Developmental Neuropsychology

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/hdvn20

Developmental Variation in Amygdala Volumes Among Children With Posttraumatic Stress

Carl F. Weems a, Brandon G. Scott a, Justin D. Russell a, Allan L. Reiss b & Victor G. Carrión b

a Department of Psychology, University of New Orleans, New Orleans, Louisiana
b School of Medicine, Stanford University, Stanford, California

Published online: 18 Oct 2013.

To cite this article: Carl F. Weems, Brandon G. Scott, Justin D. Russell, Allan L. Reiss & Victor G. Carrión (2013) Developmental Variation in Amygdala Volumes Among Children With Posttraumatic Stress, Developmental Neuropsychology, 38:7, 481-495, DOI: 10.1080/87565641.2013.820307

To link to this article: http://dx.doi.org/10.1080/87565641.2013.820307

PLEASE SCROLL DOWN FOR ARTICLE

Taylor & Francis makes every effort to ensure the accuracy of all the information (the “Content”) contained in the publications on our platform. However, Taylor & Francis, our agents, and our licensors make no representations or warranties whatsoever as to the accuracy, completeness, or suitability for any purpose of the Content. Any opinions and views expressed in this publication are the opinions and views of the authors, and are not the views of or endorsed by Taylor & Francis. The accuracy of the Content should not be relied upon and should be independently verified with primary sources of information. Taylor and Francis shall not be liable for any losses, actions, claims, proceedings, demands, costs, expenses, damages, and other liabilities whatsoever or howsoever caused arising directly or indirectly in connection with, in relation to or arising out of the use of the Content.

This article may be used for research, teaching, and private study purposes. Any substantial or systematic reproduction, redistribution, reselling, loan, sub-licensing, systematic supply, or distribution in any form to anyone is expressly forbidden. Terms & Conditions of access and use can be found at http://www.tandfonline.com/page/terms-and-conditions
Developmental Variation in Amygdala Volumes Among Children With Posttraumatic Stress

Carl F. Weems, Brandon G. Scott, and Justin D. Russell

Department of Psychology, University of New Orleans, New Orleans, Louisiana

Allan L. Reiss and Victor G. Carrión

School of Medicine, Stanford University, Stanford, California

This article examined associations between indices of maturation (age and Tanner stage) and amygdala volumes in 24 youth (aged 7–14) with posttraumatic stress disorder symptoms and a matched control group. Fifteen of the youth with exposure to trauma were also re-evaluated one year later. A positive association between maturation and right amygdala volumes was observed in the trauma group but not in controls. Associations with maturation remained when controlling for a number of possible covariates and over time. Developmentally younger youth (Tanner stage 1 and 2) showed increases and older (Tanner stage 3 and 4) decreases in right amygdala volumes.

The amygdala is a brain region of the anterior portion of the temporal lobes. Its main functions are thought to be involved in the evaluation of the emotional significance of incoming stimuli (Tottenham, 2011; Tottenham & Sheridan, 2010). The amygdala projects to several brain structures in the frontal cortex, the hippocampus, the striatum, the hypothalamus, and brain stem (see Gordon & Hen, 2004; LeDoux, 2000; Tottenham, 2011; Tottenham & Sheridan, 2010). Because of its structural connections and functional roles, the amygdala is considered to have critical involvement in the behavioral activation and inhibition system that characterizes emotional responses such as fear and anxiety (Gray 1994; Gray & McNaughton, 2000). Over activation of the behavioral inhibition system is associated with excessive fear, anxiety, and negative emotionality and so the amygdala has received particular attention in biological theories of anxiety disorders and other disorders involving emotion (e.g., Davis, 1998; LeDoux, 2000).

Electroencephalography research has demonstrated increased anterior temporal region activation during the experience of negative emotion and fear (Davidson, 1998). Similar findings have emerged in anxious youth using functional magnetic resonance imaging (fMRI) techniques. For example, youth with posttraumatic stress disorder (PTSD) symptoms have shown faster right amygdala activation in response to angry faces than age and gender matched controls (Garrett et al., 2012, see also Tottenham & Sheridan, 2010). In terms of structural findings, volumetric imaging work in pediatric patient populations, such as youth with autism, bipolar disorder, and...
PTSD symptoms has tended to focus on amygdala volume comparisons with age- and gender-matched comparison youth (Chen et al., 2004; Groen Teluji, Buitelaar, & Tenderk, 2010; DeBellis et al., 1999; Carrión et al., 2001; Carrión, Weems, Richert, Hofman, & Reiss, 2010). However, studies examining volumetric differences of the amygdala have often produced highly inconsistent findings. For example, Morey et al. (2012) reviewed 12 studies on amygdala volumes among individuals (9 adult samples and 3 youth samples) with posttraumatic stress and examination of effect sizes revealed four studies indicating relatively smaller volumes (in both sides); one relatively smaller in the left, but larger in the right; one study relatively smaller in the right, larger in left; and six relatively larger in both sides (but only 1 of the 12 reported $p$-values reaching the typical .05 alpha). An important possible reason for inconsistencies (especially among youth samples) is developmental variation in amygdala volumes.

Structurally, there is evidence to suggest that the amygdala may continue to develop until late childhood. Table 1 summarizes nine studies we found in a literature search (e.g., Medline, PsycINFO, Google Scholar, & previous publications), that have examined normal developmental variation in amygdala volumes or otherwise correlated age with amygdala volumes in various patient and control samples. Six studies reported a statistically significant association with age in a community or control sample. However, the nature of the association is complex. In a widely cited initial study, the amygdala was observed to be related to age in youth 4–18 years old but only for male subjects and only in the left side (Giedd et al., 1996). Most recently, Uematsu et al. (2012) examined age’s association with amygdala volumes among 109 healthy participants aged from infancy to 25 years and found that a curvilinear association best defined the relationship. The nature of the association in Uematsu et al. (2012) has critical developmental implications for research on patient samples. The authors report a strong linear association from infancy to around age 11 years that leveled off or declined at that point (implying no correlation or a weak negative correlation from around 10 or 11 and beyond) and observed this association in both the right and left amygdala. The peak growth (end of a linear association in the scatterplots) was earlier in the left side generally and earlier in females (years 9 left – 11 right) than in males (years 11 left – 12 right). Theoretically, maturational variation in the amygdala may be altered by exposure to early life stress (Tottenham, 2011; Tottenham & Sheridan, 2010).

Tottenham and Sheridan (2010) have argued that adverse experiences can produce long-term changes in the amygdala structurally. In their view there is a stress-induced kindling of the amygdala where repeated stimulation produces greater future excitability and this implies potential differences in morphology. For example, among children adopted from orphanage care, Tottenham et al. (2009) reported a significant linear association between amygdala volumes and the time spent in the institution and the effect was observed when controlling for current age (volume correlation with age was not reported). In the full sample, amygdala volumes did not differ between institutionalized children and matched comparison children, but larger volumes were found among those who spent greater than 15 months in institutionalized care (Tottenham et al., 2009). The work of Tottenham and colleagues (Tottenham et al., 2009; Tottenham & Sheridan, 2010) coupled with the idea of maturational associations with amygdala volumes in school aged youth (Geidd et al., 1996; Uematsu et al., 2012) implies that an important approach to understanding group differences in the amygdala is clarifying developmental variation in amygdala volumes. This is highlighted by work in another patient population with potential for elevated stress. Chen et al. (2004; see Table 1) found a trend for smaller left amygdala volumes in adolescent bipolar patients versus matched control subjects (ages 10–21) but there was a positive
**TABLE 1**

Summary of Previous Research Linking Age/Maturation With Amygdala Volumes

<table>
<thead>
<tr>
<th>Study</th>
<th>N</th>
<th>Age Range</th>
<th>% Females</th>
<th>Sample Type</th>
<th>Salient Associations With Age</th>
<th>Laterality of Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Giedd et al., 1996</td>
<td>99</td>
<td>4.7–17.8</td>
<td>46</td>
<td>Healthy Community</td>
<td>Slope = .06, p = .01 (Males Only)</td>
<td>Left Only</td>
</tr>
<tr>
<td>Uematsu et al., 2012</td>
<td>109</td>
<td>Infants–25 years</td>
<td>48</td>
<td>Healthy Community</td>
<td>$R^2 = .48$ (Right Cubic Model), $R^2 = .60$ (Left Cubic Model)</td>
<td>Right and Left</td>
</tr>
<tr>
<td>Van der Plas et al., 2010</td>
<td>116</td>
<td>7–17</td>
<td>48</td>
<td>Healthy Community</td>
<td>$r = .36, p &lt; .05$ (Right; Males Only)</td>
<td>Right Only</td>
</tr>
<tr>
<td>Chen et al., 2004</td>
<td>37</td>
<td>10–21</td>
<td>50 (Bipolar)</td>
<td>43 (Control)</td>
<td>$r = .50, p=.047$ (Bipolar) $r = −.48, p =.03$ (Healthy Controls)</td>
<td>Left Only</td>
</tr>
<tr>
<td>Karchemskiy et al., 2011</td>
<td>44</td>
<td>Bipolar Offspring: $M = 12.3$ Controls: $M = 13.1$</td>
<td>Bipolar Offspring: 47 Children of parents with bipolar disorder &amp; Healthy Controls</td>
<td>“No correlation between age and brain volume in any region” ($r$ and $p$ value not reported; p. 322)</td>
<td>Right and Left</td>
<td></td>
</tr>
<tr>
<td>Keller et al., 2008</td>
<td>64</td>
<td>8–21</td>
<td>63 (MDD)</td>
<td>63 (MDD w/o psychosis) MDD w/o Psychosis, &amp; Healthy Controls</td>
<td>Smaller amygdala in young (&lt;35.5) MDD w/psychosis patients vs. healthy controls ($p = .001$). No difference between older MDD w/psychosis (&gt;35.5) and controls.</td>
<td>Right and Left</td>
</tr>
<tr>
<td>MacMaster et al., 2008</td>
<td>77</td>
<td>8–21</td>
<td>63 (MDD)</td>
<td>Psychotropic Naïve Patients w/MDD &amp; Healthy Controls</td>
<td>“No significant correlation for either group” ($r$ and $p$ value not reported; p. 389)</td>
<td>N/A</td>
</tr>
<tr>
<td>Mosconi et al., 2009</td>
<td>83</td>
<td>2 years old followed to 4 years old</td>
<td>14 (Autism)</td>
<td>27 (Typical) 9 (Delayed) Autism Disorder, Developmentally Delayed, &amp; Typical Healthy Controls</td>
<td>Volumes increased from age 2 to 4 in Autism and Control Group ($\beta = .14, p &lt;.001$)</td>
<td>Right and Left</td>
</tr>
<tr>
<td>Schumann et al., 2004</td>
<td>98</td>
<td>7.5–18.5</td>
<td>0 (All Male)</td>
<td>Low and High Functioning Autism, Asperger Disorder, Healthy Controls</td>
<td>$r = .67$ (Right), $r = .77$ (Left) both $p$’s less than .05 (Control Only)</td>
<td>Right and Left</td>
</tr>
</tbody>
</table>

*Note. MDD = Major Depressive Disorder.*
correlation between left amygdala volumes and age ($r = .50, p = .047$) in patients, whereas in healthy controls there was a negative correlation ($r = -.48, p = .03$). The negative correlation in the control sample might represent the tail end of the curvilinear trend observed in Uematsu et al. (2012) with the positive association possibly representing an atypical or prolonged growth period.

The recent theory implicating maturational differences (Tottenham, 2011; Tottenham & Sheridan, 2010) and research suggesting differential age relations in amygdala volumes (Chen et al., 2004) highlights the importance of testing the association between indices of maturation and amygdala volumes in youth exposed to traumatic stress. However, the extant research with age begs the question of the biological factors associated with amygdala growth (and/or the end of growth/pruning) and true change in volumes. Blakemore, Burnett, and Dahl (2010) have argued for the need to examine neural maturation in the context of pubertal development because chronological age only approximates important pubertal stages (e.g., variation in ages are seen at different Tanner stages of pubertal development) whereas the biological maturation seen in puberty more directly indexes a theoretical mechanism of neural development than chronological age. Previous research has not examined pubertal development and amygdala volumes and we found no studies examining change in amygdala volumes among school aged youth. We found only one study that examined longitudinal change (Table 1; Mosconi et al., 2009). These children were assessed at age two and again at age four years and results showed that volumes increased in each group (Autism and control groups).

In summary, this study aimed to examine the associations between indices of maturation (age and Tanner stage) and amygdala volumes among children ages 7–14 years with exposure to traumatic stress, re-evaluate the amygdala volumes one year later to provide replication of the initial findings, and examine change over time. The ages of 7–14 years represent a potentially critical transition period in amygdala growth. Drawing from Uematsu et al., (2012) this age range is the major arch in the curve from strong linear (infancy to ∼11 years) to flat or decreasing (ages 11–25) and often represents a period of relatively rapid changes in puberty (Marshall & Tanner, 1969, 1970). A positive association between maturation (both age and pubertal development) and amygdala volumes among youth exposed to traumatic stress (as well as controlling for total cerebral volumes and total right temporal volumes) and a less strong association among youth not exposed to stress was predicted. Theoretically, if stress is driving atypical maturation, controlling for stress symptoms would mediate (or partially mediate/reduce) the association between maturation and amygdala volumes. Thus, analyses examined the association between amygdala volumes and PTSD symptoms and controlled for PTSD as well as other potential covariates (e.g., total brain volumes). Finally, the youth with exposure to trauma were re-evaluated one year later to provide replication of the initial findings and examine change over time. Based on the extant non-longitudinal work (Uematsu et al., 2012), we predicted differential change based on Tanner stage.

**METHOD**

**Participants**

The sample of youth exposed to traumatic stress consisted of 14 boys and 10 girls. The mean age of the children was 10.96 years with a range of 7 to 14 years. All of the children in this sample
were referred to the project due to exposure to traumatic events. The clinical group was recruited from local departments of social services and mental health clinics and all of the subjects fulfilled the following criteria: (1) At least one episode of exposure to trauma, as defined by DSM-IV criterion A1; “the person experienced, witnessed, or was confronted with an event or events that involved actual or threatened death or serious injury, or a threat to the physical integrity of self or others” (American Psychiatric Association, 1994), (2) the trauma episode or episodes for which the individual is referred must have occurred at least 6 months prior to referral, and (3) a severity score of 12 or above on the PTSD Reaction Index. Exclusion criteria consisted of a PTSD Reaction Index severity score below 12; history of mental retardation; history of schizophrenia or autism, presence of any metal or electrical conductive implants or foreign bodies; current history of substance dependence and history of clinically significant head trauma, epilepsy, or other documented neurological disorder.

Most of the trauma-exposed sample (83%) experienced multiple traumatic events. Traumatic events included witnessing violence (50%), physical abuse (46%), separation and loss (38%), sexual abuse (21%), physical neglect (13%), emotional abuse (13%). In terms of family income, 43% reported incomes between 0 and $20,000, 28% reported incomes between $21,000 and $40,000, and 27% of the families reported incomes over $41,000. In terms of education, caregivers reported partial high school education (2.9%), a high-school education (17.6%), partial college (26.5%), college (20.6%) or graduate school education (20.6%). Ethnic composition was Euro-American (n = 14), African American (n = 6), Hispanic (n = 3), and Asian (n = 1). Children’s average pubic hair Tanner stage was 2.2 (range 1 to 4). WASI full scale IQs ranged from 65 to 142, average score 90, (only 3 subjects scored below 70 and these were included in the sample because they had no prior history of mental retardation and were able to participate in the clinical evaluation). The imaging data for a control group were obtained from an archived sample. Twenty-four healthy control subjects were individually age and gender matched to experimental subjects within 1.5 years of age. The mean age for the control group was 11.04; range 8 to 14 years of age.

Follow-up (Time 2; 12–18 months later) evaluation was conducted on 6 boys and 9 girls in the trauma-exposed sample for a total follow-up sample of 15 children. The mean age of the children was 10.4 years with a range of 8 to 14 years. In terms of family income, 54% reported incomes between 0 and $21,000, 30% reported incomes between $21,000 and $41,000, and 15% of the families reported incomes over $41,000. Ethnic composition was Euro-American (n = 7), African American (n = 6), Hispanic (n = 1), and Asian (n = 1). Comparison of the participants lost to attrition (n = 9) indicated that they did not differ on age, Tanner stage, gender, income, parent educational attainment, IQ, right or left amygdala volumes, or PTS symptoms from those who completed the follow up (n = 15).

Measures

Exposure to traumatic stress and PTSD symptoms were assessed with the Clinician Administered PTSD scale for Children and Adolescents. The CAPS-CA interview assesses the 17 symptoms for PTSD outlined in DSM-IV and was designed to be a developmentally adjusted counterpart to the CAPS for adults (Blake et al., 1990; Blake, Weathers, Nagy, & Kaloupeck, 1995; Nader et al., 1996). An intra-class correlation coefficient of .97 was established on a sub-sample (n =
10) of the interviews in the current sample with one of the designers of the instrument who rated videotaped recordings of the interviews. The PTSD Reaction Index (Pynoos et al., 1987; Nader, Pynoos, Fairbanks, & Frederick, 1990) was used to initially screen the PTS group for inclusion in this study and is a 20-item self-report instrument used to assess PTSD symptoms after exposure to trauma. The measure uses a 5-point Likert rating scale to assess frequency ranging from “none” to “most of the time.” The reaction index is a widely used instrument that has been shown to be a valid and reliable measure of PTSD symptoms (e.g., inter-rater reliability kappa = .88, internal consistency alpha = .78, see Nader et al., 1990).

Participants’ pubertal development was determined by self-report. Participants selected from drawings with written descriptions representing the five Tanner stages (Marshall & Tanner, 1969, 1970) of pubic hair development and genital development for boys and breast development for girls. Previous research has demonstrated that self-report Tanner staging is a valid and reliable method that has been shown to correlate with physician ratings (Duke, Litt, & Gross, 1980). In this study we focused on pubic hair as an indicator given its applicability to both boys and girls. Tanner stage I is no pubic hair at all and is typical at age 10 and younger; Tanner II, involves a small amount of downy hair [ages 10–11.5], Tanner III, hair becomes more coarse and curly, and begins to extend laterally [ages 11.5–13]; Tanner IV; adult-like hair quality, extending across pubis but sparing medial thighs [ages 13–15]; none of the participants in this sample were Tanner V (hair extends to medial surface of the thighs, ages 15+). Children’s average pubic hair Tanner stage was 2.1 (range 1-4).

The Wechsler Abbreviated Scales of Intelligence (WASI) was used to determine intelligence (The Psychological Corporation, 1999). The WASI is a nationally standardized (N = 2,245) test of intelligence that yields Verbal, Performance, and Full Scale IQ scores that correlate with subscales of the Wechsler Intelligence Scale for Children–Third Edition (WISC–III) and the Wechsler Adult Intelligence Scale–Third Edition (WAIS–III).

Procedures MRI Acquisition and Image Analysis

All subjects and their legal guardians were presented with an institutional review board (IRB)–approved informed consent and agreed to participate. All imaging and image analysis was identical in the PTS and control groups. Data were acquired using a 1.5 Tesla G.E.-Signa scanner (General Electric, Milwaukee, Wisconsin). Coronal 3D volumetric spoiled gradient echo (SPGR) series (TR = 35, TE = 6, flip angle = 45°; number of excitations = 1, FOV = 24, matrix = 256 × 192, 124 - 1.5 mm contiguous slices) were acquired on all subjects and used for all measurements and analysis. Morphometric analysis was done at the Center for Interdisciplinary Brain Sciences Research (CIBSR) at Stanford (additional details can be found in Carrión et al., 2001). Volumetric assessment of segmented image data in the software program, BrainImage (Subramaniam et al., 1997), requires a stepwise process of data importation, removal of non-brain voxels, correction of image non-uniformity, positional normalization, and fuzzy tissue segmentation (Reiss et al., 1998). Individual tissue compartments of Talairach-defined brain regions were then measured. Since regions of interest occur bilaterally in the brain, areas of the right and left hemisphere were also measured individually. The assessment of amygdala in BrainImage requires a manual delineation of regions of interest. Brain tissue was isolated (Andreasen et al., 1996; Kaplan et al., 1997; Reiss et al., 1998; Talairach & Tournoux, 1988) and coronal images were oriented...
perpendicular to the anterior commisure-posterior commisure plane. Inter-rater reliability testing was done between two well-trained raters blind to diagnoses to insure accuracy in measurements. A single rater then circumscribed regions of the amygdala and hippocampus for all subjects on coronal images oriented perpendicular to the anterior commisure-posterior commisure plane and according to a protocol previously developed in our lab (Eliez et al., 2000). To increase the resolution at which the regions of interest could be drawn, the matrix sizes of the coronal datasets were expanded from $256^2$ pixels to $512^2$ pixels using a bicubic interpolation algorithm. The image contrast was increased so that the amygdala was clearly distinguishable from the surrounding white matter and cerebrospinal fluid (CSF). Volume measures recorded total tissue, gray tissue, and white tissue volumes for both the amygdala and. The amygdala was circumscribed coronally and proceeded from the posterior portion, beginning on the slice where the anterior commissure first crosses the midline of the brain. The surrounding white matter tract defined the inferior border of the amygdala while the medial border of the amygdala was defined as the CSF/gray border. The delineation continued superiorly and laterally around the amygdala following the gray/white border. In the posterior regions of the amygdala, the superior border was partially defined by the presence of the entorhinal sulcus. The amygdala was drawn until it disappeared posteriorly. Amygdala volumes are reported in cubic centimeters.

RESULTS

Cross Sectional Time 1 Findings

Means and standard deviations for amygdala volumes are presented in Table 2. No significant main effect differences for controls versus the PTS youth were found. Left and right amygdala volumes were first correlated with age separately for the control and PTS youth. Given the potential for curvilinear associations, parametric linear analyses (i.e., Pearson’s $r$) were supplemented by Spearman’s rho correlations, and an emphasis on visually plotting associations. Results indicated a positive correlation between age and right amygdala volumes in the PTS youth ($r = .43$, $\rho = .49$) beyond the .05 level, but not in the control group where correlations were negative ($r = -.09$, $\rho = -.08$) but did not reach statistical significance. Scatter plot of the correlation between age and right amygdala volumes is depicted in Figure 1. Fisher’s $R$ to $Z$ tests indicated that the differences in the correlation between age and right amygdala volumes were statistically significant ($z$ for $r = 1.78$, $z$ for $\rho = 1.99$, $p < .05$) across the two groups. The pattern of association with age observed in the right amygdala was not observed in left amygdala volumes [left amygdala correlation with age in the PTS youth ($r = .09$, $\rho = .13$) and control group ($r = .15$, $\rho = .15$) no correlations reached statistical significance]. Figure 2 presents the scatterplot of the association of age with right amygdala volumes within the PTS group ($n = 24$) by gender and shows a trend for a stronger association in females.

To examine specificity of the findings and because we previously found (Carrión et al., 2001) that the PTS participants had significantly smaller cranial volumes [$t (46) = 3.19$, $p < .005$] and cerebral volumes than controls [$t (46) = 3.19$, $p < .005$], separate partial correlations between age and right amygdala volumes were calculated for the PTS group controlling for total cranial volumes (partial $r = .46$, $p < .05$), total cerebral volumes (partial $r = .46$, $p < .05$), and right temporal lobe volumes (partial $r = .48$, $p < .05$). The partial correlation between age and right
### TABLE 2
Means and Standard Deviations for Amygdala Regions by PTS and Control

<table>
<thead>
<tr>
<th></th>
<th>Control (n = 24)</th>
<th></th>
<th>PTS (n = 24)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>Mean</td>
<td>SD</td>
</tr>
<tr>
<td>Amygdala Total</td>
<td>4.23</td>
<td>.73</td>
<td>4.03</td>
<td>.57</td>
</tr>
<tr>
<td>Left—Gray</td>
<td>1.91</td>
<td>.39</td>
<td>1.82</td>
<td>.29</td>
</tr>
<tr>
<td>Left—White</td>
<td>0.25</td>
<td>.08</td>
<td>0.23</td>
<td>.08</td>
</tr>
<tr>
<td>Left—Total</td>
<td>2.16</td>
<td>.41</td>
<td>2.05</td>
<td>.34</td>
</tr>
<tr>
<td>Right—Gray</td>
<td>1.84</td>
<td>.36</td>
<td>1.75</td>
<td>.27</td>
</tr>
<tr>
<td>Right—White</td>
<td>0.23</td>
<td>.08</td>
<td>0.23</td>
<td>.06</td>
</tr>
<tr>
<td>Right—Total</td>
<td>2.07</td>
<td>.39</td>
<td>1.98</td>
<td>.31</td>
</tr>
</tbody>
</table>

Note. Volumes are reported in cubic centimeters. PTS = posttraumatic stress.

**FIGURE 1** Scatterplot of age and right amygdala volumes with regression lines by posttraumatic stress (PTS) (n = 24) and control (n = 24) groups.
FIGURE 2 Scatterplot of age and right amygdala volumes with regression lines by sex within the trauma exposed group (n = 24).

As a complementary way of showing the effect implied by the Scatterplot in Figure 1, a 2 (age split in the middle of the age distribution; 7–10 years and 11–14 years) by 2 (PTS versus control) ANOVA indicated a significant age group × PTS interaction \( [F (1, 43) = 7.05, p = .011] \) and the interaction was significant controlling for either total cranial volumes \( [F (1, 43) = 7.93, p < .01] \) or total cerebrum volumes \( [F (1, 43) = 7.79, p < .01] \). Follow up analyses controlling for total cerebrum volumes indicated significantly \( [F (1, 17) = 4.87, p = .041] \) larger right amygdala volumes in controls \( (M = 2.21, SD = .35) \) than PTS \( (M = 1.82, SD = .29) \) in the younger group and no significant \( [F (1, 25) = 0.70, p > .1] \) differences in the older youth [controls \( (M = 1.96, SD = .39) \) and PTS \( (M = 2.09, SD = .28) \)] but the direction of the effect was controls showing smaller volumes than PTS. No effects were found on left amygdala volumes.

Correlations were next conducted between Tanner stage and right amygdala volumes. Results indicated a significant positive correlation between Tanner Stage and right amygdala volumes in the PTS youth \( (r = .45, \rho = .55) \) beyond the .05 level. This association is depicted in
Figure 3 (panel A) and this association similarly held when controlling for cranial volumes, cerebral volumes, right temporal lobe volumes, CAPS-CA scores (PTSD symptoms), and IQ (partial rs ranging from .45 to .5 all ps < .05). The data in Figure 3 were fit with both linear and quadratic lines and the superior fit of the quadratic fit line (R-square = .266 quadratic versus .202 linear, both ps < .05) suggests that the association may taper off in older (more pubertal advancement) youth. The pattern of association with Tanner Stage observed in the right amygdala was not observed in left amygdala volumes [left amygdala correlation with Tanner stage in the PTS youth (r = .14, rho = .20) where no correlations reached statistical significance and curve fitting did not improve fit to a statistically significant level; R-square = .08 quadratic versus .02 linear].

Time 2 Findings

As noted, 15 of the youth with PTS were reevaluated at time 2. Intraclass correlation coefficients (ICC absolute agreement) indicated a fairly high level of stability in both left and right amygdala volumes (right Time 1 to 2 ICC = .71; left ICC = .63). Among the 15 youth with PTS reassessed

1Given the high degree of co-variation of Tanner stage with age (r = .7) it is statistically difficult to unambiguously determine which one is the more important predictor of volume. This is because high co-variation leads to multicollinearity in regression or partial correlations and thus underestimation of relative predictive power; that is, in this sample separately they are statistically significant predictors but together they are both non-significant (e.g., partial r for age with right amygdala volumes was .15 and with Tanner stage .17). One suggestion is to run parallel analyses (see Tabachnick & Fidell, 2001) and simply compare effect sizes. In this sample, Tanner stage tended to show larger effect sizes.
at Time 2. Tanner stage remained significantly correlated with right amygdala volumes at Time 2 ($r = .61$, $\rho = .62$, both $p$s < .05), but less so with age ($r = .38$, $\rho = .44$, both $p$s > .1).

Longitudinal Change Findings

A repeated measures ANOVA was utilized to examine changes in right amygdala volumes across time with Tanner stages 1 and 2 ($n = 10$) versus Tanner stages 3 and 4 ($n = 5$) as a grouping variable for the $n = 15$ youth with PTS. Results indicated a significant main effect of Tanner stage group [$F (1, 13) = 12.66, p = .004$], no significant main effect of time [$F (1, 13) = 1.12, p = .31$], but a significant Tanner group x time interaction [$F (1, 13) = 9.25, p = .009$].² Figure 3, panel B, visually depicts the interaction showing decreases in the older group and increases in the younger group. The increase was significant [$t (9) = 2.06, p < .05$ one tailed] but given the small $n$ the decrease was not [$t (4) = 1.93, p > .05$ one tailed]; however, the decrease was larger in effect size (Cohen’s $d = .65$ increase and Cohen’s $d = .85$ for the decrease). The interaction term remained significant when controlling for PTSD symptoms and effect sizes for increase and decrease were virtually identical. In terms of left amygdala volumes, results indicated no significant effect of Tanner stage group [$F (1, 13) = 1.30, p > .1$], no significant effect of time [$F (1, 13) = 0.13, p > .1$], and no significant Tanner group x time interaction [$F (1, 13) = 3.09, p > .1$].

DISCUSSION

Results indicated a positive association between age and right amygdala volumes among the trauma exposed sample, but a non-significant correlation within a sample of controls. This association in the PTS group was observed for Tanner Stage, as well as controlling for total cerebral volumes and total right temporal volumes, IQ, and PTSD symptoms. Associations with Tanner stage held at time 2, and there was a Tanner stage by time interaction in change in amygdala volumes from time 1 to time 2 with developmentally younger youth showing increases and older youth decreases. While age and pubertal development are highly related, the results using Tanner staging are consistent with the suggestion that pubertal development may provide an important picture of the role of maturational variation in neural development generally (Blakemore et al., 2010) and amygdala volumes in particular.

These findings also may help contextualize future research on youth with a history of traumatic stress and offer suggestions for the use of age in comparing other patient populations. In particular, the findings augment the work of Tottenham and colleagues (Tottenham, 2011; Tottenham & Sheridan, 2010) linking exposure to stress with atypical developmental variation in amygdala volumes and are similar to the work of Chen et al. (2004) who found a positive correlation between amygdala volumes and age. Like Chen et al. (2004), the non-significant correlation in our control sample might represent the tail end of the curvilinear trend observed in Uematsu et al., (2012) with the positive correlation in the PTS group representing atypical or prolonged growth. The

²A similar ANOVA with age group (oldest 5 versus youngest 10) was conducted and the age by time interaction term was not significant. However, the direction of effect was the same with relative increases in the younger and decreases in the older youth.
larger effects found with the rho correlation with Tanner stage may represent the tapering off of the linear association reported in Uematsu et al. (2012). This interpretation is consistent with the improved fit of a curvilinear regression line over a linear one (Figure 3). Future research on developmental variation in amygdala volumes may benefit from employing nonlinear analyses or augmenting linear analysis with curve fitting.

The Tanner stage by time interaction, with developmentally younger youth showing relative increases and older youth showing decreases in amygdala volumes also helps to demonstrate that the tail end of the curvilinear trend observed in Uematsu et al. (2012) may indeed be decreases. Specifically, a positive correlation with age at younger ages followed by no correlation or negative correlation at older ages implies increases followed by decreases, but only longitudinal designs can truly assess change (e.g., they help to exclude cohort effects driving age differences). As far as we are aware our study is the first to report change in amygdala volumes in this age range.

One implication of our findings (and those of Chen and colleagues 2004) is the need to not simply match patient populations on age (or co-vary age) when examining amygdala differences, but to also examine maturation as a potential moderator of differences. Specifically, the different regression lines in Figure 1 imply that, depending on developmental stage, youth with exposure to stress could have relatively smaller (younger) and relatively larger (older) amygdala volumes than controls (and this was consistent with the age group by PTS group ANOVA interaction). Our interpretation is that these findings are suggestive of the need to consider age as a potential moderator of group differences. Future studies testing maturation as a moderator of the main effects (or lack of main effects) of clinical groupings may help address some of the inconsistencies in the literature noted by previous researchers (see Morey et al., 2012).

The findings add to the existing knowledge base on amygdala development in youth but also raise additional questions. Our results do not allow us to conclude if the findings are simply delayed maturation or atypical amygdala development due to stress. We found no evidence that the amygdala volume’s link to age and Tanner stage was attributable to level of PTSD symptoms. PTSD symptom levels were not associated with amygdala volumes in this sample. Finding that controlling for PTSD symptoms did not affect the association between age and amygdala suggests that this association may be due to trauma exposure, not PTSD per se. However, clarifying delayed versus atypical amygdala maturation may have important implications. Specifically, if the differences are simply delayed amygdala maturation then the PTS group may simply catch up, while atypical amygdala development might imply more negative long-term outcomes. As Belsky and de Haan (2011) have argued, there is a need for an increased research focus on the normal developmental processes (they emphasized parenting) associated with differential brain development. Future research is also needed that follows both health community children and those exposed to stress to disentangle stress effects versus normal maturational variation in amygdala volume changes and examines traumatic stress timing issues and other potential mechanisms of differential brain development such as cortisol levels (see Weems & Carrión, 2007).

Findings are limited by the relatively small sample size and the lack of a normative comparison sample followed over time. Although this sample represents the first longitudinal study of amygdala volumes in traumatized youth to date, the study and range of conclusions about additional confounding variables is limited by the sample size and findings should be considered preliminary until replicated. Moreover, the longitudinal and Tanner stage data is limited by the lack of a comparison sample and the comparison sample was not matched on socioeconomic
status and IQ scores. Conclusions about the role of exposure versus natural or delayed development would have been strengthened by a longitudinal control group. Another limitation is that there are likely a number of intermediary factors, not assessed in this study, that could influence the link between maturation and amygdala volumes. While no single study can hope to capture all the potential influences and the present findings help identify maturation as a potential source of inconsistencies in the literature, findings also raise questions about the mechanisms (mediators) whereby stress and maturation exerts its influences on brain development. Our findings support the notion of pubertal development as a possible mechanism. Despite the limitations, a clear implication is that future research may benefit from not just matching or controlling for age/maturity, but examining maturation as a moderator of group differences and employing additional indicators of maturation, such as pubertal status.

REFERENCES


