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Tentative Evidence for Striatal Hyperactivity in Adolescent Cannabis Using Boys: A Cross-Sectional Multicenter fMRI Study

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Abstract

Adolescents' risk-taking behavior has been linked to a maturational imbalance between reward (“go”) and inhibitory-control (“stop”) related brain circuitry. This may drive adolescent drug-taking, such as cannabis use. In this study we assessed the non-acute effects of adolescent cannabis use on reward-related brain function. We performed a two-site (United States and Netherlands; pooled data) functional magnetic resonance imaging (fMRI) study with a cross-sectional design. Twenty-one abstinent but frequent cannabis-using boys were compared with 24 non-using peers on reward-related brain function, using a monetary incentive delay task with fMRI. Focus was on anticipatory and response stages of reward and brain areas critically involved in reward processing like the striatum. Performance in users was normal. Region-of-interest analysis indicated striatal hyperactivity during anticipatory stages of reward in users. Intriguingly, this effect was most pronounced during non-rewarding events. Striatal hyperactivity in adolescent cannabis users may signify an overly sensitive motivational brain circuitry. Frequent cannabis use during adolescence may induce diminished ability to disengage the motivational circuit when no reward can be obtained. This could strengthen the search for reinforcements like drugs of abuse, even when facing the negative (non-rewarding) consequences.

Keywords

cannabis; adolescence; fMRI; reward; striatum

Introduction

During adolescence, the propensity for risk taking and impulsive behavior is higher than at other stages of life. Developmental neuroscience findings convincingly demonstrate that this has its origin in immaturities of the brain systems involved in reward processing and inhibitory control (Geier & Luna 2009). The triadic model (Casey, Jones & Hare 2008; Ernst, Pine & Hardin 2006) posits that during adolescence reward and novelty seeking, in the face of uncertain or potential negative outcome, can be explained by an earlier functional

maturation of reward-related circuitry (ventral striatum and orbitofrontal cortex) compared to harm-avoidant (amygdala) and inhibitory control-related brain circuitry (medial/ventral prefrontal cortex). The result is an imbalance in “go” versus “stop” neurocircuitry, and one can easily conceive how this may drive adolescent drug taking or other risky behavior, despite rational knowledge and awareness of the potential negative consequences. Exposure to drugs like nicotine, alcohol and cannabis may, in turn, further interfere with neurodevelopment.

In this respect, the effect of cannabis on developing brain functions during adolescence deserves greater attention. Its psychoactive compound, delta-9 tetrahydrocannabinol (THC) acts on the endogenous cannabinoid system. This system reaches functional maturity during adolescence and plays an active role in activating or suppressing other developing neurotransmitter systems, in particular mesocortical dopaminergic and opioid systems (Fernandez-Ruiz, Berrendero & Hernandez 2000). Perturbations by exogenous cannabinoids can lead to functional deficits, as demonstrated by developmental studies in rodents. Here, chronic periadolescent treatment with exogenous cannabinoids resulted in persisting deficits in locomotor activity, motivation, attention and short-term memory (Jager & Ramsey 2008). The few studies in human adolescents suggest cannabis-induced neurophysiological alterations in the domains of executive and inhibitory processing, but more research is needed to warrant conclusive statements (Jager & Ramsey 2008).

In the present study we investigated the impact of frequent adolescent cannabis use on non-drug reward processing and related brain function. Neuroimaging studies in humans reveal that reward receipt enhances activity of subcortical structures (i.e. striatum and amygdala), and of the mesial frontal cortex (mFC). Reward omission tends to reduce activity in these same structures (Ernst, Pine & Hardin 2006; Bjork et al. 2004). We performed blood-oxygen-level-dependent (BOLD) fMRI using a modified monetary incentive delay (MID) task (Van Hell et al. 2010; Bjork et al. 2004). This task allows for separation of brain activity during anticipation of rewards (motivational component; robustly activates the ventral striatum) from activity following the response to a target upon which the reward depends (consummatory component; activates the mFC) (Bjork et al. 2004; Knutson et al. 2001). As there is evidence for gender differences in the rate and timing of neurodevelopment (Giedd et al. 2006) and neurodevelopmental responses to cannabinoid exposure (Rodriguez de Fonseca et al. 1993), only boys were included in this study. As our primary interest was in the long-lasting effects compared to the acute intoxicating effects, we examined frequent cannabis using boys that were abstinent at the time of the study, and compared them with age-matched non-using peers. Given that adolescents may exhibit an imbalance in “go” versus “stop” neurocircuitry with a hypersensitive ventral striatum, we hypothesized that cannabis use would further amplify imbalance. That is, in comparison to non-using controls, we expected cannabis users to display relative striatal overactivation during reward anticipation, with relative hypoactivity in prefrontal inhibition control neurocircuitry. We further expected that earlier onset of cannabis use would lead to a stronger deviation in reward-related brain activity from non-using peers.

Method

This study was a joint venture of the University Medical Center Utrecht (The Netherlands) and the University of Iowa (United States). Data were pooled to increase statistical power. The study involved a single fMRI session where participants performed three tasks on working memory, associative memory and reward processing. The memory studies have been reported in a separate paper (Jager et al. 2010).

Participants

A total of 47 boys participated in the study (13 – 19 years) including 23 regular cannabis users (11 Dutch, 12 US) and 24 controls (12 Dutch, 12 US). Eligibility was assessed through a screening procedure involving questionnaires on drug use, medical history and mental health. Table 1 reports inclusion and exclusion criteria. For details we refer to a previous report (Jager et al. 2010). The local Institutional Review Boards approved the study. Written informed consent was obtained from adolescents and their parent or legal guardian in conformance with Institutional Review Board requirements and the Helsinki Declaration of 2008. Subjects were reimbursed for participation.

Procedure

The study procedure has been described in detail in a previous report (Jager et al. 2010). In short, subjects participated in two sessions separated by one week. The first involved screening for inclusion and exclusion criteria, using drug questionnaires, a semi-structured psychiatric interview (NIMH Diagnostic Interview Schedule for Children; C-DISC-version IV) (Shaffer et al. 2000), and IQ measurements – four subtests (similarities, block design, vocabulary and matrix reasoning) of the Wechsler Intelligence Scale for Children – 4th edition (WISC-IV) (Baron 2005). The second session included neuropsychological testing and fMRI scanning. Participants abstained from cannabis, alcohol and other substances for at least 24 hours prior to the first session and remained abstinent until the second session was finished. Smoking was allowed until two hours before the scanning session (to avoid nicotine withdrawal). In both sessions, clearance of drugs was tested by means of a urine sample. Exclusion followed on positive testing on any psychoactive substance on the second test day, except for cannabis that can linger in the body for several weeks due to its lipophilic properties, and thus, can induce a positive test result even after one week of abstinence (Goodwin et al. 2008). Instead, cannabis-using subjects were excluded only when their urine toxicology test failed to show a decrease in quantified levels of cannabinoid metabolites (THCCOOH) between the first and second urine samples. Based on the lab results, two subjects had to be excluded from analyses. Two other subjects had a positive urine test on cannabinoids during the second session, but THCOOH-levels were decreased compared to the first measurement, consistent with self-reported abstinence. All other subjects had negative urine tests. Thus 45 subjects were included in the study.

Task

The monetary incentive delay (MID) task (Van Hell et al. 2010; Bjork et al. 2004; Knutson et al. 2001) consisted of 72 trials (duration 3 – 10 sec). At the beginning of each trial a cue was presented signalling a potentially rewarding or a neutral trial (Figure 1). Following this cue, a target was presented to which subjects had to respond as fast as possible by pressing a button. Finally, feedback on trial performance was given (1 euro/US dollar or no reward), in tandem with cumulative task earnings as the task proceeded. Prior to the experiment, ten practice trials were presented to familiarize subjects with the task. From these data, the shortest reaction time to the target determined the individually adapted time limit allowed for responses during the task. Hence, in case of a reward cue subjects could win money when responding within the time limit. The task was designed to deliver reward in fifty percent of trials, to ensure that all subjects received the same number of rewards. This was achieved by increasing the time limit with 400 ms in half of the trials to ensure subjects would be fast enough to win the trial, and decreasing the time limit with 400 ms in the other half of the trials to make sure subjects would miss the trial. Anticipation time (the time between cue and target) and inter-trial interval were varied (3 – 10 s; mean 6 s, and 0 – 30 s; mean 4.2 s, respectively). All subjects earned a comparable amount of money (mean 17 euro/US dollar ($SD \pm 1.5$)). The Institutional Review Board required that all participants were paid the same amount of money upon concluding the experiment, but prior to scanning

participants were informed they would receive the amount of money earned during the task. Hence, participants believed their winnings to be dependent upon task performance.

FMRI acquisition

Imaging data were collected using two clinical 3.0 Tesla MRI scanners, both with an 8-channel head coil but from different vendors (Philips Achieva and Siemens Magnetom Trio). The approach adopted to assess scanner compatibility and to overcome problems resulting from scanner differences is described in a Supplement to a previous report (Jager et al. 2010).

Scan parameters used were: a T2* sensitive echoplanar sequence, TE/TR 35/2000 ms, flip angle 70°, FOV 256 × 256 mm, acquisition matrix 64 × 64, slice thickness 3.6 (plus a 0.4 mm gap), voxel size 4.0 mm isotropic, 26 slices, scan orientation transaxial. For MID a single run of 504 scans was acquired over a period of 16.8 minutes. In addition, a volumetric T1-weighted MR anatomical scan was acquired for spatial localization (TR 25 ms, flip angle 30°, FOV read 256 mm, voxel size 1.0 mm isotropic, 176 slices, scan duration 7.8 minutes).

Statistical analysis

Sample characteristics and drug use—Cannabis parameters were: age of onset, cumulative use lifetime (estimated number of joints), current or recent use (number of joints last year), frequency and duration of use, and abstinence (weeks since last use). Additional drug use data included last year use of alcohol (average number of drinks/week), tobacco (average number of cigarettes/week) and lifetime use of other illegal substances (episodes lifetime). Other variables included age, estimated IQ, country (United States versus Netherlands) and a diagnosis of conduct disorder (yes/no). Group differences were tested using t-tests and non-parametric Kolmogorov-Smirnov Z tests.

Task performance—Outcome measures included reaction times (RT) on target responses. General linear model (GLM) repeated measures analysis was applied, with task condition (reward and neutral) as a within-subject factor, and group (user versus control) as a between-subject factor. Gain (amount of money won) was not included in the analysis, as this variable was manipulated and showed minimal variance between subjects.

FMRI—Imaging data were collected with two scanners in different locations and from different vendors. Data were pooled to increase statistical power. The approach adopted to overcome potential problems resulting from scanner differences included quantitative comparison of temporal signal-to-noise-ratio (tSNR) maps, a smoothing step of 8 mm FWHM during pre-processing, and inclusion of country (Netherlands/United States) as a covariate in fMRI data analysis. The rationale of this approach is described in a Supplement to a previous report (Jager et al. 2010).

Imaging data were pre-processed using SPM5 (<http://www.fil.ion.ucl.ac.uk/spm>). Pre-processing consisted of realignment and unwarping of functional images and co-registration with the anatomical volume. After realignment, functional scans were normalized using parameters obtained from spatial normalization into standard MNI space of the anatomical volume. After normalization, functional scans were spatially smoothed with an 8 mm (FWHM) Gaussian kernel.

First level analysis involved modelling individual time-series with canonical hemodynamic response functions using a factor matrix containing factors time-locked to anticipatory stages (reward or neutral), and three types of response stages depending on neutral, positive (reward trials) or negative feedback (missed reward trials). Due to a fixed time interval (1

sec) between target presentation and feedback, separate factors coding for the motor responses to targets and feedback would have resulted in high collinearity between these factors. Therefore these responses were modelled together with the feedback stimulus as a single event, yielding three factors for positive, negative and neutral feedback. Reference to these factors thus includes the motor responses. To correct for drifts in the signal, a high-pass filter with cut-off frequency of 0.0025 Hz was applied to the data. Individual activation maps were generated for all five conditions (anticipation of neutral and reward trials, feedback on either neutral, rewarded or non-rewarded trials). Subsequent group analysis centered on two linear contrasts of signal change (hereafter 'contrasts') calculated for the MID task: 1) reward versus neutral anticipation, and 2) positive feedback vs. negative feedback (in reward trials). Contrast maps were then used in a second-level brain analysis to test for effects of cannabis use on brain activity. Age and a dichotomized variable for country (0=NL; 1=US) were added as covariates, to take into account potential systematic effects of age and/or differences in MRI-scanners across sites.

Region of interest analysis—As the dorsal (caudate nucleus and putamen) and ventral striatum (including the nucleus accumbens) constituted primary regions-of-interest (ROI), we performed an ROI analysis on BOLD signal in these areas, using the Marsbar toolbox (Brett et al. 2002) and the Automated Anatomical Labelling (AAL) ROI library in SPM5. The AAL ROI library contains ROIs in Marsbar format, anatomically defined by hand on a single brain matched to the MNI template (Tzourio-Mazoyer et al. 2002). We selected the AAL ROIs on the left and right caudate nucleus (CN) and left and right putamen (see Figure 2). No separate ROIs were defined for the nucleus accumbens, which is located where the head of the caudate and the anterior portion of the putamen meet. Hence, a separate mask for the nucleus accumbens would show significant overlap with the caudate (head) mask.

ROIs were marked and activity values for all subjects were obtained per ROI by averaging beta-values across all contained voxels for the anticipatory (neutral and reward) and feedback (positive and negative) task conditions. This resulted in sixteen values per subject, i.e. mean activity values of four ROIs (left and right caudate and putamen) in four conditions (anticipation neutral and reward, positive and negative feedback). These variables were entered into two GLM repeated measures analyses (separate GLM's for anticipation and response-related variables) in SPSS 15.0.1, using country as a covariate.

Results

Table 2 summarizes sample characteristics and drug use for users (United States and Dutch) and non-users (United States and Dutch). In the user group, estimated number of joints smoked lifetime was generally high, but showed considerable variation. Number of joints smoked lifetime was on average 4,006 (SD 7,555), with a median of 1,400 and a range between 224 – 32,850 joints. For use in the year preceding inclusion, the estimated number of joints was on average 741 (SD 772), with a median of 485 and a range between 208 – 3,528. Hence, these boys smoked on average 2 joints per day, but some extreme cases reported closer to smoking 8–10 joints per day. All users, however, were abstinent on the day of scanning. On average, abstinence was 5.1 weeks (SD $\pm$ 4.2, range 1 – 16 weeks). In the non-user group, 17 boys reported no previous cannabis use. Four controls reported cannabis use on less than 5 occasions, whereas three boys had used between 5 – 15 times lifetime.

Groups did not differ in age, but users had significantly lower IQ-scores than controls ($p < 0.01$), and reported significantly higher last year consumption of alcohol (mean alcoholic drinks per week 13.3 ($\pm$ 13.6) for users compared to 3.4 ($\pm$ 5.8) for controls) and tobacco (mean number of cigarettes smoked per week 63.8 ($\pm$ 53.4) for users compared to 6.1 ($\pm$ 14.9) for controls). Additionally, demographic and cannabis use parameters were compared

for the user and non-user group separately, comparing United States subjects with Dutch subjects, to identify potentially site related biases. Dutch users were older than United States users (mean age $17.9 (\pm 0.9)$ and $16.7 (\pm 0.8)$ respectively, $p < 0.05$), but no significant differences were found on IQ scores or cannabis use parameters. Nine United States users had a C-DISC diagnosis of conduct disorder, whereas none of the Dutch users or the controls (both United States and Dutch) did.

Task performance

Figure 3 displays reaction times for users and non-users across task conditions. Repeated measures GLM analysis with task condition (reward vs. neutral) as within- and group (users vs. non-users) as between-subject factors yielded a main effect of condition ($F(1,43)=29.6$, $p < 0.001$) indicating that both groups responded faster to the target during reward trials compared to neutral trials. The group by condition interaction term revealed a trend ($F(1,43)=3.04$, $p=0.09$, indicating that users tended to show larger differences in reaction times between reward and neutral trials (mean of 25 ms for users, and 12 ms for non-users). Both groups earned an equal amount of money (17 euro/US dollars).

fMRI data

To check whether users and non-users activated similar networks of brain regions during reward processing (as expected, as there is no a priori reason to assume that cannabis users show significant functional reorganization of the brain), we obtained group activation maps of the contrast reward versus neutral anticipation for users and non-users separately ($p < 0.05$, FWE corrected). Visual inspection of the group activation maps indicated no differential activation patterns between users and controls. Subsequent group activation maps were derived of all subjects combined ($p < 0.05$, FWE corrected). Group-wise analysis centered on two contrasts: 1) reward versus neutral anticipation, and 2) positive feedback vs. negative feedback. See Fig. 4 for a graphical overview of group activation maps. The contrast reward versus neutral anticipation yielded large areas of significant activation including striatal areas (caudate nucleus, putamen), thalamus, and areas in the inferior prefrontal gyrus, insula, medial prefrontal cortex, anterior cingulate cortex and orbitofrontal cortex. In addition, we found activation in the posterior cingulate cortex, the occipital cortex and cerebellum.

The feedback contrast (positive minus negative) predominantly showed clusters in nucleus caudate, putamen and thalamus. Smaller clusters were found in posterior cingulate cortex, and the occipital lobe (lingual gyrus, cuneus).

Region of interest analysis—A Region of Interest (ROI) analysis was performed of reward-related brain activity in the areas critically involved in (anticipation of) reward, that is, the left and right caudate nucleus (CN) and left and right putamen, using anatomically defined masks (see Methods section for details). For anticipation-related activity, GLM repeated measures analysis with group as between-subjects factor, ROI (left and right CN, left and right putamen) and task condition (neutral and reward anticipation) as within-subjects factors, yielded a marginally significant main effect of group ($F(1,42)=3.22$, $p=0.08$), indicating that users tended to show increased levels of activity during anticipatory stages of the task. Posthoc ANOVA (see Figure 5) revealed that during anticipation of neutral trials, users showed significantly higher levels of activity than controls in the left CN ($F(1,43)=5.36$, $p=0.03$), whereas levels of activity were marginally increased compared to controls in the right CN ($F(1,43)=2.82$, $p=0.10$), and right putamen ($F(1,43)=3.74$, $p=0.06$). Activity levels during anticipation of rewarding trials also seemed to be elevated in users (see Fig. 5), but none of the ANOVAs reached significance. Hence, the two groups showed comparable activation levels during anticipation of reward. For feedback no significant main or interaction effects of group were observed.

Hence, ROI analysis showed that compared to controls, cannabis users displayed marginally higher levels of brain activity in striatal regions during the anticipatory stages of the MID task. However, this was predominantly due to increased levels during anticipation of neutral trials instead of reward trials.

Whole brain analyses—A whole brain voxelwise comparison in SPM5 (two-sample t-test with country as covariate, $p < 0.05$, FWE corrected) failed to show any significant group differences in anticipation or feedback.

Correlation analysis—The relation between increased activity levels during anticipation of neutral trials in users and cannabis use parameters (age of onset, lifetime use, last year use) was explored using non-parametric correlation analysis. Results showed that brain activity in the right CN during anticipation of neutral trials was inversely related to age of onset of cannabis use ($\rho = -0.45$, $p = 0.04$; see Figure 6), indicating that earlier onset corresponded with higher activity levels. Correlations of activity in the left CN, and right and left putamen with age of onset had the same direction but did not reach significance. No other correlations with parameters of cannabis use were found.

Discussion

This study assessed the non-acute effects of frequent adolescent cannabis use on reward processing. This was done by examining brain activity in the striatum during monetary reward anticipation and reward feedback.

The key finding of the study was that frequent cannabis using boys displayed striatal hyperactivity during anticipatory components of reward processing, whereas no such effect was found on processing of reward outcome. There was tentative evidence that in the caudate altered activity patterns in users were inversely related to age of onset of cannabis use. The earlier the onset of use, the more prominent the effects were on caudate activity. Significance of effects on anticipatory activity disappeared, however, in whole brain group comparisons. Neither did whole brain group comparisons yield significant effects of cannabis use on brain activation patterns in other relevant brain regions like orbitofrontal, lateral and medial prefrontal and anterior cingulate cortex.

At the behavioral level both users and non-users reacted faster during trials when cues signalled a potential reward than during neutral trials. Since reaction time is strongly affected by motivation, this implies that users were at least as motivated as non-users.

Even if the finding of striatal hyperactivity in users during reward anticipation does not stand up to rigorous statistical thresholding (whole brain analysis), it is compellingly consistent with the triadic model of risky behavior in adolescence (Casey, Jones & Hare 2008; Ernst, Pine & Hardin 2006). Striatal hyperactivity in adolescent cannabis users may signify an overly sensitive motivational brain circuitry. Striatal hyperactivity was, unexpectedly, most prominent during anticipation of neutral/non-rewarding trials instead of during anticipation of reward. Gatzke-Kopp and colleagues (Gatzke-Kopp et al. 2009) recently compared neurobiological correlates of reward responding in adolescents with and without externalizing behavior disorders (Attention-Deficit Hyperactivity Disorder (ADHD) and conduct disorder (CD)). They found that externalizing adolescents exhibited striatal activation during both reward and non-reward task trials. Controls exhibited striatal activation during reward trials, which decreased during non-reward trials and shifted from the striatum to the anterior cingulate cortex. Hence, externalizing psychopathology could be characterized by deficits in processing the omission of predicted reward, reflected by the sustained striatal activation during both reward and non-reward trials. Insensitivity to the

omission of predicted reward may promote perseverative responding to rewards, even after reward contingencies become unfavourable (Gatzke-Kopp et al. 2009).

In the present sample nine (all United States subjects) out of twenty-one cannabis using boys met criteria for a diagnosis of CD according to their scores on the DISC-IV interview. Still, none of them had received a formal diagnosis from a medical professional or was prescribed psychostimulant medication. We cannot exclude the possibility that the occurrence of externalizing psychopathology in nine users has influenced our results. Yet, it seems implausible for two reasons. For one, the remaining twelve cannabis users in the sample did not display externalizing behavior disorder symptoms. Secondly, presence of conduct disorder was country-specific. As effects of country and other site-related differences on brain activity data were regressed out in all analyses, it seems unlikely that conduct disorder played a major role in the observed group differences in striatal activation.

Two recent studies on the effects of chronic cannabis use on reward brain function in adult users reported contrasting findings. Nestor and colleagues (Nestor, Hester & Garavan 2010) observed increased activity in the right ventral striatum (putamen) during reward anticipation in frequent cannabis users with variable abstinence periods ranging from 12 hrs to three weeks. Van Hell et al. (Van Hell et al. 2010) observed an opposite effect, and reported attenuated brain activity during reward anticipation in the nucleus accumbens in one-week abstinent users compared to non-smoking controls, and decreased reward anticipation activity in the caudate nucleus compared to both smoking and non-smoking controls. This study also included a smoking control group, to disentangle the effects of nicotine from those of cannabis. It was proposed that nicotine could be responsible for attenuated activity in the accumbens, but that the blunted response in the caudate was associated with use of cannabis. The relevance of taking nicotine into account is also demonstrated by a recent study on striatal activation during reward anticipation in adolescent smokers. Here, it was found that adolescent smokers had lower neural responses in the ventral striatum during reward anticipation than non-smokers, and this response was correlated with smoking frequency (Peters et al. 2011). Whether these effects were fully due to nicotine use is, unfortunately, hard to say as the smokers also reported cannabis use, whereas the controls did not.

The discrepancies between our present findings in adolescent cannabis users and the findings reported in the studies mentioned above point out several factors of importance. For one, effects of chronic cannabis use on reward-related brain function in adult users may be of an altogether different nature in adolescents. Second, duration of abstinence likely plays a role, as differential findings could be related to the longer abstinence periods in the present study (on average 5 weeks), as compared to the average abstinence period of one week (Van Hell et al. 2010) and four days (Nestor, Hester & Garavan 2010). Finally, cannabis is commonly mixed with tobacco. Interactions between cannabis and nicotine use can lead to synergistic effects that are difficult to disentangle.

While acknowledging that the present findings are not very robust, one can speculate about a possible underlying mechanism that may explain striatal hyperactivity in cannabis users during *non-reward* trials. To select appropriate behaviors leading to rewards, the brain needs to learn associations between stimuli, actions, and rewards. Neuroimaging studies have revealed that the dorsal striatum plays an important role in learning such stimulus-action-reward associations, with a functional difference between the putamen and the caudate nucleus. The putamen is thought to be particularly involved in associating a reward with an appropriate action in response to a stimulus, whereas the caudate nucleus is implicated in reward-prediction error (Haruno & Kawato 2006). Reward prediction error occurs when an expected reward is not received (omitted). In the MID task, a cue is presented that signals (a

chance of) reward, but subjects cannot predict winning or not winning. Still, a missed target (as it is preceded by a reward cue) can be perceived as a reward prediction error. A neutral cue signals altered contingencies between stimulus (the cue), action and reward, as the monetary incentive is removed. Hence, a possible explanation for the current findings is that users do not show abnormalities in reward processing, as both groups showed comparable activations in striatal regions during rewarded trials, but in adjusting to reward omission in neutral trials and failing to disengage striatal regions when cues do not signal monetary incentives. This is in line with studies that reported increased response perseveration and a less adaptive response allocation to the changing reinforcement contingencies on a concurrent-reinforcement task both after acute delta-9 THC administration (Lane & Cherek 2002) and in heavy cannabis-smoking adolescents (Lane et al. 2007).

The present study has several limitations. For one, despite our efforts to match groups on key variables like age, IQ, and use of other substances, they differed on a number of them, with users displaying lower estimated IQ scores (albeit within the normal range), and greater alcohol and tobacco smoking histories. Unfortunately, these factors systematically correlated with parameters of cannabis use (age of onset, cumulative use, recent use) and could, therefore, not be disentangled. Nonetheless, we cannot exclude the possibility that group differences in IQ, use of alcohol and tobacco contributed to the effects of cannabis use on striatal functioning.

A relevant issue is also whether to allow smokers to smoke tobacco before scanning or not. This is a matter of choosing between acute nicotine effects (which we have shown previously to occur in adults, Van Hell et al. 2010) and withdrawal. Here we chose to also allow smoking to avoid withdrawal, to make some comparison across studies possible. Note that the difference between groups in the present study cannot be explained by nicotine since cannabis users exhibited an elevated anticipation response in nucleus accumbens as opposed to a blunted response associated with tobacco use in the 2010 study.

In addition, as mentioned previously, there was an unintended but high incidence of symptoms of conduct disorder in the United States users group. This may have been the consequence of differences in recruitment strategies. Recruitment in the Netherlands included advertisement on the internet and asking help of schools. In the United States, however, this strategy was not feasible and recruitment was switched to local substance abuse and adolescent health and resource centers offering education and treatment programs to minors that got involved with the legal system because of possession of cannabis or other drugs. Boys ending up in these programs may display more externalizing behavior problems and more often meet criteria for conduct disorder.

The multi-center design poses, besides the advantage of increased power, a challenge in terms of scanner compatibility. We believe we adopted an adequate approach to minimize and/or quantify systematic effects due to site related differences in scanner equipment (Jager et al. 2010; Friedman et al. 2006) that may have relevance for future multi-site studies. In the present study, pooling data from two centers had the drawback of scanner related differences in between-subject variability, predominantly in the orbito- and ventromedial regions and extending into one of the regions of interest, the head of the caudate containing the nucleus accumbens. This has likely limited sensitivity to detect cannabis-related effects in these particular areas. For this reason, we have refrained from drawing any conclusions on these regions based on the present findings. Nonetheless, sensitivity for cannabis-related effects on brain activation in body and tail of the caudate and the putamen do not seem to be compromised by the multi-center approach, and thus, benefits from the increased power.

Another issue is that we did not match users and controls on genotype. Recent studies on reward (dys)function indicate a role for genotypes putatively associated with low dopamine signaling capacity, for example the TaqIA allele. Persons carrying an AI allele show lower D2 receptor density and less responsivity of reward regions that primarily rely on dopamine signaling, denoted reward deficiency syndrome (Blum et al. 2012; Stice et al. 2012; Le Foll et al. 2009, Kirsch et al. 2006). This genotype is linked to a higher risk for substance abuse. In adolescents, due to control from parents or other influential adults, an endophenotype of reward deficiency could be expressed in other behaviors than substance abuse per se (e.g. overeating, internet gaming, gambling). As we did not test for genotypes, it could well be that some of the controls carry a genetic risk for drug abuse but refrain either because of age or environmental pressure. Hence some of the controls might exhibit the same striatal hyperactivity. Matching on genotyping could have avoided this possibility. Nevertheless, possible inclusion of 'at risk' controls would reduce the difference between groups, and therefore does not pose a bias toward finding an effect that is not true. In effect, both this and use of nicotine may well have led to our finding only a marginal effect, so more strict matching and exclusion of tobacco use are strongly recommended for future studies.

Also, the cross-sectional design limits conclusions on causal direction, as we cannot exclude the possibility that the observed differences in striatal brain function predated the onset of regular cannabis use. Finally, the exclusion of females prevented exploration of gender-specific differences in the impact of adolescent cannabis use on cognition and brain function, for which there is tentative evidence from animal studies (Giedd et al. 2006; Rodriguez de Fonseca et al. 1993).

In conclusion, striatal hyperactivity in adolescent cannabis users may signify an overly sensitive motivational brain circuitry and a diminished ability to disengage this circuit in the context of non-rewarding events. This could drive risk-seeking behavior like drug taking, even when facing the negative (non-rewarding) consequences.

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Figure 1.

Schematic presentation of the Monetary Incentive Delay (MID) task. Each trial starts with presentation of a cue signalling a potential rewarding (circle) or non-rewarding (square) trial. Following this cue a target is presented to which subjects had to respond as fast as possible by pressing a button. Finally, feedback is given on trial performance (either a reward of 1 euro/US dollar or no reward), in tandem with cumulative task earnings.

Figure 2. Anatomically defined masks of the left and right caudate nucleus (in blue and green respectively), and the left and right putamen (in purple and red). ROI masks are projected onto a normalized averaged group anatomy image (N=45) and are shown in neurological orientation (left = left). Slices display MNI (X,Y,Z)-coordinates 19, 3, 3.

Figure 3.

Mean reaction time (ms) $\pm$ standard error of mean (SEM) for responses to the target in reward trials and neutral (non-reward) trials, for cannabis users and controls. Both groups responded faster in rewarding trials compared to neutral trials ($p < 0.001$).

Figure 4.

Group activation maps (N=45) based on the contrasts anticipation reward trials versus anticipation neutral trials (left panel), and positive feedback versus negative feedback (right panel), thresholded at $T = 5.0$ ($p < 0.05$, FWE corrected). Slices displayed show MNI-coordinates (X,Y,Z) 8, 10, -1. Images are in neurological orientation, i.e. left = left.

Figure 5.

Brain activity levels (in arbitrary units (au)) in left and right caudate nucleus (CN) and putamen (Put) during anticipation of reward and neutral/non-reward targets, for cannabis users and controls. No significant group differences were found in activity during anticipation of reward targets, but cannabis users show higher levels of brain activity in the left CN and bilateral putamen in anticipation of neutral targets compared to non-using controls.

Figure 6.

In cannabis users (N=21) brain activity levels during neutral anticipation are significantly inversely correlated (Spearman's $\rho = -0.45$, $p < 0.05$) to the age of onset of cannabis use (on the X-axis in years of age). au = arbitrary units.

TABLE 1**Inclusion and Exclusion Criteria**

Inclusion criteria	Exclusion criteria
Male	Medical or neurological problems
Age between 12 – 19 years	Regular use of illegal drugs other than cannabis (> 10 episodes lifetime), except for alcohol or nicotine
Right handedness	Axis I psychiatric diagnosis, except for conduct disorder (which is a common diagnosis in cannabis using boys)
	IQ-scores below 80
	Use of psychotropic medication
	Contraindications for MRI (claustrophobia, metal objects, full dental braces)

TABLE 2

Demographics and Drug Use

	Cannabis users (n = 21) M (SD, range) or n	Cannabis-naïve controls (n = 24) M (SD, range) or n
Age	17.2 (1.0, 15 – 19)	16.8 (1.3, 13 – 19)
IQ *	101 (10.7, 82 – 116)	111 (11.6, 94 – 138)
Cannabis		
Lifetime (number of joints) **	4006 (7555, 224 – 32,850)	1.8 (4.0, 0 – 15)
Last year (number of joints) **	741 (772, 208 – 3528)	1.0 (2.9, 0 – 12)
Age of onset	13.2 (2.3, 8 – 16)	15.0 (1.6, 12 – 17)
Abstinence (weeks since last use)	5.1 (4.2, 1 – 16)	
Other substances (use last year)		
Alcohol (drinks/week) *	13.3 (13.6, 0 – 46)	3.4 (5.8, 0 – 24)
Tobacco (cigarettes/week) **	63.8 (53.4, 0 – 144)	6.1 (14.9, 0 – 53)
Ecstasy (episodes lifetime)	0.3 (0.9, 0 – 4)	0.1 (0.4, 0 – 2)
Amphetamines (episodes lifetime)	1.3 (2.4, 0 – 7)	--
Cocaine (episodes lifetime)	0.4 (0.9, 0 – 4)	--
Psilocybin ^A (episodes lifetime)	0.4 (0.6, 0 – 2)	0.1 (0.4, 0 – 2)
LSD (episodes lifetime)	0.1 (0.2, 0 – 1)	--
Laughing gas (episodes lifetime)	0.7 (2.1, 0 – 9)	0.1 (0.4, 0 – 2)
Benzodiazepines (episodes lifetime)	2.9 (8.8, 0 – 40)	0.2 (0.8, 0 – 4)
C-DISC		
Conduct disorder (yes/no) **	n = 9 (yes), n = 12 (no)	n = 24 (no)

Significance of differences calculated using independent samples T-tests (age, IQ, age of onset) and non-parametric Kolmogorov-Smirnov Z tests, two-tailed. Values between parentheses denote standard deviation and range.

^A psilocybin = magic mushrooms.

* p < 0.01 for cannabis users vs. cannabis-naïve controls.

** p < 0.001 for cannabis users vs. cannabis-naïve controls.