PLOS ONE

RESEARCH ARTICLE

Brain Cortical Thickness Differences in Adolescent Females with Substance Use Disorders

Peter K. Boulos[1], Manish S. Dalwani[2], Jody Tanabe[3], Susan K. Mikulich-Gilbertson[2], Marie T. Banich[4], Thomas J. Crowley[2], Joseph T. Sakai[2]*

1 University of Colorado Denver School of Medicine, Aurora, Colorado, United States of America, **2** Division of Substance Dependence, Department of Psychiatry, University of Colorado Denver School of Medicine, Aurora, Colorado, United States of America, **3** Department of Radiology, University of Colorado Denver School of Medicine, Aurora, Colorado, United States of America, **4** Institute of Cognitive Science, Department of Psychology and Neuroscience, University of Colorado Boulder, Boulder, Colorado, United States of America

* joseph.sakai@ucdenver.edu

Citation: Boulos PK, Dalwani MS, Tanabe J, Mikulich-Gilbertson SK, Banich MT, Crowley TJ, et al. (2016) Brain Cortical Thickness Differences in Adolescent Females with Substance Use Disorders. PLoS ONE 11(4): e0152983. doi:10.1371/journal.pone.0152983

Editor: Kewei Chen, Banner Alzheimer's Institute, UNITED STATES

Received: August 31, 2015

Accepted: March 22, 2016

Published: April 6, 2016

Data Availability Statement: Data are available upon request to qualified investigators, so long as the proposed work aligns with the approved study aims as consented to by study subjects. Requests may be sent to the corresponding author, Dr. Joseph T. Sakai (joseph.sakai@UCDenver.edu).

Funding: This work was supported by NIDA grants DA009842, DA011015, DA034604 and the Kane Family Foundation. Drs. Sakai, Crowley, Tanabe, and Mikulich-Gilbertson and Mr. Dalwani are also supported by NIDA grant DA031761. Dr. Sakai's lab is also supported by the Hewit Family Foundation.

Abstract

Some youths develop multiple substance use disorders early in adolescence and have severe, persistent courses. Such youths often exhibit impulsivity, risk-taking, and problems of inhibition. However, relatively little is known about the possible brain bases of these behavioral traits, especially among females.

Methods

We recruited right-handed female patients, 14–19 years of age, from a university-based treatment program for youths with substance use disorders and community controls similar for age, race and zip code of residence. We obtained 43 T1-weighted structural brain images (22 patients and 21 controls) to examine group differences in cortical thickness across the entire brain as well as six *a priori* regions-of-interest: 1) medial orbitofrontal cortex; 2) rostral anterior cingulate cortex; and 3) middle frontal cortex, in each hemisphere. Age and IQ were entered as nuisance factors for all analyses.

Results

A priori region-of-interest analyses yielded no significant differences. However, whole-brain group comparisons revealed that the left pregenual rostral anterior cingulate cortex extending into the left medial orbitofrontal region (355.84 mm^2 in size), a subset of two of our *a priori* regions-of-interest, was significantly thinner in patients compared to controls (vertex-level threshold $p = 0.005$ and cluster-level family wise error corrected threshold $p = 0.05$). The whole-brain group differences did not survive after adjusting for depression or externalizing scores. Whole-brain within-patient analyses demonstrated a positive association between cortical thickness in the left precuneus and behavioral disinhibition scores (458.23 mm^2 in size).

Peter Boulos' effort on this project was supported under R25DA033219.

Competing Interests: The authors of this manuscript have the following competing interests: Dr. Crowley recently served on the National Advisory Council of the National Institute on Drug Abuse, and on a Task Force of the American Psychiatric Association for drafting the Diagnostic and Statistical Manual of Mental Disorders, Edition 5. Dr. Sakai received reimbursement in 2012 for completing a policy review for the WellPoint Office of Medical Policy & Technology Assessment (OMPTA), WellPoint, Inc., Thousand Oaks, CA. He previously served on the board of the ARTS Foundation. The other authors report no conflict of interest. Dr. Crowley's and Dr. Sakai's competing interests do not alter the authors' adherence to all PLOS ONE policies on sharing data and materials.

Conclusions

Adolescent females with substance use disorders have significant differences in brain cortical thickness in regions engaged by the default mode network and that have been associated with problems of emotional dysregulation, inhibition, and behavioral control in past studies.

Introduction

Some individuals have onset of substance use disorder (SUD) early in adolescence, develop multiple SUD diagnoses and have severe persistent courses [1, 2]. These youth exhibit problems of self-control and risk taking in real life and laboratory settings [2–4] and such problems of inhibition may stem from measurable brain differences. Brain structural differences associated with these behavioral phenotypes are poorly understood in females. Therefore, we tested whether adolescent females with early onset substance use problems differ from controls in brain cortical thickness.

A Focus on Youths with Child/Adolescent-Onset Substance Use Problems

Despite important and recent advances [5], our understanding of the neurobiology of SUDs remains insufficient. SUDs are common in the general population [6], are a source of great morbidity and mortality [6, 7], and exact a huge cost to society in drug-related crime, health care costs, and productivity losses [8]. Although many youths experiment with substances [9], most will not progress to develop a SUD [6]. While it is well documented that these disorders cluster within families [10] and are moderately heritable [11–14], it is not soundly understood at a biological level why some youth appear more prone to develop a SUD.

Considering those who develop a SUD, the peak age of onset is in later adolescence or young adulthood, with less common onset after age of 25 [15]. However, some individuals have onset of SUD early in adolescence, develop multiple SUD diagnoses, and have severe persistent courses [1, 2]. Youths in this population are likely to have a number of precursors, characteristic co-morbidities, and associated cognitive deficits. For example, youths with poor self-control [16], low constraint [17], and early problems with inhibition [18, 19] are at an increased risk for later developing SUDs. Youths with SUDs also display risk-taking [3], impulsivity [20], difficulty delaying gratification [21], and impaired performance on laboratory cognitive tasks [22, 23]. Youths of both genders with SUDs are also very likely to have co-morbid conduct disorder [24, 25] and individuals with conduct disorder on average initiate substance use at an early age [26]. While conduct disorder is more common in males, it is still prevalent in adolescent girls, representing the second most common psychiatric diagnosis in female adolescents [27]. This clustering of high externalizing behavior problems within individuals with SUD is sometimes referred to as behavioral disinhibition (BD), a highly heritable (h^2>0.8) latent trait [11,14, 19, 28] which has a strong genetic correlation with laboratory-measured problems of executive control [29].

A Female-Only Sample

Adolescence is a time in which many sex differences begin to emerge with regards to psychopathology (e.g., rates of depression; [30]) and these sex differences appear to be mirrored by sex

differences in brain development [31, 32]. Becker et al. review the broad literature of neural networks mediating addiction, highlighting clear sex-differences in dopaminergic, noradrenergic, corticotropic, opioid, and cholinergic pathways. The authors suggest that these differences may correlate with distinct clinical presentations of addiction in females and emphasize the importance of studying males and females separately [33]. Hardee et al. recently presented their findings from a longitudinal fMRI study showing clear differences between males and females in amygdala and premotor cortex activation, in support of the proposition that the development of SUD in females is more likely to be related to negative affectivity, whereas in males, risk may be more likely mediated by impulsiveness [34]. While there has been some examination of differences in behavior and brain anatomy/function in boys with and without SUD or related phenotypes [35–43], little research has examined girls [36, 44]. Behaviorally, SUD in girls looks somewhat different than boys. For example, although the prevalence of substance use is similar between young adolescent males and females, with increasing age a higher SUD prevalence develops in males [6]. Females also show telescoping effects, having faster rates of progression from use to dependence, resulting in more severe clinical profiles upon presentation to treatment despite less or equivalent total substance use [45]. Other studies of SUD suggest male-female differences in genetic contributors [46] and environmental risks [47].

Considering these differences in behavior, and the clear sex differences in brain anatomy/function during adolescence, it is reasonable that males and females may have different, as well as overlapping, biological underpinnings to SUD. As anatomical differences between boys with and without SUD have been clearly documented, the current paper focuses on anatomy in females with and without SUD. In addition, given the literature on externalizing problems and, especially in females, affect regulation, we also seek here to explore whether patient-control differences covary with the severity of these comorbidities.

Brain Cortical Thickness

Several brain regions have been implicated in *volumetric* studies of youths with serious SUD, youths with high BD, or similar phenotypes. These include the insula [35, 36], dorsolateral prefrontal cortex (DLPFC) [37], orbitofrontal cortex (OFC) [38], and anterior cingulate cortex (ACC) [39–43], among others. This literature of volumetric studies is rapidly growing but, to our knowledge, few of these studies have focused on adolescent females specifically [36, 44]. In addition, relatively few studies of cerebral *cortical thickness* have been previously conducted on these adolescent phenotypes. Adolescent heavy marijuana users reportedly have cortical thinning in right caudal middle frontal regions, bilateral insula, and bilateral superior frontal cortex along with increased cortical thickness in the lingual, superior temporal, inferior parietal and paracentral regions [48]. Adolescents with "gaming addiction" [49] and "internet addiction" [50] have shown cortical thinning in the orbitofrontal cortex and elsewhere. However, to our knowledge, none of these previous studies focus on cortical thickness in female-only samples. Instead most prior studies have used male-only or mixed-sex samples. Although the literature on cortical thickness is more limited, available volumetric studies strongly suggest that prefrontal cortex, including the ACC, DLPFC, and OFC sub-regions are involved in SUD. These regions participate in behavior inhibition, executive functions, and decision-making [51]; localized lesions in these regions are associated with significant impairment in neuropsychological function, similar to those discussed with BD patients [52]. Thus, the ACC, DLPFC, and OFC are logical targets for region-of-interest analyses.

While volume and thickness are related, they are distinct phenotypes. According to the radial unit hypothesis, cells with the same origin are organized into columns, which run perpendicular to the brain's surface [53–56]. The number of columns determines surface area,

which is strongly related to grey matter volume, while the number of cells within a column determines cortical thickness [57, 58]. Both surface area and cortical thickness are heritable but available twin work supports that they have different genetic determinants [57, 59]. Thus, studying cortical thickness as we do here provides important complementary information to our recently published volumetric work [60].

Study Hypothesis

We hypothesized that whole-brain and region-of-interest analyses would identify differences in cortical thickness in prefrontal (especially anterior cingulate, middle frontal gyrus and orbitofrontal cortex) brain regions in female adolescents with early onset SUD, compared to controls.

Methods

The Colorado Multiple Institutions Review Board (COMIRB) approved all procedures and the study consents. Subjects 18 years of age or older provided written consent; those under 18 provided written assent while their parents provided written consent to study participation. Different data from this sample are reported in Dalwani et al., (volumetric results) [60] and Crowley et al., (fMRI data results) [61].

Sample

We report on 22 patients and 21 controls. All were female, aged 14–19 years, had an estimated IQ $\geq$ 80, and adequate English proficiency to understand the study consents.

Patients were recruited from a university-based adolescent treatment program for youths with serious substance and conduct problems and were required to meet criteria for at least one non-nicotine DSM-IV-TR substance abuse or dependence diagnosis [62]. To reduce confounds from intoxication or recent drug use, we required patients to have multiple negative urine (AccuTest™ for THC, cocaine, methamphetamine, amphetamine, barbiturates, benzodiazepines, MDMA, methadone, other opioids, PCP) and saliva (AlcoScreen™ for alcohol) tests for at least 7 days prior to scanning. 26 patients were enrolled in the study but did not complete MRI scanning for reasons including positive urinalysis, not meeting substance use disorder screening criteria, IQ $<$ 80, MRI contraindications, epilepsy, positive pregnancy test, court-ordered ankle monitor that could not be removed, or simply no longer willing to participate.

Controls, contacted first by advertising or by a research marketing company, were similar to the patient group with respect to age, race, and zip code of residence. Exclusion criteria for controls included previous court conviction (excluding minor traffic or curfew offenses), a substance-related arrest or treatment, school expulsions, meeting DSM-IV-TR criteria for a non-nicotine substance abuse or dependence diagnosis, meeting DSM-IV-TR criteria for conduct disorder in the last year, or a positive test for a non-prescribed substance about 7 days before and immediately prior to scanning using the same urine and saliva tests mentioned above. Four controls were enrolled in the study but did not complete MRI scanning for reasons including IQ $<$ 80 or meeting criteria for a non-nicotine SUD

For all subjects, we applied standard MRI exclusion criteria (e.g. orthodontic braces, claustrophobia, ferric metal in the body) for adolescents. Subjects with a positive pregnancy test, history of serious neurological illness, prior neurosurgery, or a history of unconsciousness lasting greater than 15 minutes were also excluded. Because the scanning session also acquired fMRI data using a paradigm requiring subjects to distinguish green from red for use in another study, color blindness was an additional exclusion criterion. Prior work showing cortical asymmetry amongst right- and left-handed individuals resulted in the exclusion of left-handed

adolescents from these analyses [63, 64]. Exclusion criteria for all subjects also included current high risk of suicide, psychosis, violence, or fire setting.

Assessments

Each participant completed numerous psychosocial assessments before MRI scanning [65]. Parents of each adolescent completed the Child Behavior Checklist (CBCL) and an updated Hollingshead Four-Factor Index of socioeconomic status [66]. The CBCL assessed attention-deficit/hyperactivity disorder (ADHD) symptoms and associated problems [67]. Each adolescent completed the vocabulary and matrix reasoning subtests of the Wechsler Abbreviated Scale of Intelligence to provide an estimate of IQ [68], the Youth Self Report (YSR), the Composite International Diagnostic Interview—Substance Abuse Module (CIDI-SAM), the Diagnostic Interview Schedule for Children (DISC), a Peak Aggression Scale (PAS) [2], the Eysenck Junior Impulsiveness Scale (EJIS) for a measure of impulsivity [69], and finally the Carroll Rating Scale for Depression (CRS). The YSR measure of ADHD symptoms was substituted for those participants (n = 6, all in the patient group) that did not have a CBCL available [70]. The CIDI-SAM and supplement served to generate DSM-IV SUD diagnoses and to determine recency of substance use, respectively [71]. From CIDI-SAM data, we also calculated SUMDEP, the total number of substance dependence symptoms across 10 different categories (range 0 to 70). We have used this measure in previous studies to compare groups on substance use severity [2]. The DISC assessed lifetime DSM-IV conduct disorder diagnoses [72] and the CRS estimated depression scores [73]. These assessments were completed in one session lasting approximately 3 hours.

Behavioral disinhibition (BD) scores combined information from 4 behavioral measures: DSM-IV symptom counts for conduct disorder, CBCL/YSR-measured scores of inattention (questions 8, 13,17, 61, and 80) and impulsivity (questions 1, 10, 36, 41, 45, 46, 62, 93, and 104), and sum of abuse/dependence symptoms across 10 drug categories. Subjects' scores were normed to a community sample of 414 adolescent females (i.e. number of standard deviations from the community sample mean). Utilizing this community sample, principal component analyses extracted the maximum covariance among the 4 behavioral measures and the resulting standardized factor loadings (on the first principal component) were utilized to weight our 4 standardized behavioral measures and sum them to generate BD scores (see http://ibgwww.colorado.edu/cadd/bd.html for details; [61]). We chose this validated measure of externalizing behavior, as opposed to other broader measures, as it takes into account those specific externalizing traits commonly comorbid with SUD (see Introduction, A Focus on Youths with Child/Adolescent-Onset Substance Use Problems).

MRI Parameters

A General Electric 3T MRI scanner was used to acquire high-resolution 3D T1-weighted images, taken along the coronal plane, using an SPGR-IR sequence and a standard quadrature head coil. The parameters were: TR = 9 ms, TE = 1.9 ms, T1 = 500 ms, flip angle = 10°, FOV = 220 mm^2, slice thickness = 1.7 mm, and matrix = 256x256, 0.97 x 0.97 mm^2 in plane. Scan time was 9 minutes and 12 seconds to acquire 124 slices.

Data Analyses

We compared groups for differences in demographic (e.g. age, race, SES) and diagnostic data (e.g. attention-deficit/hyperactivity disorder, conduct disorder, substance use disorder diagnoses) using SPSS software (IBM SPSS Statistics, Version 21. Chicago, IL: IBM Corp; 2012). Chi-square or Fisher's Exact tests were used to compare categorical variables and t-tests or Mann-

Whitney U tests were appropriately performed for continuous variables. We conducted all of these analyses using two-tailed tests at a 0.05 significance level.

FreeSurfer Analyses

MRI scans were reconstructed to measure cortical thickness using FreeSurfer software version 5.3.0. FreeSurfer reconstructs the images by first fitting the image to Talairach space, stripping non-brain structures from the image, forming the outermost grey matter boundary, and finally forming a white/grey matter boundary. The program utilizes triangular tessellation and surface deformation algorithms to form the boundaries. Cortical thickness is measured as the distance from the outer grey boundary, the pial surface, to the white/grey boundary [74]. A single team member blinded to the subjects' group affiliation ensured that the software performed the reconstructions properly by conducting a slice-by-slice visual inspection of each step of the reconstruction for all subjects in 3 planes (coronal, sagittal, and horizontal). As needed, edits were performed consistently throughout the sample and then the edited images were run through the program again. Necessary edits included: ensuring proper fit into Talairach space, manually stripping skull that the program missed, and adding control points to areas that were assuredly white matter but were not appropriately recognized as white matter. The temporal lobe commonly demanded edits. The effects of these edits on the results were examined (See S1 Text. Testing the Effects of Edits). The program then automatically parfcellated the reconstructed brain into regions according to Desikan's atlas [75].

Brain Morphometry Analysis

We conducted whole-brain and region-of-interest (ROI) analyses. The whole-brain analysis was performed using FreeSurfer's QDEC program while adjusting for age and IQ by entering them as nuisance factors. QDEC smoothed the data with a 10 mm full width at half maximum Gaussian kernel, while enforcing a Monte Carlo cluster correction (250 mm^2) with a vertex-level threshold of $p < 0.005$. SPSS was used to conduct the ROI analyses on extracted regions. We examined 3 ROIs bilaterally (total of 6 ROIs) defined by the Desikan's atlas [75] for our *a priori* predictions based on published literature (see Introduction, Brain Cortical Thickness, paragraph 1). These regions were: 1) medial orbitofrontal cortex (mOFC); 2) rostral anterior cingulate cortex (RACC); 3) middle frontal gyrus (MFG). In order to calculate MFG cortical thickness we combined surface-area-adjusted values for rostral middle frontal cortex and caudal middle frontal cortex as measured according to the Desikan's atlas. Regression analyses tested for group differences while controlling for age and IQ for each ROI. This approach to perform both whole-brain and *a priori* identified ROI analyses follows procedures used in past studies [31, 48].

Secondary Analyses

Patient-only regression analyses. We explored differences among patients that affect cortical thickness. To do this, we conducted within-patient regression analyses examining association of cortical thickness with recency of drug use (a single variable calculated from number of days since last use of any non-tobacco substance) and separately with severity of BD after adjusting for age and IQ. This was done as both a whole-brain vertex-level analysis and also utilizing a virtual mask to include only those regions that differed significantly in patient-control comparisons.

Testing how patient-control cortical thickness differences relate to differences in internalizing and externalizing behavior problems. To investigate how differences in cortical thickness between patients and controls might relate to internalizing and externalizing measures we performed additional QDEC analyses using the same procedures as for our primary whole-

brain analysis (described in section 2.6), with the same Monte Carlo cluster correction (250 mm^2) and vertex-level threshold ($p < 0.005$). In addition to age and IQ, we evaluated depression scores from the CRS, total anxiety scores from the YSR, total affectivity scores from the YSR, or total externalizing scores from the YSR as covariates in 4 separate analyses.

Testing for sex differences. Lastly, we have previously published very similar analyses testing brain cortical thickness patient-control differences in cortical thickness in a male adolescent sample [76]. This study used essentially the same recruitment procedures, inclusion/exclusion criteria and imaging parameters (see S1 Table. Comparing Inclusion and Exclusion Criteria for Our Female Sample with Male Sample Published Previously). Males and females from our patient, and separately control samples, were similar for demographic and clinical measures, except conduct disorder prevalence in patients (see S2 Table. Comparing Males and Females Within Patients and Within Controls for Demographics and Key Clinical Measures). Although our focus in this study is squarely on patient-control differences in a female sample, and we do not wish to duplicate reports of these previously published male patient-control findings, we utilized this male sample in these secondary analyses to explore sex differences. We therefore completed female vs male comparisons for cortical thickness differences, while controlling for age and IQ, within-patients and within-controls. Again, we used the same procedures and parameters as our primary whole-brain analysis.

Results

Demographic and Clinical Assessments

Table 1 compiles demographic, diagnostic, and substance use data along with other sample characteristics. There was a trend for age to differ between groups ($p = 0.08$) with controls being slightly older (16.67 years) than patients (16.09 years). Controls had significantly higher IQ than patients ($p = 0.004$; Mean IQ controls: 103.95; Mean IQ patients: 94.26). As a result we adjusted for age and IQ in all analyses. As expected, patients and controls significantly differed on various clinical measures including combined ADHD raw scores, lifetime conduct disorder diagnoses, aggression scores, impulsivity scores, and depression scores.

Region-of-Interest Analysis

Female patients and controls did not differ significantly in cortical thickness in regression analyses of the 6 regions of interest while controlling for age and IQ (Left-mOFC Beta = -0.16, $p = 0.37$; Right-mOFC Beta = -0.07, $p = 0.69$; Left-RACC Beta 0.22, $p = 0.24$; Right-RACC Beta = -0.06, $p = 0.76$; Left-MFG Beta 0.09, $p = 0.62$; Right-MFG Beta = 0.31, $p = 0.08$).

Whole-Brain Analysis

With specified vertex-level $p < 0.005$ and cluster threshold (250 mm^2), female patients had significantly less cortical thickness than controls in left pregenual rostral anterior cingulate cortex extending into the medial orbitofrontal region, including parts of Brodmann Areas 24, 32 and 10 (MNI coordinates for center of region: x = -6.7, y = 39.5, z = 2.6; see Fig 1). The region was 355.84 mm^2 in area and is a subset of both our RACC and mOFC *a priori* defined ROIs, but is not circumscribed by strict anatomical boundaries from either ROI.

Secondary Analyses

Patient-only regression analyses. Regression analyses within the patient group, adjusted for age and IQ, revealed no correlation between either recency of substance use nor BD scores with cortical thickness of the RACC-mOFC cluster identified in patient-control whole-brain

Table 1. Adolescent controls and patients: comparing demographic and diagnostic differences.

Measure	Controls (n = 21) mean(SEM) or n	Patients (n = 22) mean(SEM) or n	Test Statistic	p-value
Demographic Data				
Age in years	16.67 (0.25)	16.09 (0.20)	$t_{41} = 1.84$	0.08
Race				
Caucasian	13	12		
African American	1	1		
Hispanic	1	7		
Other	6	2		
Caucasian vs. non-Caucasian			$\chi^2 = 0.24$	0.62
Education-Highest grade completed	10.00 (0.30)	8.77 (0.17)	Mann-Whitney U	0.0021
Socioeconomic status[1]	36.14 (3.57)	45.19 (3.34)	$t_{35} = 1.80$	0.08
Diagnostic Data				
Estimated IQ	103.95 (2.26)	94.26 (2.23)	$t_{41} = 3.02$	0.004
Combined ADHD	1.48 (0.40)	5.68 (0.81)	$t_{30.60} = -4.66$	<0.001
CD lifetime diagnosis	0/21	14/22	$\chi^2 = 19.82$	<0.0001
Aggression[2]	0/21	21/22	$\chi^2 = 39.18$	<0.0001
Impulsivity	5.62 (1.00)	14.68 (1.23)	$t_{41} = 5.69$	<0.0001
Depression	4.33 (0.78)	10.95 (1.23)	$t_{35.12} = 4.50$	<0.0001
Lifetime DSM-IV-defined SUD				
Alcohol	0	19	$\chi^2 = 32.49$	<0.0001
Tobacco	0	10	Fisher's Exact	0.0005
Cannabis	0	20	$\chi^2 = 35.69$	<0.0001
Club Drugs	0	10	Fisher's Exact	0.0005
Cocaine	0	4	Fisher's Exact	0.11
Hallucinogens	0	1	Fisher's Exact	1
Amphetamine	0	4	Fisher's Exact	0.11
SUMDEP	0.24 (0.24)	13.09 (1.66)	Mann-Whitney U	<0.0001
Length of substance dependence[3]	N/A	1.53 years (0.29)		

[1] For 6 patients, parents did not complete questionnaires (SES and CBCL).

[2] Note: all controls had aggression scores of 0 (mean = 0/SE = 0). Twenty-one patients had recorded aggression scores >0 (range: 1-9/mean = 5.73/SE = 0.55).

[3] Length of substance dependence was calculated using these steps for each of the n = 20 patients meeting at least 1 substance dependence diagnosis. For one subject, considering all 10 drug categories, earliest age of substance dependence onset was subtracted from exact age at assessment.

Abbreviations: CD = conduct disorder; club drugs = ecstasy or MDMA, GHB, ketamine, rohypnol as defined by the CIDI-SAM; Combined ADHD = DSM-IV-TR defined attention-deficit/hyperactivity disorder raw scores measured using the CBCL or YSR (n = 6) if CBCL unavailable; estimated IQ = intelligence quotient estimated using the vocabulary and matrix reasoning subtests of the Wechsler Abbreviated Scale of Intelligence; SEM = standard error of the mean; SES = socioeconomic status measured using the Hollingshead Four-Factor Index; SUD = substance use disorders; SUMDEP = total number of substance dependence symptoms across 10 drug categories.

doi:10.1371/journal.pone.0152983.t001

analyses. Performing these regression analyses on a whole-brain basis did not show any associations between cortical thickness and recency of use. However, these whole-brain regression analyses identified a positive correlation between BD severity and cortical thickness in a cluster in the left precuneus measuring 458.23 mm^2 (MNI coordinates for center of region: x = -21.1, y = -61.3, z = 17.7; see Fig 2).

Testing how patient-control cortical thickness differences relate to differences in internalizing and externalizing behavior problems. The RACC-mOFC finding identified in our patient-control whole-brain analysis did not survive after additionally adjusting for either CRS

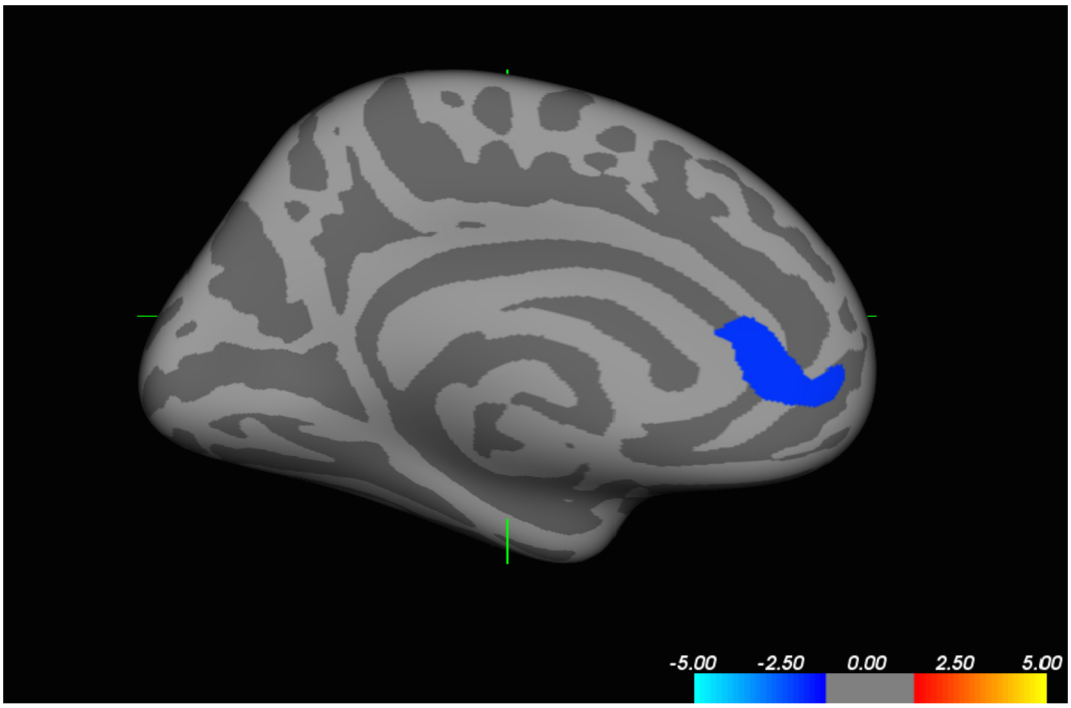

Fig 1. Whole-brain analyses testing for female patient-control differences in cortical thickness in QDEC using a vertex-level threshold of p<0.005 and Monte-Carlo simulation generated cluster level threshold. Medial view of left hemisphere here shows control>patient differences in cortical thickness of the pregenual rostral anterior cingulate cortex extending to the medial orbitofrontal cortex.

doi:10.1371/journal.pone.0152983.g001

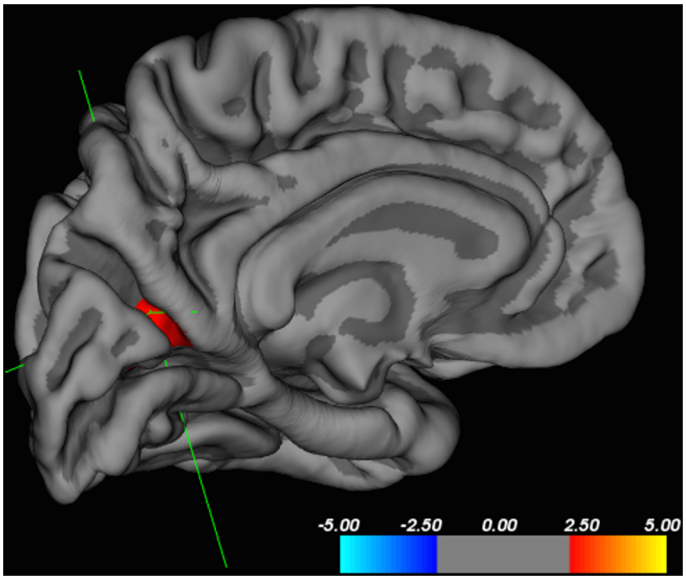

Fig 2. Whole-brain regression analyses within the patient group for correlation between cortical thickness and BD severity in QDEC (see Methods, Data Analyses and Discussion, ROI vs. Whole Brain Results for explanation of BD scores). Medial view of left hemisphere here shows positive correlation between BD scores and cortical thickness of the precuneus.

doi:10.1371/journal.pone.0152983.g002

depression scores or total externalizing scores from the YSR. The RACC-mOFC finding did survive, but was smaller, when additionally adjusting for either total anxiety (313.79 mm^2) or total affectivity scores (299.86 mm^2) from the YSR.

Testing for sex differences. Using our previously-published male sample [76] for comparisons, we found no differences in cortical thickness between male and female patients and no differences between male and female controls.

Discussion

Our results suggest that adolescent females with serious substance use problems have reduced cortical thickness in pregenual regions of the left rostral anterior cingulate and medial orbitofrontal cortex (RACC-mOFC) and that left precuneus cortical thickness is positively associated with BD scores within patients.

Current Understanding of RACC, mOFC, and Default Mode Network

The anterior cingulate and medial frontal cortices benefit from both a central anatomic location and rich interconnections with important regions and circuits, suggesting their potential importance in information processing and regulation. PET and functional MRI studies suggest an anatomical division of the anterior cingulate into rostral and caudal portions [77], with the RACC having dense connections to nucleus accumbens [78], limbic and affective systems, among other regions [43, 77]. RACC appears to play an important role in limbic regulation with respect to emotional processing and emotional conflict resolution [79, 80]. RACC is further implicated as the region primarily activated by self-referential thought and reflection [81].

A medial-lateral distinction has been proposed in OFC with the lateral OFC evaluating punishments and playing a role in response inhibition, and mOFC subserving monitoring and learning related to reinforcer valuation [82]. OFC has efferent or reciprocal connections with various brain regions including ACC, amygdala, caudate, and ventral tegmental area [83]. Through those connections, OFC may play a role in motivated behavior and assigning emotional valence to possible actions [84]. Thus together RACC and mOFC can be conceived of as an important hub, integrating sensory and visceral information, valuing reward of potential choices, and driving emotional reflection and response; these regions also play important roles in learning from errors, and engaging cognitive control regions (e.g. lateral prefrontal cortex) when necessary [79, 80, 82, 85, 86].

However, as shown in Fig 1, our RACC-mOFC cortical thickness finding from our whole-brain analyses, covers only a small portion of these regions and based on size and location appears to implicate an important hub of the Default Mode Network (DMN) [87, 88]. The DMN activates in task-free periods of rest and is characterized by functional connectivity between the posterior cingulate/precuneus, inferior parietal cortex, and the ventromedial prefrontal cortex [87]. These connections develop with age from a "local to a distributed" network. Connectivity and activation in DMN regions are seen in children as young as 8 years of age, and adolescence represents a time of DMN maturation [89]. The DMN is associated with intrinsic thought, self-reflection, and emotional processing with instructions such as, "focus on one's feelings, one's character, one's memories, and one's aspirations" activating the network [87]. Thus, the observed group differences in cortical thickness within RACC-mOFC map well to a hub of the DMN and may indicate patient-control differences in DMN, though resting state functional data would be required to confirm this.

Relating Our Findings to Female Youths with Substance Use Disorders

Our finding of control>patient cortical thickness in the left pregenual RACC-mOFC is consistent with two complementary lines of research. First, available neuroimaging work on

substance use disordered populations implicates patient-control differences in this important region. Second, available data on roles subserved by RACC and OFC fit with phenotypic descriptions of female youths with SUD.

Past MRI studies of both healthy and SUD populations point to this medial prefrontal region. For example, in normative populations, past work has suggested a link between children's anterior cingulate volume with performance on a Go/No-Go task [43]. A negative relationship has been demonstrated between OFC cortical thickness and impulsivity in adults [90], and also between RACC cortical thickness and impulsive aggression in children [41]. Studies of SUD populations compared to controls have shown significant hypoactivity of RACC during Go/No-Go tasks [39, 40] and less grey matter concentration in mOFC and ACC, among other regions, in cocaine-dependent adults [91, 92]. Studies of grey matter volume have also linked smaller OFC volume and conduct disorder [38], though other studies implicate other frontal [37] or temporal [35] regions. Compellingly, cannabis and ecstasy users show reduced deactivation in DMN during Go/No-Go task performance compared to healthy controls indicating a failure to inhibit default-mode circuitry [93]. Thus, several lines of research link hypoactivity, less grey matter volume, and less cortical thickness within the region identified in our whole-brain analyses as affected or altered in SUD.

As described above (see Discussion, Current Understanding of RACC, mOFC, and Default Mode Network) RACC-OFC has been proposed as one important hub for integration of emotional, sensory and visceral information, aiding in valuation of expected rewards for competing potential choices. Therefore, individuals with deficits in these regions might be hypothesized to have difficulty with affective control. Relating this with our sample of adolescent females, we see a phenotypic link with emphasis placed on emotional dysregulation. Congruent with this idea, development of SUD in females is associated with negative affectivity and emotional reactivity, along with externalizing behavior problems, while in adolescent males SUD is mainly associated with externalizing problems [47]. Patients with depression have a blunted ability to down-regulate DMN activity compared to controls, associating abnormal DMN activity with depressive rumination [94, 95]. Our patient group indeed showed significantly greater depression scores compared with controls (see Table 1).

SUD involves compulsive pursuit of the drug along with craving. Such characteristics have been hypothesized to be related to OFC and its strong connections with limbic and reward pathways [84]. Individuals with lesions to ACC/OFC may exhibit externalizing behaviors, including problems of inhibition and poor behavioral control [84], problems often seen in youths with or at risk for SUD [3, 16, 20]. Patients like ours may display emotional dysregulation [96], problems of executive control and inhibition [29], impulsiveness [20, 97], problems with error processing, and difficulty learning from punishment [98]. ACC and OFC dysfunction can be reasonably related to all of these traits.

We attempted to test empirically whether our patient-control cortical thickness difference in left pregenual RACC-mOFC was better explained by problems of affect regulation or externalizing behavior problems. However, we found that controlling for both depression severity and severity of externalizing behavior problems eliminated the patient-control difference. While this result is not simple or straightforward and does not link one co-morbidity to our brain finding, it suggests that risk for SUD in female adolescents may be complex, implicating both depression and externalizing problems.

Sex Differences from Secondary Analyses

We unexpectedly found no differences in cortical thickness between our prior male and current female samples (see Results, Secondary Analyses). Although sex differences have been

demonstrated in regions relevant to the current study (e.g., medial oribitofrontal cortex) [99,100], sex differences in cortical thickness appear much less prominent than sex differences in cortical volume and surface area [100]. Males and females differ in cortical thickness developmental trajectories [99] but in many instances those trajectories appear to decussate in the adolescent years (see Figure 4 in publication [99]). For these reasons, detection of sex differences for cortical thickness in SUD populations may require larger samples than those utilized here and perhaps a focus on younger or older populations; in other words, the lack of sex differences demonstrated here may be because of limited power and our adolescent focus. These results might support that future studies focusing on brain cortical thickness in this population might consider studying males and females together. However, we would suggest caution in this approach. Our male and female patients differed significantly in the prevalence of conduct disorder. Thus, as expected from the extant literature, the pattern of co-morbidity in adolescent males and females with SUD differed and prior authors suggest important potential phenotypic differences in males and females (see Introduction, A Female-Only Sample). Studying females specifically allowed examining the unique contributions of internalizing and externalizing scores on our cortical thickness finding. As mentioned above, our analyses may have missed important but smaller male:female differences in cortical thickness due to our modest sample sizes (see Discussion, Limitations).

The Meaning of Cortical Thinning in Relation to Function

Although we demonstrate that patients compared to controls had thinner cortex in a portion of the RACC/OFC, it is not clear what the functional relevance of such "thinning" represents. There are certainly normative age related changes in cortical thickness, including both synaptogenesis and then thinning in the adolescent years. That thinning is hypothesized to be related to important maturational changes that improve synaptic efficiency such as increased myelination and synaptic pruning [101, 102]. Therefore, in some instances cortical thinning has been associated with improved function, (e.g., with improved general verbal intellectual functioning; [103]), and failure of normative cortical thinning has also been linked to problems of emotional control and behavioral regulation [104]. In many other instances, cortical thinning is associated with neurological decline [105–107]. Differences seen here could relate to developmental differences in synaptogenesis, early or more extensive synaptic pruning, substance-related injury, among other possible explanations. Longitudinal imaging designs of this important population are needed to better place such findings in a developmental context.

ROI vs. Whole Brain Results

It is important to note that while our whole brain analyses demonstrated patient-control differences in left RACC/OFC, our region of interest analyses (which also looked at ACC and OFC) did not yield significant group differences. While this appears contradictory on first blush, the two approaches have many important differences. Our whole brain analyses search for areas of group difference where each vertex differs in cortical thickness (at a vertex-level $p<0.005$) while also requiring a Monte-Carlo-simulation-determined minimum number of contiguous vertices meeting the vertex-level requirement (250 mm^2). This approach allows us to find smaller focal regions of greater patient-control differences and allows identified brain regions of group differences to cross boundaries (e.g. different Brodmann Areas or different gyri). Our region of interest analyses test for group differences in average cortical thickness across larger pre-defined brain regions. This approach, compared to our whole brain analyses, can identify less extreme patient-control differences in cortical thickness ($p<0.05$ at the ROI level vs. $p<0.005$ at each vertex) within these regions. Hence, the two approaches are complementary.

Regression Analyses and Cortical Thickness

We completed regression analyses between cortical thickness with recency of drug use and separately with severity of externalizing behavior problems (BD scores) on a whole-brain vertex-level and within the left RACC-mOFC region identified in our primary patient-control comparisons. The purpose of this was to examine whether there was a significant component of cortical thinning secondary to direct substance effects that recovered with abstinence. Alternatively, we expected a correlation between BD severity and cortical thinning in the setting of predisposing brain differences. The utilization of BD scores, a measure that has been shown in the past to effectively capture a strongly heritable component of externalizing behaviors, allows us to propose the potential meaning of this correlation [28].

In performing these analyses at the whole-brain level we demonstrate no association with recency of use and one cluster showing a positive correlation between BD severity and cortical thickness within the left precuneus. This region is considered as the "functional core" of the DMN playing a "pivotal role" in the appropriate functioning of the network [108, 109]. Prior work has found increased precuneus connectivity with DMN regions in depressed subjects [110], which aligns with the hypothesis that our findings in our adolescent female population are associated with problems of affective control. We see both increased cortical thickness in the precuneus (in association with BD) and decreased thickness in the pregenual RACC-mOFC (between groups). Both are regions critical to the DMN, suggesting this network may be important to understanding patient-control differences in SUD risk in adolescent females.

Strengths and Limitations

This study provides the opportunity to add to our understanding of brain morphometry related specifically to SUD in adolescent females. Studying adolescents conveys some advantages. These adolescent patients have substance problems severe enough to merit treatment entry early in life (see Table 1). But unlike adult studies of samples with many more years of chronic substance exposure, these youths had relatively few years of heavy substance exposure (see bottom row Table 1); if the brain differences identified in this study are substance induced, such brain changes occur with relatively few years of heavy exposure in adolescence. Studying only females is another strength of this work. As highlighted in section 1.2, there is mounting evidence of important sex differences at the phenotypic level that may play a role in SUDs.

Our study also has several limitations. To our knowledge, this is one of the first studies testing differences in cortical thickness in SUD female youths, but our sample of 22 patients and 21 controls may have relatively modest power to detect whole-brain cortical thickness differences [111]. For example, Pardoe and colleagues [111] estimate under certain assumptions (e.g., alpha 0.05, two-sided test, 10mm surface-based smoothing, etc.) that 20 subjects per group has 80% power to detect mean cortical thickness differences of about 0.4–0.5 mm, with some variability based on lobe of interest. Therefore, regions with modest between-group differences may not have been identified in our current analyses. Future studies with larger samples will reduce these concerns. Also, we study female adolescents, and our results should not be extrapolated to male adolescents or to adults with serious substance problems. Additionally, although we describe less cortical thickness in patients versus controls, the functional relevance of such cortical thinning is not fully understood (as described in Section 4.3). Finally, as is apparent from our inclusion/exclusion criteria, we did not clean our sample of comorbid mental health concerns, broadly including female adolescents with SUD. By taking this "broad" approach, we are able to recruit a sample that is more representative of the treatment population of interest. Given that SUD, externalizing behavior problems, as well as problems of impulsivity and/or high novelty seeking tend to cluster within individuals [65, 97] in a highly

heritable fashion [11, 14, 19, 28], removing all comorbidity would lead to an atypical, less severely affected sample [112]. Nevertheless, our "broad" approach does result in some limitations, most notably that we cannot assess the contributions of specific diagnosis to a specific finding. However, our "broad" approach provides complementary information to studies employing a "narrow" strategy to recruit subjects with a single SUD and no other co-morbid disorders.

Future Directions

Various studies of brain morphometry in SUD youths, or similar phenotypes, now suggest either cortical thinning [48, 113–117] or less grey matter volume [35, 37, 38] in such youths, with some important exceptions [118]. Many of these studies further implicate various frontal regions important to decision-making and executive control. Unfortunately, no brain morphometric changes appear clearly pathognomonic. Future studies could benefit from larger sample sizes to identify group differences with smaller effect sizes. Longitudinal designs may also better separate predisposing from substance-induced changes and identify patient-control differences in developmental trajectories. If clear and replicable brain differences associated with SUD are identified, approaches such as transcranial direct current stimulation might be employed to test potential mitigation of such behavior problems.

Supporting Information

S1 Table. Comparing Inclusion and Exclusion Criteria for Our Female Sample with Male Sample Published Previously.
(DOCX)

S2 Table. Comparing Males and Females Within Patients and Within Controls for Demographics and Key Clinical Measures.
(DOCX)

S1 Text. Testing the Effects of Edits.
(DOCX)

Author Contributions

Conceived and designed the experiments: PKB MSD JT SKM MTB TJC JTS. Performed the experiments: PKB MSD JT SKM MTB TJC JTS. Analyzed the data: MSD PKB JTS SKM. Wrote the paper: PKB MSD JT SKM MTB TJC JTS.

References

1. Chen CY, Storr CL, Anthony JC. Early-onset drug use and risk for drug dependence problems. Addict Behav. 2009; 34(3):319–22. doi: 10.1016/j.addbeh.2008.10.021 PMID: 19022584
2. Crowley TJ, Mikulich SK, Ehlers KM, Whitmore EA, MacDonald MJ. Validity of structured clinical evaluations in adolescents with conduct and substance problems. J Am Acad Child Adolesc Psychiatry. 2001; 40(3):265–73. PMID: 11288767
3. Crowley TJ, Raymond KM, Mikulich-Gilbertson SK, Thompson LL, Lejuez CW. A risk-taking "set" in a novel task among adolescents with serious conduct and substance problems. J Am Acad Child Adolesc Psychiatry. 2006; 45(2):175–83. PMID: 16429088
4. Thurstone C, Salomonsen-Sautel S, Mikulich-Gilbertson SK, Hartman CA, Sakai JT, Hoffenberg AS, et al. Prevalence and predictors of injection drug use and risky sexual behaviors among adolescents in substance treatment. Am J Addict. 2013; 22(6):558–65. doi: 10.1111/j.1521-0391.2013.12064.x PMID: 24131163
5. Volkow ND, Baler RD. Addiction science: Uncovering neurobiological complexity. Neuropharmacology. 2014; 76, Part B(0):235–49.

6. Substance Abuse and Mental Health Services Administration. Results from the 2012 National Survey on Drug Use and Health: Summary of National Findings. Rockville, MD: Substance Abuse and Mental Health Services Administration. 2013.
7. Substance Abuse and Mental Health Services Administration. The DAWN Report: Highlights of the 2011 Drug Abuse Warning Network (DAWN) Findings on Drug-Related Emergency Department Visits. Rockville, MD: Center for Behavioral Health Statistics and Quality; 2013.
8. United States Department of Justice. The Economic Impact of Illicit Drug Use on American Society. Washington, DC: National Drug Intelligence Center; 2011.
9. Johnston LD, O'Malley PM, Bachman JG, Schulenberg JE. Monitoring the Future National Survey Results on Drug Use, 1975–2011. Volume I: Secondary School Students. Ann Arbor, MI: Institute for Social Research, University of Michigan; 2012.
10. Tyrfingsson T, Thorgeirsson TE, Geller F, Runarsdóttir V, Hansdóttir I, Bjornsdottir G, et al. Addictions and their familiality in Iceland. Ann N Y Acad Sci. 2010; 1187:208–17. doi: 10.1111/j.1749-6632.2009.05151.x PMID: 20201855
11. Hicks BM, Krueger RF, Iacono WG, McGue M, Patrick CJ. Family transmission and heritability of externalizing disorders: a twin-family study. Arch Gen Psychiatry. 2004; 61(9):922–8. PMID: 15351771
12. Hicks BM, Iacono WG, McGue M. Index of the transmissible common liability to addiction: heritability and prospective associations with substance abuse and related outcomes. Drug Alcohol Depend. 2012; 123 Suppl 1:S18–23. doi: 10.1016/j.drugalcdep.2011.12.017 PMID: 22245078
13. Kendler KS, Prescott CA, Myers J, Neale MC. The structure of genetic and environmental risk factors for common psychiatric and substance use disorders in men and women. Arch Gen Psychiatry. 2003; 60(9):929–37. PMID: 12963675
14. Krueger RF, Hicks BM, Patrick CJ, Carlson SR, Iacono WG, McGue M. Etiologic connections among substance dependence, antisocial behavior, and personality: modeling the externalizing spectrum. J Abnorm Psychol. 2002; 111(3):411–24. PMID: 12150417
15. Compton WM, Thomas YF, Stinson FS, Grant BF. Prevalence, correlates, disability, and comorbidity of DSM-IV drug abuse and dependence in the United States: results from the national epidemiologic survey on alcohol and related conditions. Arch Gen Psychiatry. 2007; 64(5):566–76. PMID: 17485608
16. Moffitt TE, Arseneault L, Belsky D, Dickson N, Hancox RJ, Harrington H, et al. A gradient of childhood self-control predicts health, wealth, and public safety. Proc Natl Acad Sci U S A. 2011; 108(7):2693–8. doi: 10.1073/pnas.1010076108 PMID: 21262822
17. Krueger RF. Personality traits in late adolescence predict mental disorders in early adulthood: a prospective-epidemiological study. J Pers. 1999; 67(1):39–65. PMID: 10030020
18. McGue M, Iacono WG. The association of early adolescent problem behavior with adult psychopathology. Am J Psychiatry. 2005; 162(6):1118–24. PMID: 15930060
19. Iacono WG, Carlson SR, Taylor J, Elkins IJ, McGue M. Behavioral disinhibition and the development of substance-use disorders: findings from the Minnesota Twin Family Study. Dev Psychopathol. 1999; 11(4):869–900. PMID: 10624730
20. Thompson LL, Whitmore EA, Raymond KM, Crowley TJ. Measuring impulsivity in adolescents with serious substance and conduct problems. Assessment. 2006; 13(1):3–15. PMID: 16443715
21. Field M, Christiansen P, Cole J, Goudie A. Delay discounting and the alcohol Stroop in heavy drinking adolescents. Addiction. 2007; 102(4):579–86. PMID: 17309540
22. Squeglia LM, Jacobus J, Tapert SF. The influence of substance use on adolescent brain development. Clin EEG Neurosci. 2009; 40(1):31–8. PMID: 19278130
23. Tapert SF, Granholm E, Leedy NG, Brown SA. Substance use and withdrawal: neuropsychological functioning over 8 years in youth. J Int Neuropsychol Soc. 2002; 8(7):873–83. PMID: 12405538
24. Young SE, Mikulich SK, Goodwin MB, Hardy J, Martin CL, Zoccolillo MS, et al. Treated delinquent boys' substance use: onset, pattern, relationship to conduct and mood disorders. Drug Alcohol Depend. 1995; 37(2):149–62. PMID: 7758404
25. Crowley TJ, Riggs PD. Adolescent substance use disorder with conduct disorder and comorbid conditions. NIDA Res Monogr. 1995; 156:49–111. PMID: 8594479
26. Hopfer C, Salomonsen-Sautel S, Mikulich-Gilbertson S, Min SJ, McQueen M, Crowley T, et al. Conduct disorder and initiation of substance use: a prospective longitudinal study. J Am Acad Child Adolesc Psychiatry. 2013; 52(5):511–8.e4. doi: 10.1016/j.jaac.2013.02.014 PMID: 23622852
27. Pajer K, Stein S, Tritt K, Chang CN, Wang W, Gardner W. Conduct disorder in girls: neighborhoods, family characteristics, and parenting behaviors. Child Adolesc Psychiatry Ment Health. 2008; 2(1):28. doi: 10.1186/1753-2000-2-28 PMID: 18837974

28. Young SE, Stallings MC, Corley RP, Krauter KS, Hewitt JK. Genetic and environmental influences on behavioral disinhibition. American journal of medical genetics. 2000; 96(5):684–95. PMID: 11054778

29. Friedman NP, Miyake A, Robinson JL, Hewitt JK. Developmental trajectories in toddlers' self-restraint predict individual differences in executive functions 14 years later: a behavioral genetic analysis. Dev Psychol. 2011; 47(5):1410–30. doi: 10.1037/a0023750 PMID: 21668099

30. Rutter M, Caspi A, Moffitt TE. Using sex differences in psychopathology to study causal mechanisms: unifying issues and research strategies. J Child Psychol Psychiatry. 2003; 44(8):1092–115. PMID: 14626453

31. Tanabe J, York P, Krmpotich T, Miller D, Dalwani M, Sakai JT, et al. Insula and orbitofrontal cortical morphology in substance dependence is modulated by sex. AJNR Am J Neuroradiol. 2013; 34 (6):1150–6. doi: 10.3174/ajnr.A3347 PMID: 23153869

32. Goldstein JM, Seidman LJ, Horton NJ, Makris N, Kennedy DN, Caviness VS, et al. Normal sexual dimorphism of the adult human brain assessed by in vivo magnetic resonance imaging. Cereb Cortex. 2001; 11(6):490–7. PMID: 11375910

33. Becker JB, Perry AN, Westenbroek C. Sex differences in the neural mechanisms mediating addiction: a new synthesis and hypothesis. Biol Sex Differ. 2012; 3(1):14. doi: 10.1186/2042-6410-3-14 PMID: 22676718

34. Hardee J, Cope LM, Zucker R, Heitzeg M, editors. Gender differences in the development of emotion circuitry in youth at risk for substance abuse: A longitudinal fMRI study. College on Problems of Drug Dependence 77th Annual Meeting; 2015 June 16, 2015; Phoenix, AZ.

35. Sterzer P, Stadler C, Poustka F, Kleinschmidt A. A structural neural deficit in adolescents with conduct disorder and its association with lack of empathy. Neuroimage. 2007; 37(1):335–42. PMID: 17553706

36. Fairchild G, Hagan CC, Walsh ND, Passamonti L, Calder AJ, Goodyer IM. Brain structure abnormalities in adolescent girls with conduct disorder. J Child Psychol Psychiatry. 2013; 54(1):86–95. doi: 10.1111/j.1469-7610.2012.02617.x PMID: 23082797

37. Dalwani M, Sakai JT, Mikulich-Gilbertson SK, Tanabe J, Raymond K, McWilliams SK, et al. Reduced cortical gray matter volume in male adolescents with substance and conduct problems. Drug Alcohol Depend. 2011; 118(2–3):295–305. doi: 10.1016/j.drugalcdep.2011.04.006 PMID: 21592680

38. Huebner T, Vloet TD, Marx I, Konrad K, Fink GR, Herpertz SC, et al. Morphometric brain abnormalities in boys with conduct disorder. J Am Acad Child Adolesc Psychiatry. 2008; 47(5):540–7. doi: 10.1097/CHI.0b013e3181676545 PMID: 18356764

39. Kaufman JN, Ross TJ, Stein EA, Garavan H. Cingulate hypoactivity in cocaine users during a GO-NOGO task as revealed by event-related functional magnetic resonance imaging. J Neurosci. 2003; 23(21):7839–43. PMID: 12944513

40. Forman SD, Dougherty GG, Casey BJ, Siegle GJ, Braver TS, Barch DM, et al. Opiate addicts lack error-dependent activation of rostral anterior cingulate. Biol Psychiatry. 2004; 55(5):531–7. PMID: 15023582

41. Ducharme S, Hudziak JJ, Botteron KN, Ganjavi H, Lepage C, Collins DL, et al. Right anterior cingulate cortical thickness and bilateral striatal volume correlate with child behavior checklist aggressive behavior scores in healthy children. Biol Psychiatry. 2011; 70(3):283–90. doi: 10.1016/j.biopsych.2011.03.015 PMID: 21531391

42. Benegal V, Antony G, Venkatasubramanian G, Jayakumar PN. Gray matter volume abnormalities and externalizing symptoms in subjects at high risk for alcohol dependence. Addict Biol. 2007; 12 (1):122–32. PMID: 17407506

43. Bush G, Luu P, Posner MI. Cognitive and emotional influences in anterior cingulate cortex. Trends Cogn Sci. 2000; 4(6):215–22. PMID: 10827444

44. Fein G, Greenstein D, Cardenas VA, Cuzen NL, Fouche JP, Ferrett H, et al. Cortical and subcortical volumes in adolescents with alcohol dependence but without substance or psychiatric comorbidities. Psychiatry Res. 2013; 214(1):1–8. doi: 10.1016/j.pscychresns.2013.06.001 PMID: 23916536

45. Greenfield SF, Back SE, Lawson K, Brady KT. Substance abuse in women. Psychiatr Clin North Am. 2010; 33(2):339–55. doi: 10.1016/j.psc.2010.01.004 PMID: 20385341

46. Hicks BM, Blonigen DM, Kramer MD, Krueger RF, Patrick CJ, Iacono WG, et al. Gender differences and developmental change in externalizing disorders from late adolescence to early adulthood: A longitudinal twin study. J Abnorm Psychol. 2007; 116(3):433–47. PMID: 17696699

47. Silberg J, Rutter M, D'Onofrio B, Eaves L. Genetic and environmental risk factors in adolescent substance use. J Child Psychol Psychiatry. 2003; 44(5):664–76. PMID: 12831111

48. Lopez-Larson MP, Bogorodzki P, Rogowska J, McGlade E, King JB, Terry J, et al. Altered prefrontal and insular cortical thickness in adolescent marijuana users. Behav Brain Res. 2011; 220(1):164–72. doi: 10.1016/j.bbr.2011.02.001 PMID: 21310189

49. Yuan K, Cheng P, Dong T, Bi Y, Xing L, Yu D, et al. Cortical thickness abnormalities in late adolescence with online gaming addiction. PLoS One. 2013; 8(1):e53055. doi: 10.1371/journal.pone.0053055 PMID: 23326379

50. Hong SB, Kim JW, Choi EJ, Kim HH, Suh JE, Kim CD, et al. Reduced orbitofrontal cortical thickness in male adolescents with internet addiction. Behav Brain Funct. 2013; 9:11. doi: 10.1186/1744-9081-9-11 PMID: 23497383

51. Crews FT, Boettiger CA. Impulsivity, frontal lobes and risk for addiction. Pharmacol Biochem Behav. 2009; 93(3):237–47. doi: 10.1016/j.pbb.2009.04.018 PMID: 19410598

52. Bonelli RM, Cummings JL. Frontal-subcortical circuitry and behavior. Dialogues Clin Neurosci. 2007; 9(2):141–51. PMID: 17726913

53. Mountcastle VB. The columnar organization of the neocortex. Brain. 1997; 120 (Pt 4):701–22. PMID: 9153131

54. Rakic P. Specification of cerebral cortical areas. Science. 1988; 241(4862):170–6. PMID: 3291116

55. Rakic P. A small step for the cell, a giant leap for mankind: a hypothesis of neocortical expansion during evolution. Trends Neurosci. 1995; 18(9):383–8. PMID: 7482803

56. Rakic P. The radial edifice of cortical architecture: from neuronal silhouettes to genetic engineering. Brain Res Rev. 2007; 55(2):204–19. PMID: 17467805

57. Winkler AM, Kochunov P, Blangero J, Almasy L, Zilles K, Fox PT, et al. Cortical thickness or grey matter volume? The importance of selecting the phenotype for imaging genetics studies. Neuroimage. 2010; 53(3):1135–46. doi: 10.1016/j.neuroimage.2009.12.028 PMID: 20006715

58. Pakkenberg B, Gundersen HJ. Neocortical neuron number in humans: effect of sex and age. J Comp Neurol. 1997; 384(2):312–20. PMID: 9215725

59. Panizzon MS, Fennema-Notestine C, Eyler LT, Jernigan TL, Prom-Wormley E, Neale M, et al. Distinct genetic influences on cortical surface area and cortical thickness. Cereb Cortex. 2009; 19(11):2728–35. doi: 10.1093/cercor/bhp026 PMID: 19299253

60. Dalwani MS, McMahon MA, Mikulich-Gilbertson SK, Young SE, Regner MF, Raymond KM, et al. Female adolescents with severe substance and conduct problems have substantially less brain gray matter volume. PLoS One. 2015; 10(5):e0126368. doi: 10.1371/journal.pone.0126368 PMID: 26000879

61. Crowley TJ, Dalwani MS, Mikulich-Gilbertson SK, Young SE, Sakai JT, Raymond KM, et al. Adolescents' Neural Processing of Risky Decisions: Effects of Sex and Behavioral Disinhibition. PLoS One. 2015; 10(7):e0132322. doi: 10.1371/journal.pone.0132322 PMID: 26176860

62. American Psychiatric Association. Diagnostic and statistical manual of mental disorders, (DSM-IV-TR). Washington, DC: American Psychiatric Pub; 2000.

63. Jung P, Baumgärtner U, Bauermann T, Magerl W, Gawehn J, Stoeter P, et al. Asymmetry in the human primary somatosensory cortex and handedness. Neuroimage. 2003; 19(3):913–23. PMID: 12880820

64. Sörös P, Knecht S, Imai T, Gürtler S, Lütkenhöner B, Ringelstein EB, et al. Cortical asymmetries of the human somatosensory hand representation in right- and left-handers. Neurosci Lett. 1999; 271 (2):89–92. PMID: 10477109

65. Crowley TJ, Dalwani MS, Mikulich-Gilbertson SK, Du YP, Lejuez CW, Raymond KM, et al. Risky decisions and their consequences: neural processing by boys with Antisocial Substance Disorder. PLoS One. 2010; 5(9):e12835. doi: 10.1371/journal.pone.0012835 PMID: 20877644

66. Hollingshead AB. Four-Factor Index of Social Status. New Haven: Yale University Department of Sociology. Unpublished manuscript. 1975.

67. Achenbach TM. Manual for the Child Behavior Checklist/4-18 and 1991 Profile. Burlington, VT: University of Vermont Department of Psychiatry; 1991.

68. Wechsler D. Wechsler Abbreviated Scale of Intelligence (WASI) manual. San Antonio, TX: The Psychological Corporation; 1999.

69. Eysenck SB, Eysenck HJ. Impulsiveness and venturesomeness: their position in a dimensional system of personality description. Psychol Rep. 1978; 43(3 Pt 2):1247–55. PMID: 746091

70. Achenbach TM. Manual for the youth self-report and 1991 profile. Burlington, VT: University of Vermont Department of Psychiatry; 1991.

71. Robins LN, Wing J, Wittchen HU, Helzer JE, Babor TF, Burke J, et al. The Composite International Diagnostic Interview. An epidemiologic Instrument suitable for use in conjunction with different diagnostic systems and in different cultures. Arch Gen Psychiatry. 1988; 45(12):1069–77. PMID: 2848472

72. Shaffer D, Fisher P, Lucas CP, Dulcan MK, Schwab-Stone ME. NIMH Diagnostic Interview Schedule for Children Version IV (NIMH DISC-IV): description, differences from previous versions, and reliability

of some common diagnoses. J Am Acad Child Adolesc Psychiatry. 2000; 39(1):28–38. PMID: 10638065

73. Carroll BJ, Feinberg M, Smouse PE, Rawson SG, Greden JF. The Carroll rating scale for depression. I. Development, reliability and validation. Br J Psychiatry. 1981; 138:194–200. PMID: 7272609
74. Fischl B, Dale AM. Measuring the thickness of the human cerebral cortex from magnetic resonance images. Proc Natl Acad Sci U S A. 2000; 97(20):11050–5. PMID: 10984517
75. Desikan RS, Ségonne F, Fischl B, Quinn BT, Dickerson BC, Blacker D, et al. An automated labeling system for subdividing the human cerebral cortex on MRI scans into gyral based regions of interest. Neuroimage. 2006; 31(3):968–80. PMID: 16530430
76. Chumachenko SY, Sakai JT, Dalwani MS, Mikulich-Gilbertson SK, Dunn R, Tanabe J, et al. Brain cortical thickness in male adolescents with serious substance use and conduct problems. Am J Drug Alcohol Abuse. 2015:1–11.
77. Margulies DS, Kelly AM, Uddin LQ, Biswal BB, Castellanos FX, Milham MP. Mapping the functional connectivity of anterior cingulate cortex. Neuroimage. 2007; 37(2):579–88. PMID: 17604651
78. Rushworth MF, Behrens TE, Rudebeck PH, Walton ME. Contrasting roles for cingulate and orbitofrontal cortex in decisions and social behaviour. Trends Cogn Sci. 2007; 11(4):168–76. PMID: 17337237
79. Etkin A, Egner T, Peraza DM, Kandel ER, Hirsch J. Resolving emotional conflict: a role for the rostral anterior cingulate cortex in modulating activity in the amygdala. Neuron. 2006; 51(6):871–82. PMID: 16982430
80. Etkin A, Egner T, Kalisch R. Emotional processing in anterior cingulate and medial prefrontal cortex. Trends Cogn Sci. 2011; 15(2):85–93. doi: 10.1016/j.tics.2010.11.004 PMID: 21167765
81. D'Argembeau A, Collette F, Van der Linden M, Laureys S, Del Fiore G, Degueldre C, et al. Self-referential reflective activity and its relationship with rest: a PET study. Neuroimage. 2005; 25(2):616–24. PMID: 15784441
82. Kringelbach ML. The human orbitofrontal cortex: linking reward to hedonic experience. Nat Rev Neurosci. 2005; 6(9):691–702. PMID: 16136173
83. Kringelbach ML, Rolls ET. The functional neuroanatomy of the human orbitofrontal cortex: evidence from neuroimaging and neuropsychology. Prog Neurobiol. 2004; 72(5):341–72. PMID: 15157726
84. London ED, Ernst M, Grant S, Bonson K, Weinstein A. Orbitofrontal cortex and human drug abuse: functional imaging. Cereb Cortex. 2000; 10(3):334–42. PMID: 10731228
85. Parsons CE, Stark EA, Young KS, Stein A, Kringelbach ML. Understanding the human parental brain: a critical role of the orbitofrontal cortex. Soc Neurosci. 2013; 8(6):525–43. doi: 10.1080/17470919.2013.842610 PMID: 24171901
86. Ridderinkhof KR, Ullsperger M, Crone EA, Nieuwenhuis S. The role of the medial frontal cortex in cognitive control. Science. 2004; 306(5695):443–7. PMID: 15486290
87. Whitfield-Gabrieli S, Ford JM. Default mode network activity and connectivity in psychopathology. Annu Rev Clin Psychol. 2012; 8:49–76. doi: 10.1146/annurev-clinpsy-032511-143049 PMID: 22224834
88. Andrews-Hanna JR, Reidler JS, Sepulcre J, Poulin R, Buckner RL. Functional-anatomic fractionation of the brain's default network. Neuron. 2010; 65(4):550–62. doi: 10.1016/j.neuron.2010.02.005 PMID: 20188659
89. Fair DA, Cohen AL, Power JD, Dosenbach NU, Church JA, Miezin FM, et al. Functional brain networks develop from a "local to distributed" organization. PLoS Comput Biol. 2009; 5(5):e1000381. doi: 10.1371/journal.pcbi.1000381 PMID: 19412534
90. Schilling C, Kühn S, Romanowski A, Schubert F, Kathmann N, Gallinat J. Cortical thickness correlates with impulsiveness in healthy adults. Neuroimage. 2012; 59(1):824–30. doi: 10.1016/j.neuroimage.2011.07.058 PMID: 21827861
91. Franklin TR, Acton PD, Maldjian JA, Gray JD, Croft JR, Dackis CA, et al. Decreased gray matter concentration in the insular, orbitofrontal, cingulate, and temporal cortices of cocaine patients. Biol Psychiatry. 2002; 51(2):134–42. PMID: 11822992
92. Matochik JA, London ED, Eldreth DA, Cadet JL, Bolla KI. Frontal cortical tissue composition in abstinent cocaine abusers: a magnetic resonance imaging study. Neuroimage. 2003; 19(3):1095–102. PMID: 12880835
93. Roberts GM, Garavan H. Evidence of increased activation underlying cognitive control in ecstasy and cannabis users. Neuroimage. 2010; 52(2):429–35. doi: 10.1016/j.neuroimage.2010.04.192 PMID: 20417713

94. Sheline YI, Barch DM, Price JL, Rundle MM, Vaishnavi SN, Snyder AZ, et al. The default mode network and self-referential processes in depression. Proceedings of the National Academy of Sciences. 2009; 106(6):1942–7.

95. Zhu X, Wang X, Xiao J, Liao J, Zhong M, Wang W, et al. Evidence of a dissociation pattern in resting-state default mode network connectivity in first-episode, treatment-naive major depression patients. Biological psychiatry. 2012; 71(7):611–7. doi: 10.1016/j.biopsych.2011.10.035 PMID: 22177602

96. Pope AW, Bierman KL. Predicting adolescent peer problems and antisocial activities: the relative roles of aggression and dysregulation. Dev Psychol. 1999; 35(2):335–46. PMID: 10082005

97. Krueger RF, Markon KE, Patrick CJ, Benning SD, Kramer MD. Linking antisocial behavior, substance use, and personality: an integrative quantitative model of the adult externalizing spectrum. J Abnorm Psychol. 2007; 116(4):645–66. PMID: 18020714

98. Thompson LL, Claus ED, Mikulich-Gilbertson SK, Banich MT, Crowley T, Krmpotich T, et al. Negative reinforcement learning is affected in substance dependence. Drug Alcohol Depend. 2012; 123(1–3):84–90. doi: 10.1016/j.drugalcdep.2011.10.017 PMID: 22079143

99. Mutlu AK, Schneider M, Debbané M, Badoud D, Eliez S, Schaer M. Sex differences in thickness, and folding developments throughout the cortex. Neuroimage. 2013 Nov; 82:200–7. doi: 10.1016/j.neuroimage.2013.05.076 PMID: 23721724

100. Wierenga LM, Langen M, Oranje B, Durston S. Unique developmental trajectories of cortical thickness and surface area. Neuroimage. 2014 Feb; 87:120–6. doi: 10.1016/j.neuroimage.2013.11.010 PMID: 24246495

101. Blakemore SJ, Choudhury S. Brain development during puberty: state of the science. Dev Sci. 2006; 9(1):11–4. PMID: 16445389

102. Blakemore SJ, Choudhury S. Development of the adolescent brain: implications for executive function and social cognition. J Child Psychol Psychiatry. 2006; 47(3–4):296–312. PMID: 16492261

103. Sowell ER, Thompson PM, Leonard CM, Welcome SE, Kan E, Toga AW. Longitudinal mapping of cortical thickness and brain growth in normal children. J Neurosci. 2004; 24(38):8223–31. PMID: 15385605

104. Wilde EA, Merkley TL, Bigler ED, Max JE, Schmidt AT, Ayoub KW, et al. Longitudinal changes in cortical thickness in children after traumatic brain injury and their relation to behavioral regulation and emotional control. Int J Dev Neurosci. 2012; 30(3):267–76. doi: 10.1016/j.ijdevneu.2012.01.003 PMID: 22266409

105. Doré V, Villemagne VL, Bourgeat P, Fripp J, Acosta O, Chetélat G, et al. Cross-sectional and longitudinal analysis of the relationship between Aβ deposition, cortical thickness, and memory in cognitively unimpaired individuals and in Alzheimer disease. JAMA Neurol. 2013; 70(7):903–11. doi: 10.1001/jamaneurol.2013.1062 PMID: 23712469

106. Hardan AY, Libove RA, Keshavan MS, Melhem NM, Minshew NJ. A preliminary longitudinal magnetic resonance imaging study of brain volume and cortical thickness in autism. Biol Psychiatry. 2009; 66(4):320–6. doi: 10.1016/j.biopsych.2009.04.024 PMID: 19520362

107. Verstraete E, Veldink JH, Hendrikse J, Schelhaas HJ, van den Heuvel MP, van den Berg LH. Structural MRI reveals cortical thinning in amyotrophic lateral sclerosis. J Neurol Neurosurg Psychiatry. 2012; 83(4):383–8. doi: 10.1136/jnnp-2011-300909 PMID: 21965521

108. Utevsky AV, Smith DV, Huettel SA. Precuneus is a functional core of the default-mode network. J Neurosci. 2014; 34(3):932–40. doi: 10.1523/JNEUROSCI.4227-13.2014 PMID: 24431451

109. Fransson P, Marrelec G. The precuneus/posterior cingulate cortex plays a pivotal role in the default mode network: Evidence from a partial correlation network analysis. Neuroimage. 2008; 42(3):1178–84. doi: 10.1016/j.neuroimage.2008.05.059 PMID: 18598773

110. Greicius MD, Flores BH, Menon V, Glover GH, Solvason HB, Kenna H, et al. Resting-state functional connectivity in major depression: abnormally increased contributions from subgenual cingulate cortex and thalamus. Biol Psychiatry. 2007; 62(5):429–37. PMID: 17210143

111. Pardoe HR, Abbott DF, Jackson GD, Initiative AsDN. Sample size estimates for well-powered cross-sectional cortical thickness studies. Hum Brain Mapp. 2013; 34(11):3000–9. doi: 10.1002/hbm.22120 PMID: 22807270

112. Krueger RF. The structure of common mental disorders. Arch Gen Psychiatry. 1999; 56(10):921–6. PMID: 10530634

113. Makris N, Gasic GP, Kennedy DN, Hodge SM, Kaiser JR, Lee MJ, et al. Cortical thickness abnormalities in cocaine addiction—a reflection of both drug use and a pre-existing disposition to drug abuse? Neuron. 2008; 60(1):174–88. doi: 10.1016/j.neuron.2008.08.011 PMID: 18940597

114. Thompson PM, Hayashi KM, Simon SL, Geaga JA, Hong MS, Sui Y, et al. Structural abnormalities in the brains of human subjects who use methamphetamine. J Neurosci. 2004; 24(26):6028–36. PMID: 15229250

115. Koester P, Tittgemeyer M, Wagner D, Becker B, Gouzoulis-Mayfrank E, Daumann J. Cortical thinning in amphetamine-type stimulant users. Neuroscience. 2012; 221:182–92. doi: 10.1016/j.neuroscience.2012.06.049 PMID: 22750208

116. Kühn S, Schubert F, Gallinat J. Reduced thickness of medial orbitofrontal cortex in smokers. Biol Psychiatry. 2010; 68(11):1061–5. doi: 10.1016/j.biopsych.2010.08.004 PMID: 20875635

117. Durazzo TC, Tosun D, Buckley S, Gazdzinski S, Mon A, Fryer SL, et al. Cortical thickness, surface area, and volume of the brain reward system in alcohol dependence: relationships to relapse and extended abstinence. Alcohol Clin Exp Res. 2011; 35(6):1187–200. doi: 10.1111/j.1530-0277.2011.01452.x PMID: 21410483

118. De Brito SA, Mechelli A, Wilke M, Laurens KR, Jones AP, Barker GJ, et al. Size matters: increased grey matter in boys with conduct problems and callous-unemotional traits. Brain. 2009; 132(Pt 4):843–52. doi: 10.1093/brain/awp011 PMID: 19293245