memory resources (McKiernan et al., 2003). Thus, subtle attentional and working memory deficits among MAUD youths may result in the need for greater resource allocation to brain regions subserving SWM, and therefore greater deactivation in anterior cingulate cortex.

MAUD teens evidenced diminished SWM activation compared to controls in the right inferior frontal and right superior temporal/supramarginal gyri, both of which have been implicated in attentional response to salient stimuli (Downar et al., 2002). In particular, the right temporo-parietal junction, including parts of the supramarginal and superior temporal gyri, may be crucial for identifying and shifting attention to relevant stimuli (Downar et al., 2002; Perry and Zeki, 2000). The right inferior frontal gyrus and anterior portions of the insula may be involved in evaluating stimulus relevance and inhibiting irrelevant responses (Downar et al., 2002), and increased activation has been associated with better performance (Casey et al., 2001). The lack of SWM activation in these regions among MAUD teens suggests decrements in attention-orienting systems. The absence of such attentional recruitment may necessitate increased activation in other areas involved with attention and SWM, which could result in reorganization of attention and working memory circuits.

In addition to possible neural inefficiency and reorganization among MAUD teens, aberrant regional cerebral blood flow could produce the observed BOLD response differences. Diminished resting cerebral blood flow has been demonstrated among adult marijuana users during short-term abstinence, particularly in frontal and cerebellar regions (Block et al., 2000; Loeber and Yurgelun-Todd, 1999; Lundqvist et al., 2001). Since BOLD fMRI contrasts a resting condition and an active condition, changes in blood flow during rest alter the magnitude of the observed BOLD response (Cohen et al., 2002). Specifically, reductions in

resting blood flow produce a larger BOLD response change between resting and active conditions (Cohen et al., 2002; Myers et al., 1998). Therefore, the observed greater dorsolateral prefrontal activation among MAUD teens compared to controls could be due to reduced resting frontal blood flow associated with marijuana use. However, the current study contrasted SWM with an active baseline condition (simple attention), possibly limiting the impact of resting blood flow changes on the BOLD response difference between SWM and simple attention. Deactivation abnormalities among MAUD teens could also be related to resting blood flow differences between groups. One study demonstrated increased anterior cingulate blood flow among chronic marijuana users (Block et al., 2000). It has been proposed that regional BOLD response changes will be evidenced as activation if resting activity is low, and deactivation if resting activity is high (LaBar et al., 1999). Thus, high resting blood flow in the anterior cingulate associated with marijuana use could account for enhanced deactivation among MAUD teens in this region. In sum, resting blood flow changes, in addition to neural dysfunction, could underlie the observed fMRI abnormalities among MAUD teens.

MAUD youths showed similar differences relative to AUD teens as they did to controls in most regions, including reduced SWM BOLD response in right inferior frontal and superior temporal cortices, as well as greater SWM deactivation in the anterior cingulate. AUD youths did not demonstrate altered response compared to control teens in these regions. Thus, it appears that teens with marijuana and alcohol use disorders have aberrant patterns of functional response not observed in teens with AUD alone, especially in frontal systems. Heavy marijuana use during adolescence may adversely affect frontal functioning more than other brain regions (Kanayama et al., 2004; Loeber and Yurgelun-Todd, 1999; Lundqvist et al., 2001), and may be related to problems with attention (Solowij et al., 1991, 1995; Tapert et al., 2002) as well as working memory (Schwartz et al., 1989). Further, protracted recent marijuana and alcohol use during adolescence appears associated with disrupted attention and working memory networks above and beyond the abnormalities observed in teens with AUD alone.

While AUD teens demonstrated greater parietal SWM response than controls (Tapert et al., 2004), MAUD youths did not show such a pattern, although MAUD and AUD teens were equivalent on lifetime and recent drinking characteristics. One previous study indicates that adults with MAUD may perform better than those with AUD alone on working memory, visuospatial, and problem solving tasks (Nixon et al., 1998). These findings give rise to a possibility that marijuana use may moderate some parietal abnormalities related to heavy alcohol use (Tapert et al., 2001, 2004) among MAUD youths. However, while alcohol-related parietal changes may not be apparent in MAUD youths, other abnormalities were observed, suggesting that concomitant heavy marijuana and alcohol use during adolescence presents a unique profile of functional alterations.

Previous research has suggested that impaired neuropsychological functioning among marijuana using adults may largely reflect recent use (Pope et al., 2001). We observed a trend for recent marijuana use to be associated with decreased activation in the right middle temporal gyrus, where MAUD teens showed reduced response relative to AUD teens. This could indicate that, at least in this region, abnormal response in MAUD teens may be due to residual drug effects from recent use. We did not find significant relationships between recency of use and brain response patterns in other regions, suggesting that some observed group differences may be unrelated to residual effects. This lack of significant relationships could be due to the limited number of participants reporting distal use, as the majority of participants had used in the month before the scan. However, whether the results in the current study are accounted for by lingering effects of recent use or long-term changes in brain functioning, important clinical implications can be drawn. Seven percent of 11th graders report using marijuana at least 10 days per month (Austin and Skager, 2004), reflecting that sizeable numbers of youths may consistently

experience shorter-term after-effects of marijuana use, involving altered brain functioning during school and other activities.

Several limitations of this study need to be considered. First, our sample size of 49 is relatively small for examining moderator factors, such as gender and family history. Second, although symptom severity was relatively mild, it is possible that group differences in conduct disorder prevalence could have influenced results. Third, by not including teens that heavily use marijuana alone, we were unable to delineate effects solely related to marijuana use. Fourth, our cross-sectional design does not permit the evaluation of functional differences that may have existed before the onset of substance use. Finally, as discussed above, the MAUD adolescents in the current investigation evidenced neural dysfunction after a minimum of 48 h abstinence, yet it is unclear whether the observed differences would persist with more extended sobriety (Pope et al., 2001; Schwartz et al., 1989; Yurgelun-Todd et al., 1999). Future studies might attempt to disentangle the residual and longer term effects of adolescent marijuana use by requiring longer periods of monitored abstinence before assessment.

Despite these limitations, the current investigation raises several questions to be addressed by future studies. First, we observed increased frontal activation during SWM among MAUD youths in the context of intact performance. It is unclear how brain response patterns would differ if performance was impaired. Future studies might attempt to parametrically alter task difficulty in order to characterize neural patterns associated with changing behavioral performance (Callicott et al., 1999; Jansma et al., 2000). Although both SWM and attention deficits may have influenced brain response abnormalities among MAUD teens, the task utilized in this study was not designed to directly assess attention. Thus, future neuroimaging studies of heavy marijuana using youths might attempt to examine sustained and divided attention more closely, as well as other cognitive abilities. Further, as resting blood flow abnormalities may have contributed to some fMRI differences between groups, future fMRI studies should assess resting perfusion for use in covariate analyses. In addition, the current fMRI study cannot make any direct conclusions about the neural characteristics underlying functional change. Magnetic resonance spectroscopy could elucidate the metabolic and cellular underpinnings of functional abnormalities related to combined marijuana and alcohol use. Finally, as the persisting neural effects of heavy marijuana use are unclear, longitudinal investigations should characterize the neuromaturational and functional consequences of heavy alcohol and marijuana use during youth, as well as the potential for neural recovery.

In summary, this study found aberrant brain response to a spatial working memory task among adolescents with comorbid marijuana and alcohol use disorders. Compared to non-abusing controls and teens with AUD alone, MAUD youths evidenced frontal and temporal dysfunction, suggesting that heavy marijuana use could be related to attentional decrements and compensatory responses in areas subserving spatial working memory. These neural abnormalities were not observed among AUD-only teens, despite similar drinking characteristics, indicating that combined marijuana and alcohol use may have a unique influence on brain functioning. Together, these findings demonstrate that teens with comorbid marijuana and alcohol use disorders show subtle disruptions in brain functioning after a minimum 48 h of abstinence.

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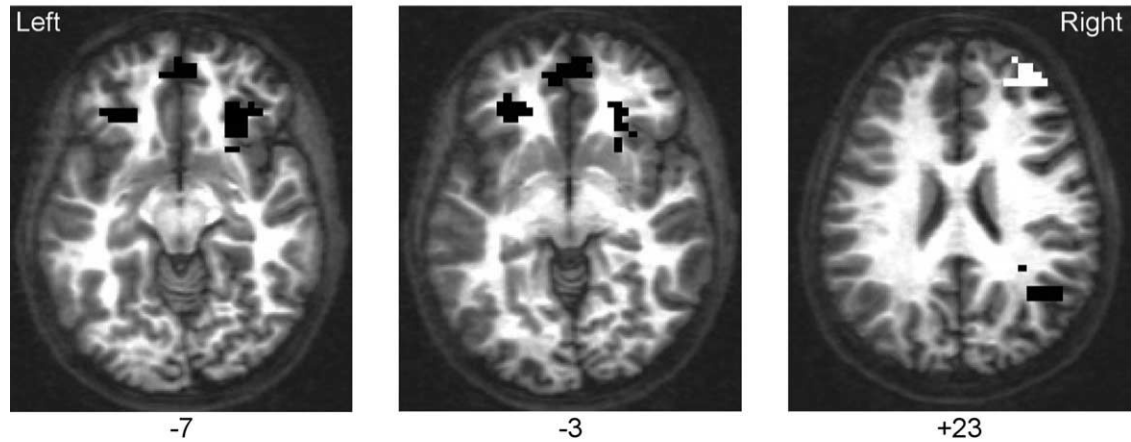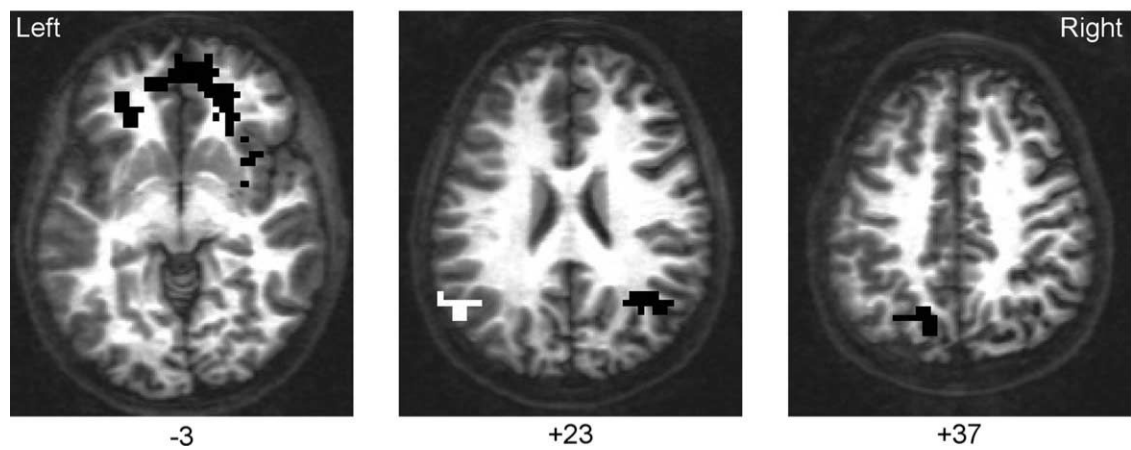
Fig. 1.
Significant clusters of group difference in spatial working memory fMRI response in MAUD teens compared to controls (top row) and AUD teens (bottom row). Black clusters indicate regions where MAUD teens showed less spatial working memory response than others, and white clusters represent areas where MAUD teens showed more spatial working memory response than others; cluster $p < .0167$, volume >1072 μl. Numbers below brain images refer to axial slice positions.

Table 1

Demographic characteristics of adolescent participants

	MAUD (*n* = 15), *M* (S.D.) or %	AUD (*n* = 15), *M* (S.D.) or %	Controls (*n* = 19), *M* (S.D.) or %	*p*
Age (range 15−17)	16.91 (0.64)	16.77 (0.66)	16.50 (0.83)	ns
Female (%)	33.3	33.3	42.1	ns
Caucasian (%)	93.3	100.0	73.7	.07
Family history negative (%)[a]	40.0	41.7	37.5	ns
Conduct disorder positive (%)	26.7	33.3	5.3	.01[c]
CBCL externalizing *T*-score	47.74 (9.76)	42.76 (4.88)	43.74 (6.85)	ns
Grades completed	9.16 (2.55)	10.07 (0.80)	9.47 (2.20)	ns
Parent annual salary (thousands)	66.92 (43.64)	103.67 (60.72)	68.11 (28.38)	.05[d]
Vocabulary scaled score[b]	11.77 (2.13)	12.53 (1.77)	12.21 (2.80)	ns

AUD: alcohol use disordered; MAUD: marijuana and alcohol use disordered; CBCL: Child Behavior Checklist.

[a] No first- or second-degree biological relative with alcohol or drug abuse or dependence.

[b] Based on Wechsler Intelligence Scale for Children-III (Wechsler, 1993) for participants ≤16 years old, and Wechsler Adult Intelligence Scale-III (Wechsler, 1997) for 17 years old.

[c] MAUD and AUD significantly different than controls.

[d] Tukey's pairwise comparisons non-significant.

Table 2

Substance use characteristics of adolescent participants

	MAUD (*n* = 15), *M* (S.D.)	AUD (*n* = 15), *M* (S.D.)	Controls (*n* = 19), *M* (S.D.)	*p* (AUD vs. MAUD)
Years since first marijuana use	3.37 (2.24)	3.03 (1.11)	1.46 (0.66)	ns
Lifetime marijuana use episodes	309.87 (255.40)	11.33 (12.93)	1.47 (4.65)	.000
Marijuana use/month, past 3 months	12.80 (10.09)	0.60 (1.30)	0.00 (0.00)	.000
Marijuana abuse/dependence criteria, past 3 months	4.33 (2.87)	0.67 (1.29)	0.00 (0.00)	.000
Days since last marijuana use[a]	7.64 (11.36)	79.67 (71.37)	145.0 (91.92)	.020
Percentage who used marijuana in past week	85.7	0	0	.000
Years since first drink[a]	3.57 (1.49)	3.73 (1.68)	2.79 (1.65)	ns
Years of regular drinking[a,b]	2.05 (0.66)	1.93 (1.00)	0.00 (0.00)	ns
Lifetime alcohol use episodes	135.47 (218.29)	128.93 (142.89)	5.11 (10.88)	ns
Drinks/month, past 3 months	42.27 (47.34)	41.47 (31.26)	0.72 (2.82)	ns
Alcohol withdrawal symptoms, past 3 months	2.00 (2.04)	2.27 (2.05)	0.05 (0.23)	ns
Alcohol abuse/dependence criteria, past 3 months	2.67 (2.64)	2.47 (1.81)	0.11 (0.32)	ns
Typical peak BAC, past 3 months[c]	.110 (.07)	.108 (.09)	.01 (.02)	ns
Days since last drink[a]	13.53 (13.45)	16.73 (14.91)	67.90 (60.46)	ns
Percentage who drank in past week (%)	46.7	33.3	5.3	ns
Smoked cigarettes, past month (%)	46.7	46.7	5.3	ns
Cigarettes per smoking day[a]	2.67 (2.73)	2.50 (2.12)	3.00 (0.00)	ns
Lifetime other drug use episodes				
Opiates	1.40 (3.87)	0.20 (0.56)	0.00 (0.00)	ns
Inhalants	1.13 (2.26)	0.07 (0.26)	0.00 (0.00)	.062
Hallucinogens	1.00 (2.24)	0.00 (0.00)	0.00 (0.00)	.078
Ketamine	0.36 (1.21)	0.00 (0.00)	0.00 (0.00)	ns
Amphetamines	0.33 (0.72)	0.20 (0.77)	0.00 (0.00)	ns
Benzodiazepines	0.18 (0.40)	0.17 (0.41)	0.00 (0.00)	ns
Cocaine	0.13 (0.52)	0.13 (0.52)	0.00 (0.00)	ns
MDMA	0.00 (0.00)	0.08 (0.29)	0.00 (0.00)	.061
Barbiturates	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	ns
PCP	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	ns

AUD: alcohol use disordered; MAUD: marijuana and alcohol use disordered; BAC: blood alcohol concentration.

[a] Figures include only those who reported use.

[b] Weekly use based on self-report; only 10 AUD teens and 10 MAUD teens reported such use.

[c] Calculated based on self-report (Fitzgerald, 1995).

Table 3

Regions showing significant BOLD response difference between controls and MAUD teens

Anatomic region	Brodmann's area	Volume (µl)	Talairach coordinates			Effect size, Cohen's *d*
			x	*y*	*z*	
SWM activation						
Controls > MAUD						
Right inferior frontal gyrus	47	1972	26R	34A	8I	0.63
Right superior temporal and supramarginal gyri	39	1544	40R	57P	28S	1.07
MAUD > controls						
Right superior frontal and middle frontal gyri	9, 10	1072	30R	48A	24S	1.02
SWM deactivation						
MAUD > controls						
Left inferior frontal gyrus	47	1672	37L	27A	11I	0.81
Bilateral medial frontal cortex and anterior cingulate	10, 32	1415	5L	48A	4I	0.86

MAUD: marijuana and alcohol use disordered; SWM: spatial working memory. Talairach coordinates and Cohen's *d* refer to maximum signal intensity group difference within the cluster; R, right; L, left; A, anterior; P, posterior; S, superior; I, inferior.

Table 4

Regions showing significant BOLD response difference between AUD and MAUD teens

Anatomic region	Brodmann's area	Volume (μl)	Talairach coordinates			Effect size, Cohen's *d*
			x	*y*	*z*	
SWM activation						
AUD > MAUD						
Right inferior frontal gyrus, claustrum, and	47	1286	30R	20A	11I	0.73
insula						
Left precuneus	7	2187	16L	64P	49S	1.23
Right middle temporal and supramarginal gyri	39	1158	44R	57P	24S	1.08
SWM deactivation						
AUD > MAUD						
Left superior temporal gyrus	22, 39	2315	54L	57P	21S	1.01
MAUD > AUD						
Bilateral middle and superior frontal gyri,	10, 11, 32, 47	6346	26L	38A	4I	0.67
anterior cingulate						

AUD: alcohol use disordered; MAUD: marijuana and alcohol use disordered; SWM: spatial working memory. Talairach coordinates and Cohen's *d* refer to maximum signal intensity group difference within the cluster; R, right; L, left; A, anterior; P, posterior; S, superior; I, inferior.