Brain cortical thickness in male adolescents with serious substance use and conduct problems

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Abstract

Background—Adolescents with substance use disorder (SUD) and conduct problems exhibit high levels of impulsivity and poor self-control. Limited work to date tests for brain cortical thickness differences in these youths.

Objectives—To investigate differences in cortical thickness between adolescents with substance use and conduct problems and controls.

Methods—We recruited 25 male adolescents with SUD, and 19 male adolescent controls, and completed structural 3T magnetic resonance brain imaging. Using the surface-based morphometry software FreeSurfer, we completed region-of-interest (ROI) analyses for group cortical thickness differences in left, and separately right, inferior frontal gyrus (IFG), orbitofrontal cortex (OFC) and insula. Using FreeSurfer, we completed whole-cerebrum analyses of group differences in cortical thickness.

Results—Versus controls, the SUD group showed no cortical thickness differences in ROI analyses. Controlling for age and IQ, no regions with cortical thickness differences were found using whole-cerebrum analyses (though secondary analyses co-varying IQ and whole-cerebrum cortical thickness yielded a between-group cortical thickness difference in the left posterior

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cingulate/precuneus). Secondary findings showed that the SUD group, relative to controls, demonstrated significantly less right>left asymmetry in IFG, had weaker insular-to-whole-cerebrum cortical thickness correlations, and showed a positive association between conduct disorder symptom count and cortical thickness in a superior temporal gyrus cluster.

**Conclusion**—Functional group differences may reflect a more nuanced cortical morphometric difference than ROI cortical thickness. Further investigation of morphometric differences is needed. If replicable findings can be established, they may aid in developing improved diagnostic or more targeted treatment approaches.

**Keywords**
substance use; adolescents; cortical thickness

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**INTRODUCTION**

**Youths with serious substance use and conduct problems**

Substance use disorders (SUD) commonly have their onset in adolescence (1, 2) and different substance use disorder diagnoses tend to cluster within individuals especially in younger males (3), or in youths whose problem behaviors are severe enough to merit referral for treatment (4, 5). For example, about three quarters of those admitted to adolescent-substance-use-disorder-treatment facilities require services for both alcohol and other drug issues (6). Substance use disorder carries with it great morbidity and risk for mortality, underscoring the importance of better understanding neural contributions to these disorders.

Adolescents affected by SUD are also very likely to meet criteria for conduct disorder (7, 8), which is characterized by aggression to people and animals, destruction of property, deceitfulness or theft, and serious rule violations (9, 10). These two disorders, conduct disorder and SUD, cluster together so often in our clinical populations, that we have termed such youth as having “antisocial substance dependence” in our previous publications (11, 12). Numerous longitudinal studies now support that an early predisposition toward behavioral under-control or disinhibition predicts a broad number of externalizing behaviors (13, 14, 15, 16). In part because of such observations, researchers have explored the covariance across externalizing behavior problems and have shown that a single factor, sometimes called “behavioral disinhibition”, may predispose individuals to externalizing psychopathology generally (17) and this single factor is highly heritable (18, 19).

Given this conceptualization, study designs typically choose a narrow or broad focus. On the one hand, selecting samples with a single externalizing disorder (i.e. cannabis use but not conduct disorder, other substance use disorder or antisocial personality disorder) allows researchers to importantly reduce some confounds (e.g. brain changes induced by substances other than cannabis), but this approach also necessitates recruiting subjects with relatively low “behavioral disinhibition”. Studying those individuals with a strong inherited general vulnerability to externalizing disorders requires recruitment of youths with high levels of diagnostic comorbidity. Here we focus on youths with severe antisocial behavior problems, multiple substance use disorder diagnoses and high impulsiveness, i.e. youths with serious substance use and conduct problems and high “behavioral disinhibition.”
Cortical Thickness

We have previously utilized this sample of adolescent males to examine SUD group-vs-control differences in grey matter volume using voxel-based morphometry (VBM) (12). Although grey matter volume is determined by the product of cortical thickness and surface area, examination of cortical thickness provides meaningful information beyond that obtained from grey-matter volume (as obtained in VBM analyses). First, the cortex is organized into ontogenic columns perpendicular to the brain surface (20), and the radial unit hypothesis supports that cells within such columns share a common origin (21, 22, 23). Thus, cortical thickness is related to the number of cells within a column, while surface area is related to the number of columns. Surface area and cortical thickness are both highly heritable, but are essentially determined by independent genetic effects (24, 25), and grey matter volume is more closely related to surface area (24, 26). Thus measuring both grey matter volume (12) and cortical thickness, as we have done here, provides complementary information.

Cortical thickness in frontal regions has been associated with impulsiveness in healthy adults (27) and differences in cortical thickness have been demonstrated among adult alcoholics in regions relevant to the brain reward circuit (28). Therefore, cortical thickness represents one logical brain phenotype in the search for brain structural differences in youths with serious substance use and conduct problems. Several studies have examined this phenotype, finding differences in cortical thickness between heavy marijuana using adolescents (29) or binge-drinking adolescents (30) and controls. Conduct disorder (31) and disruptive behavior disorders (32) have also been shown to be linked to cortical thickness differences in several regions. In more recent investigations, when controlling for (among other variables) intracranial volume (ICV) and age, cortical thickness differences were found between youths with documented externalizing behavior (33) or conduct disorder specifically (34) and controls.

Accordingly, we used brain magnetic resonance imaging (MRI) to study male youths with serious substance use and conduct problems, and controls, to search for inter-group differences in cortical thickness. Utilizing a male sample of 25 adolescents with SUD and conduct problems and 19 male adolescent controls, we tested for inter-group differences in cortical thickness. We hypothesized that youths with serious substance use and conduct problems would demonstrate thinner cortices in regions important for response inhibition and reward-related processing, namely in inferior frontal gyrus, OFC, and insula (35, 36, 37, 38). In addition, given the limited information on cortical thickness in this population of adolescents with serious substance use and conduct problems, we also completed whole-cerebrum analyses for group differences in cortical thickness.

METHODS

Sample selection and exclusion criteria

All subjects were right-handed males, between the ages of 14 and 18, and were required to score a minimum of 80 on a test of IQ. Participants were excluded if they had a history of head injury with loss of consciousness for more than 15 minutes, or history of significant
neurological illness or neurosurgery. All adolescent subjects and their parent/guardian had adequate English proficiency to understand the study procedures and provide informed assent/consent to research participation. By protocol, all subjects submitted a urine and saliva sample for on-site testing (AccuTest™ for THC, cocaine, methamphetamine, amphetamine, barbiturates, benzodiazepines, MDMA, methadone, other opioids, PCP and saliva AlcoScreen™ for alcohol) about 7 days and immediately prior to scanning. Positive results excluded controls from the study and the experimental group adolescents from participating at that time. A set of MRI-related exclusion criteria (such as the presence if implanted ferromagnetic objects) were also enforced for subject safety. Functional brain activation (11) and grey matter volume (12) of these subjects, or a subset of them, have been reported previously. Procedures and a complete explanation of inclusion and exclusion criteria are described in our previous publication (12).

Adolescent SUD sample—Adolescents with serious substance use and conduct problems (n=25; the experimental group) were recruited from a university-based treatment program that serves such youths. These experimental group adolescents were commonly referred from probation and social service agencies. Subjects were required to meet at least one non-nicotine substance use disorder (by the Diagnostic Statistical Manual, or DSM-IV criteria; 9) and to have been referred to treatment for serious conduct problems.

Control sample—Control adolescents (n=19) were recruited through a research marketing company and local advertisements. Recruitment ensured that controls resided in the same zip codes that experimental group adolescents commonly come from, and that our control sample was similar to these experimental group adolescents in age and race/ethnicity. Controls were excluded if they met criteria for conduct disorder, any non-nicotine substance use disorder (according to DSM-IV criteria) or if they had a prior court conviction or substance use-related sequelae (e.g., arrest, treatment, or school expulsion).

Assessments

All youths completed:

(1) *Youth Self Report* which produces dimensional ratings of conduct, attention, and affective problems, with excellent reliability and validity (39);

(2) *National Institute of Mental Health (NIMH) Diagnostic Interview Schedule for Children-Version IV* (DISC-IV; 40), a fully-structured computer-assisted diagnostic interview for youths. Youth-only reports of conduct-disorder symptoms from this instrument have excellent discriminative validity (7);

(3) *Composite International Diagnostic Interview-Substance Abuse Module* (41) a structured, computerized interview which provides valid (42) diagnoses of adolescent (7) DSM-IV-defined substance abuse and dependence diagnoses;
Wechsler Abbreviated Scale of Intelligence (WASI; Psychological Corporation) (43). We estimated full scale IQ from the Vocabulary and Matrix Reasoning subtests. Subjects with estimated IQ < 80 were excluded.

Eysenck Junior Impulsiveness Scale (44); and

a measure of Peak Aggressive Behavior (45).

Parents/guardians completed:

a self-reported race/ethnicity questionnaire; and

Child Behavior Checklist (46); this parent-report assessment (CBCL) is standardized for ages 4–18 and provides dimensional ratings of conduct, attention, and affective problems in children.

Magnetic Resonance Imaging

High-resolution 3D T1-weighted coronal slices were acquired in a research-dedicated 3 Tesla MR scanner (General Electric) using a standard quadrature head coil, an SPGR-IR sequence and the following parameters: TR/TE/T1/flip angle = 9 ms/1.9 ms/500 ms/10°, FOV = 220 mm\(^2\) in plane, slice thickness = 1.7 mm, 256\(^2\) matrix, and number of slices = 124. Structural acquisition took 9 minutes and 12 seconds.

Measuring Cortical Thickness

We utilized FreeSurfer v4.5, an automated morphometric program (47, 48, 49). Steps included normalization for intensity, resampling into isotropic voxels, skull stripping, and segmentation. Pial and grey-white matter surfaces were tessellated. The distance between these two surfaces estimates cortical thickness. Images were then normalized to a standard spherical space. Skull stripping and segmentation were verified through visual inspection by a study investigator blinded to group status. Errors due to topological defects, or skull-stripping were corrected using methods available in the FreeSurfer toolbox such as introducing controls points for white-matter mis-registration, pial editing for obviously erroneous inclusion of skull or dura or the use of the water-shed algorithm to improve skull-stripping. Individual subject data was registered into standard space using the FreeSurfer tool “FSAverage brain.”

Data Analyses

We evaluated the distributions of variables for outliers and for normality, when appropriate. Descriptive characteristics, including age, race, IQ, years of schooling completed, nicotine dependence diagnosis, SUMDEP (the sum of DSM-IV dependence symptoms across drug categories), lifetime conduct disorder symptom count, and impulsivity, peak aggression, and aggressive behavior scores, were compared between groups with independent t-tests (or Mann Whitney U test when appropriate) and chi-square tests. We employed two approaches: region of interest (ROI) and whole-cerebrum analyses to test for group differences in cortical thickness. The two approaches are complementary in that the ROI approach tests for group differences in a priori predicted regions and is sensitive to small magnitude differences over
larger well-defined areas. In contrast, whole-cerebrum analyses can identify large-magnitude group differences in small regions anywhere in the cerebrum.

**Region of Interest Analyses and Covariate Selection**—We used FreeSurfer automatically-generated parcellation-units (50) for ROI analyses. The FreeSurfer output divides the inferior frontal gyrus into three regions (pars orbitalis, triangularis and opercularis) and OFC into two regions (medial and lateral). Mean cortical thickness for each parcellation-unit (or ROI as a whole if it contained only a single parcellation unit) were obtained using FreeSurfer’s built in cortical thickness tools. Average cortical thicknesses for ROIs with multiple parcellation units were then calculated by the average of said parcellation units, weighted by the surface area as follows:

\[
L_{\text{OFC CT}} = \frac{[(L_{\text{med OFC CT}})(L_{\text{med OFC SA}})] + [(L_{\text{lat OFC CT}})(L_{\text{lat OFC SA}})]}{[(L_{\text{med OFC SA}}) + (L_{\text{lat OFC SA}})]}
\]

Eq. 1

where CT=cortical thickness; L=left; med=medial, lat=lateral, OFC=orbitofrontal cortex; SA=surface area.

Given that IQ is related to cortical thickness (51), as well as the very strong data supporting the importance of age on cortical thickness in adolescence (52), we included IQ and exact age (date of assessment minus date of birth) as covariates in all of our regression analyses. In preparation for completing multiple regression analyses, we investigated, across-all subjects and within-group, the relationship (Pearson correlations) between these two regression covariates, as well as whole-cerebrum average cortical thickness and ICV, and our six regions of interest (bilateral inferior frontal gyrus, OFC and insula). To evaluate potential group differences in correlations between whole-cerebrum cortical thickness and the 6 regions of interest, the Pearson correlations estimates (r’s) for each group were converted via Fisher’s Z-transformation and then compared with a z test using a standard error based on the square root of their summed variances.

Separate multiple regression analyses were completed for each of our six regions of interest (dependent variables) with group as our independent variable of interest, covarying age and IQ. Because several other reports covary whole-cerebrum average cortical thickness (53) or ICV (33, 34), these covariates were separately added to a subsequent set of exploratory models. These QDEC (“Query, Design, Estimate, Contrast;” the Free Surfer tool used for these analyses) analyses failed, likely due to multi-collinearity, possibly due to inclusion of age. Because groups did not differ significantly on age, and because we assumed that developmental differences in the small age window (14–18 years) would be captured by average cortical thickness to a limited extent, we conducted a subsequent set of exploratory models that covaried for IQ and whole-brain cortical thickness but not age.
Also, because some prior work has suggested that the link between antisocial behavior problems and low IQ is explained by shared genetic influences (54), controlling for IQ could limit our ability to find inter-group differences. Therefore, we completed exploratory analyses without including IQ as a covariate in the model.

**Whole-cerebrum analyses**—Between-group, whole-cerebrum, vertex-by-vertex analyses were completed using the FreeSurfer tool QDEC while, again, controlling for age and IQ. Family-wise error correction at cluster level (p<0.05, 2-tailed) was applied to correct for multiple comparisons, and the corresponding threshold cluster size was determined by Monte Carlo simulation (10,000 iterations) with a cluster-forming threshold vertex-level p-value of 0.005 (55).

**Exploratory analyses**—We completed exploratory regression analyses within the experimental group, testing for associations between (1) impulsivity measured by the Eysenck Junior Impulsiveness Scale, (2) lifetime conduct disorder symptom count and (3) across-drug substance use disorder symptom count and cortical thickness, enforcing the same threshold as in our whole-cerebrum analyses.

### RESULTS

#### Sample description

Adolescents with serious substance use and conduct problems, as well as control group adolescents, did not significantly differ in age (experimental group mean 16.64 years; control mean 16.59 years) but groups differed significantly in estimated IQ (experimental group mean 98.1; control mean 105.2; p=0.01; see Table 1). Adolescents with serious substance use and conduct problems also had significantly higher lifetime cross-drug substance dependence symptom counts and conduct disorder symptom counts, and scored significantly higher on measures of impulsivity and aggression. Although not shown in Table 1, 84% of experimental group adolescents met criteria for cannabis abuse or dependence, 84% for alcohol abuse or dependence, 36% for club drug abuse or dependence, 32% for cocaine abuse or dependence, and 20% for hallucinogen abuse or dependence. Across 10 drug categories, excluding nicotine, the experimental group adolescents averaged 2.7 lifetime substance use disorder diagnoses (standard deviation = 1.4). As this was an exclusion criterion, no control individuals met lifetime criteria for a substance use disorders for these 10 categories.

#### Initial Cortical Thickness analyses

Tables 2a and 2b show initial analyses completed in preparation for regression analyses. Table 2a shows mean cortical thickness for left and right hemisphere and for our six regions of interest (left and right inferior frontal gyrus (IFG), OFC, and insula), as well as the computed left-right asymmetry. Asymmetry was computed as described by Shaw et al. (56), where asymmetry = (LCT-RCT)/(0.5*(LCT+RCT)), where LCT and RCT denote cortical thicknesses of left or right ROI sides, respectively. The asymmetry calculation results suggested right>left cortical thickness asymmetry in the IFG of control adolescents, which has been demonstrated in several samples of normally developing adolescents (see Figure 2.
in Shaw et al. (56) and Table 2 in (57)) and young adults (58). However, this IFG asymmetry was not noted in adolescents with serious substance use and conduct problems, which is in line with the literature that finds disruption of frontal cortical thickness differences in adolescents with psychiatric disorders including ADHD (56) or PTSD (59). We compared the calculated IFG asymmetry values between experimental group adolescents and controls, which yielded a significant difference (t_{42}=-2.1; p=0.04). Although not part of our planned analyses, these results suggest that adolescent normative (right>left) cortical thickness asymmetry in IFG may not be present in male adolescents with serious substance use and conduct problems and high behavioral disinhibition. No significant inter-group asymmetry differences were found in the OFC (t_{42}=-0.6; p=0.58) or insula (t_{42}=0.3; p=0.74).

**Pearson correlations across and within groups**

Table 2b presents Pearson correlations between the cortical thickness of our six regions of interest and covariates in our regression analyses (age and IQ), as well as average whole-cerebrum cortical thickness and ICV (covariates used only in exploratory analyses). In this sample’s small age window (14–18 years), age was not significantly associated with cortical thickness in any of our regions of interest within-experimental group, within-controls or across-both-groups. IQ and ICV also did not significantly correlate with any of our regions of interest across groups. Also, when considering all subjects (i.e., the adolescents with serious substance use and conduct problems and controls), average cortical thickness across the entire brain was modestly and negatively correlated with age, positively correlated with all six regions of interest but not related to IQ. However, when considering experimental group adolescents and controls separately, group differences in magnitude of correlations between average cortical thickness across the whole cerebrum and our six regions of interest were provocative enough to warrant a further, more concrete investigation.

Significance values from comparing groups’ correlations between whole-cerebrum cortical thickness and the 6 regions of interest after Fisher’s Z transformation are presented in the last row of Table 2b. Several regions demonstrated between-group correlation differences with whole-cerebrum cortical thickness, including the right IFG, bilateral OFC, and the right insula. Again, although not part of our original hypotheses, these findings suggest that controls have a more cohesive pattern of cortical thickness across the whole cerebrum, and that adolescents with serious substance use and conduct problems have greater region-to-region variability.

**Region of Interest Analyses**

No ROI showed significant inter-group cortical thickness difference in our regression analyses when covarying for age and IQ (see Table 3). After separately adding whole-cerebrum cortical thickness and ICV as exploratory covariates in two additional sets of analyses, we demonstrated no significant group difference for our six ROIs. In our exploratory analyses that covaried age but not IQ, no ROI showed significant inter-group differences.
**Whole-cerebrum Analyses**

QDEC analyses were run covarying age and IQ. At the *a priori* whole-cerebrum threshold a cluster of 1,009 contiguous vertexes (392.64 mm$^2$), no significant inter-group cortical thickness differences were found.

In subsequent exploratory analyses that repeated the QDEC while covarying IQ, age, and whole-cerebrum cortical thickness, QDEC analyses failed, likely due to multi-collinearity, possibly from including age. Subsequent analyses covarying IQ and whole-cerebrum cortical thickness, but not age, did not fail, and found thinner cortical thickness in adolescents with serious substance use and conduct problems compared to controls mainly in the left posterior cingulate extending into the precuneus (Figure 1, panel A). Controls showed no significant region with thinner cortex when compared to experimental group adolescents at the set statistical threshold. QDEC analyses covarying age, IQ and ICV failed to demonstrate any significant group differences. Our exploratory analyses that covaried age but not IQ, also failed to demonstrate significant group differences.

**Exploratory Analyses**

Exploratory regression analyses within experimental group adolescents, testing for associations between (1) impulsivity, (2) conduct disorder symptom count and (3) substance use disorder symptom count and cortical thickness, yielded one significant result, showing a positive association between cortical thickness in the superior temporal gyrus and conduct disorder symptom count (see Figure 1, panel B).

**DISCUSSION**

**Main study findings**

Imaging a male adolescent sample of adolescents with serious substance use and conduct problems and controls, with or without controlling for whole-cerebrum cortical thickness or ICV, we found no significant differences in cortical thickness using region of interest analysis. In the whole-cerebrum QDEC analyses, covarying age and IQ (with or without ICV), we also did not find any significant differences. When controlling for whole-cerebrum cortical thickness in the QDEC, we demonstrated significantly greater cortical thickness in controls than experimental group adolescents in a relatively large cluster in the posterior cingulate cortex extending into the precuneus. In exploratory analyses we found one region, superior temporal gyrus, which was positively associated with conduct disorder symptom count.

Even with the positive QDEC result in the posterior cingulate cortex and the association between conduct disorder symptom count and superior temporal gyrus cortical thickness, our findings in this study are certainly weak, and primarily negative. They contrast with the previously cited cortical thickness literature on similar phenotypes (32, 29, 31, 30). Our findings also contrast with past behavioral and functional studies. Strong evidence supports the important role of ventrolateral prefrontal cortex, including the inferior frontal gyrus (IFG), in response inhibition (see Chikazoe (60) for a review), and this region along with anterior insula has been hypothesized to be critically important for guiding behavior in...
relation to risks and rewards, especially in situations with low predictability (61) and for generating emotional empathy (62). Although IFG has been implicated generally in response inhibition, there is hemispheric asymmetry with the right ventrolateral prefrontal cortex (including inferior frontal gyrus) appearing to be critical to response inhibition (60). The posterior cingulate cortex has been implicated in past studies of addiction (63), in paradigms involving moral decision-making (64), in self-appraisal and self-reflection (64), theory-of-mind (65), and in reward-related decision-making (66).

There are several explanations for why our findings contrast with this existing functional and behavioral work. First, it is of note that several past studies have covaried ICV (33, 34) or whole-cerebrum average cortical thickness (53). One conclusion that could be drawn from this incongruence of our findings to these studies is that, as opposed to experimental group adolescents having absolutely thinner cortices in the regions of interest, the inter-group difference lies in the ratio of the regional cortical thickness to whole-brain measures. However, we found cortical thickness differences in posterior cingulate when controlling for whole-cerebrum cortical thickness, but found no inter-group differences when controlling for ICV. The second and indeed simplest explanation is that the macroscopic structure of experimental group adolescents versus control ROIs is similar. The difference may be microscopic (at the cellular, synapse, or receptor level), affecting how the regions of interest function on their own, or in a network with other behaviorally important structures. However, this explanation would fail to account for the previously discussed positive cortical thickness findings. Thirdly, although we utilized a sample similarly sized to some past studies (29, 31, 34), we may have lacked adequate power (67). Lastly, between-study differences may explain our negative findings. Between-study differences included: varying phenotypes of focus (e.g. conduct disorder-only, binge drinking, callous-unemotional traits, “externalizing behavior,” or, here, serious substance use and conduct problems), various approaches to threshold selection, and differences in sample age ranges, or inclusion of mixed-sex samples (as opposed to our male-only sample). A male-only sample might be expected to reduce confounds, given that sex-differences have been demonstrated in brain imaging (68, 30) and genetic studies (69) of externalizing youths and in studies of normative brain development (70, 71).

We selected cases for high behavioral disinhibition and controls without such behavioral issues for case-control comparisons on cortical thickness. Because comorbidity is so common in these adolescent populations, researchers must choose a “narrow” versus “broad” approaches to sample selection (see Introduction, Section 1.1). Here we employ a “broad” approach, which allowed us to recruit a clinically representative sample with high behavioral disinhibition, but includes co-morbidity and reduces the ease of interpretability of findings (e.g., a broad approach cannot with certainty identify that this cortical thickness finding is related to conduct disorder but not substance use disorders). While this broad approach has yielded differences in brain structure and function in our prior publications (e.g. 11, 12, 72, 73), we did not demonstrate strong cortical thickness differences here. Findings using the broad approach to experimental group member selection may not be specific to one disorder vs. another, but instead may be indicative of non-specific neurodevelopmental problems. There are certainly examples in the literature of such indicators of neurodevelopmental problems (e.g. large cavum septum pellucidum; 74–76). In contrast, narrow approaches...
might examine disorder specific brain findings or domain specific brain correlates (e.g. National Institute of Mental Health Research Domain Criteria, or RDoC). Certainly, the successes using this narrow approach have been previously reviewed and specific network-to-disorder models have been proposed (e.g., 77). The disadvantages of the narrow approach include that selecting samples for one disorder but no others (when co-morbidity is commonplace), may result in atypical, less severely affected samples (17). Thus there are advantages and tradeoffs with each design and they might be viewed as complementary, but it is important to recognize that our broad approach may obscure easy interpretation of results and our secondary findings should be viewed through this lens. Future studies should focus on investigating the relationship between “broad” and “narrow” phenotypes and cortical thickness, and might utilize the NIMH Research Domain Criteria such as the “Response Selection, Inhibition or Suppression” subconstruct (domain: “Cognitive Systems,” construct: “Cognitive Control”).

Secondary findings

Although not part of our hypotheses, our analyses yielded two secondary findings. First, we demonstrate that adolescents with serious substance use and conduct problems have very little left-right asymmetry in inferior frontal gyrus cortical thickness. One relatively large sample of normally developing children suggests that asymmetry of inferior frontal gyrus thickness is normative and that in our sample age range, we would expect right>left cortical thickness with a mean difference of ~0.05–0.10 mm (see Figure 2 in Shaw et al. (56)). Our results in control adolescents are remarkably consistent with this. However, this IFG asymmetry is not present in our experimental group sample. According to Shaw and colleagues, this lack of asymmetry does occur in normal individuals around age ~11 years, but disappears thereafter (56). Meanwhile, others have shown that the effacement of R>L frontal volume differences in psychiatric disorders (56, 59). In addition and more generally, the development of verbal fluency is associated with thinning of the left inferior frontal gyrus (78) and that early measurement of language skills are predictive of the development of concern and disregard for others (79). In rhesus monkeys, too, a link between cortical asymmetry effacement and reactive and aggressive behavior has been demonstrated (80). However, beyond these studies, few exist looking at the effect of patient/control status (in the context of any psychiatric disorder) on frontal asymmetry. Our findings add to this limited pool of data, and suggest an opportunity for further investigation of inferior frontal asymmetry as a marker of the development of empathic concern and self-control/inhibition.

Second, experimental group adolescents and controls differed dramatically in the correlation between insula cortical thickness and average cortical thickness across the whole cerebrum. Structural covariance may signal functional connectivity and “synchronized maturation” between brain regions (81) and our method (region of interest correlation with whole-cerebrum average cortical thickness) approximates the strength of MACACC (Mapping Anatomical Correlations Across Cerebral Cortex), a statistical technique that uses whole-cortical morphometric data to examine the interrelationships between brain structures (82). Some evidence suggests that the correlation of regional cortical thickness provides information about functional connectivity (82), at least for positively correlated areas (83). Among normally developing children insula shows at least moderate MACACC strength.
scores (see Figure 3 in Lerch et al. (82)), consistent with our control sample results. Thus, although our approximate MACACC strength estimates were conducted only in preparation for our planned region of interest regression analyses of group differences, they raise the possibility that the near zero insula-to-whole-cerebrum cortical thickness correlations seen in our experimental sample may indicate relatively poor connectivity.

**Study limitations**

It is important to view this work within the context of the study’s limitations. First, as previously discussed, our study may have lacked adequate power to establish significance of inter-group cortical thickness differences. Thus, future studies with larger sample sizes may be required. Second, it should be noted that our experimental and control groups had different mean IQs. While we included IQ as a covariate in an attempt to filter out its effect on our findings, ideally experimental and control groups without IQ differences might also be studied. Such samples would allow the ability to rule out that observed group differences were not driven by IQ differences. As such, it may be fruitful to replicate our investigation in a larger sample that allows for an IQ-matched subsample. Third, it should be noted that while our ROI selection was rational and based on the existing literature, it was by no means absolutely inclusive. ROIs that may have had significant inter-group differences could have been missed. This possibility cannot be discounted by citing the negative QDEC findings, since exploratory analyses covarying whole-cerebrum cortical thickness did in fact find significant differences in the left posterior cingulate and precuneus (neither of which were selected as initial ROIs). Follow-up studies may find it fruitful to include these regions in future ROI analyses.

**CONCLUSIONS**

We attempted to characterize an empirical, quantifiable difference between control individuals and male youths with serious substance use and conduct problems. If successful, such findings could potentially be used in diagnosis and treatment of this at-risk population, or to further our understanding of the underpinnings of this phenotype. Using both ROI and whole-brain analysis, we failed to demonstrate inter-group differences in brain cortical thickness, or any such differences within the experimental group. The incongruence between our findings and those of previous studies merits further investigation of the nuances in cortical morphometric features of adolescents with substance use and conduct problems. However, our significant exploratory QDEC findings in the posterior cingulate cortex and precuneus (when controlling for whole-brain cortical thickness) and in superior temporal gyrus (exploring associations with conduct disorder symptom count), as well as our significant secondary findings (of limited asymmetry in experimental group adolescents’ IFGs, as well as group differences in the correlations of insula cortical thickness vs whole-brain cortical thickness) do hint at potential empirical inter-group differences we sought, and certainly merit further study.

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Panel A. Whole-cerebrum Analyses in QDEC (controls > experimental group adolescents; n=44, degrees of freedom=38), controlling for IQ and average cortical thickness across the whole cerebrum, using cluster-wise family-wise error correction of p < 0.05 (going in p-
value 0.005 at vertex-level with 10,000 Monte Carlo simulations); one cluster of 1,009 vertexes, maximum vertex at Talaraich x, y, z of −5.6, −44.2, 30.0. Color scale (horizontal bar, lower right of figure) indicates t-value, where t=2.5 required for vertex-level threshold). Panel B. Within SUD group adolescents, showing a positive association between lifetime conduct disorder symptom count and cortical thickness in superior temporal gyrus of the right hemisphere (same threshold enforced; cluster size is 679 vertexes).
### Table 1

Values presented are the number of participants of the given group that fit the given criterion, or mean with parenthetic standard deviation; MW = Mann-Whitney U Test; FE = Fisher’s Exact Test; SUMDEP the sum of DSM-IV dependence symptoms across drug categories.

<table>
<thead>
<tr>
<th>Demographics:</th>
<th>Experimental group (n=25)</th>
<th>Controls (n=19)</th>
<th>Statistic; p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>16.6 (1.15) yrs</td>
<td>16.6 (1.62) yrs</td>
<td>t sub {sub}31.13=−0.13; p=0.90</td>
</tr>
<tr>
<td>Race: White</td>
<td>16</td>
<td>15</td>
<td>( \chi^2=1.16; p=0.28 )</td>
</tr>
<tr>
<td>Race: All others</td>
<td>9</td>
<td>4</td>
<td>( \chi^2=2.69; p=0.01 )</td>
</tr>
<tr>
<td>Estimated IQ</td>
<td>98.1 (1.68)</td>
<td>105.2 (2.08)</td>
<td>( t_{42}=1.02; p=0.32 )</td>
</tr>
<tr>
<td>Years of schooling completed</td>
<td>9.36 (1.15)</td>
<td>9.84 (1.80)</td>
<td>( t_{58.79}=10.9; p&lt;0.001 )</td>
</tr>
<tr>
<td>Nicotine dependence diagnosis</td>
<td>13</td>
<td>1</td>
<td>( \chi^2=10.9; p=0.001 )</td>
</tr>
<tr>
<td>SUMDEP</td>
<td>11.9 (7.05)</td>
<td>0.2 (0.69)</td>
<td>MW; p&lt;0.001</td>
</tr>
<tr>
<td>Conduct disorder symptom count (lifetime)</td>
<td>6.4 (2.83)</td>
<td>0.4 (0.61)</td>
<td>( t_{26.60}=−10.33; p&lt;0.001 )</td>
</tr>
<tr>
<td>Eysenck Impulsivity scale</td>
<td>12.8 (6.13)</td>
<td>7.0 (4.53)</td>
<td>( t_{41.96}=−3.62; p=0.001 )</td>
</tr>
<tr>
<td>Peak Aggression</td>
<td>5.7 (3.11)</td>
<td>0.4 (1.01)</td>
<td>MW; p&lt;0.001</td>
</tr>
<tr>
<td>YSR Aggressive behavior t-score</td>
<td>61.6 (10.65)</td>
<td>52.6 (5.22)</td>
<td>( t_{36.70}=−3.71; p=0.001 )</td>
</tr>
</tbody>
</table>
Table 2a

Mean and standard deviation within SUD group adolescents and controls of average cortical thickness (both left and right hemispheres and of the regions of interest) (in mm) and intracranial volume (ICV, in cm³). Number of controls = 19, number of experimental group adolescents = 25. Asymm. is asymmetry, computed as the mean of experimental group adolescents’ or controls’ differences in the ROI (or hemispheric) left and right cortical thicknesses, divided by the mean of the left and right ROI (or hemispheric) cortical thicknesses (see section 3.2).

<table>
<thead>
<tr>
<th></th>
<th>Hemispheric</th>
<th>Inferior Frontal Gyrus</th>
<th>Orbitofrontal cortex</th>
<th>Insula</th>
<th>ICV</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>L CT</td>
<td>R CT</td>
<td>asymm.</td>
<td>L CT</td>
<td>R CT</td>
</tr>
<tr>
<td>L CT</td>
<td>2.478 (0.10)</td>
<td>2.487 (0.11)</td>
<td>−0.003</td>
<td>2.657 (0.13)</td>
<td>2.754 (0.14)</td>
</tr>
<tr>
<td>R CT</td>
<td>2.478 (0.07)</td>
<td>2.477 (0.07)</td>
<td>0.000</td>
<td>2.690 (0.10)</td>
<td>2.701 (0.10)</td>
</tr>
</tbody>
</table>
Pearson correlations between several covariates and either other covariates or cortical thickness of several regions of interest are presented, as well as (in the last row) the significance of SUD adolescent-control difference in correlations between the given ROI’s cortical thickness and the whole cerebrum cortical thickness.

### Table 2b

<table>
<thead>
<tr>
<th></th>
<th>Age</th>
<th>IQ</th>
<th>LIFG</th>
<th>RIFG</th>
<th>LOFC</th>
<th>ROFC</th>
<th>LINS</th>
<th>RINS</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ALL SUBJECTS</strong> (n=44)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole-cerebrum CT</td>
<td>0.16</td>
<td>0.30*</td>
<td>−0.28</td>
<td>−0.03</td>
<td>−0.08</td>
<td>−0.06</td>
<td>0.18</td>
<td>0.19</td>
</tr>
<tr>
<td>Age</td>
<td>1</td>
<td>0.22</td>
<td>−0.24</td>
<td>−0.16</td>
<td>−0.07</td>
<td>−0.12</td>
<td>−0.10</td>
<td>0.02</td>
</tr>
<tr>
<td>Estimated IQ</td>
<td>0.22</td>
<td>1</td>
<td>−0.03</td>
<td>0.08</td>
<td>−0.04</td>
<td>0.03</td>
<td>0.14</td>
<td>0.22</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Age</th>
<th>IQ</th>
<th>LIFG</th>
<th>RIFG</th>
<th>LOFC</th>
<th>ROFC</th>
<th>LINS</th>
<th>RINS</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>WITHIN-CONTROLS</strong> (n=19)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole-cerebrum CT</td>
<td>0.16</td>
<td>0.20</td>
<td>−0.15</td>
<td>0.29</td>
<td>0.18</td>
<td>0.15</td>
<td>0.27</td>
<td>0.09</td>
</tr>
<tr>
<td>Age</td>
<td>1</td>
<td>0.24</td>
<td>−0.25</td>
<td>−0.33</td>
<td>−0.26</td>
<td>−0.22</td>
<td>−0.34</td>
<td>0.04</td>
</tr>
<tr>
<td>Estimated IQ</td>
<td>0.24</td>
<td>1</td>
<td>−0.12</td>
<td>0.21</td>
<td>0.22</td>
<td>0.18</td>
<td>−0.09</td>
<td>0.10</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Age</th>
<th>IQ</th>
<th>LIFG</th>
<th>RIFG</th>
<th>LOFC</th>
<th>ROFC</th>
<th>LINS</th>
<th>RINS</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>WITHIN-SUD GROUP</strong> (n=25)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole-cerebrum CT</td>
<td>0.17</td>
<td>0.34</td>
<td>−0.37</td>
<td>−0.40*</td>
<td>−0.26</td>
<td>−0.30</td>
<td>0.07</td>
<td>0.22</td>
</tr>
<tr>
<td>Age</td>
<td>1</td>
<td>0.25</td>
<td>−0.23</td>
<td>0.09</td>
<td>0.12</td>
<td>0.04</td>
<td>0.16</td>
<td>0.01</td>
</tr>
<tr>
<td>Estimated IQ</td>
<td>0.25</td>
<td>1</td>
<td>0.19</td>
<td>−0.23</td>
<td>−0.26</td>
<td>−0.23</td>
<td>0.14</td>
<td>0.18</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>ROI to whole-cerebrum CT correlations (SUD group vs. control)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.559</td>
<td>0.032*</td>
<td>0.037*</td>
<td>0.021*</td>
<td>0.092</td>
<td>0.039*</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: CT = cortical thickness; ICV = intracranial volume; LIFG = average cortical thickness of the left inferior frontal gyrus (pars opercularis, triangularis and orbitalis); LINS = average cortical thickness of the left insula; LOFC = average cortical thickness of the left OFC; RIFG = average cortical thickness of the right inferior frontal gyrus; RINS = average cortical thickness of the right insula; ROFC = average cortical thickness of the right OFC; Whole-cerebrum CT = Average cortical thickness across the entire cerebrum. An * indicates p<0.05; ** indicates p<0.01.
Table 3

Standardized regression coefficients (β) and associated p values of group (SUD group adolescent vs control) status or the model’s covariates (IQ and age) versus mean thickness of regions of interest (L and R delineate the left and right hemisphere’s given ROI). All values given are not corrected for multiple comparisons.

<table>
<thead>
<tr>
<th></th>
<th>LIFG</th>
<th>RIFG</th>
<th>LOFC</th>
<th>ROFC</th>
<th>LINS</th>
<th>RINS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>β=0.19 (p=0.26)</td>
<td>β=−0.20 (p=0.24)</td>
<td>β=−0.04 (p=0.81)</td>
<td>β=−0.10 (p=0.58)</td>
<td>β=−0.26 (p=0.13)</td>
<td>β=−0.19 (p=0.27)</td>
</tr>
<tr>
<td>IQ</td>
<td>β=0.10 (p=0.54)</td>
<td>β=0.04 (p=0.80)</td>
<td>β=−0.04 (p=0.82)</td>
<td>β=0.03 (p=0.89)</td>
<td>β=0.07 (p=0.70)</td>
<td>β=0.16 (p=0.37)</td>
</tr>
<tr>
<td>Age</td>
<td>β=−0.26 (p=0.10)</td>
<td>β=−0.17 (p=0.30)</td>
<td>β=−0.06 (p=0.37)</td>
<td>β=−0.13 (p=0.44)</td>
<td>β=−0.10 (p=0.51)</td>
<td>β=−0.01 (p=0.93)</td>
</tr>
</tbody>
</table>