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Vigilant attention to threat, sleep patterns, and anxiety in peripubertal youth

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Background: Vigilant attention to threat is commonly observed in anxiety, undergoes developmental changes in early adolescence, and has been proposed to interfere with sleep initiation and maintenance. We present one of the first studies to use objective measures to examine associations between vigilant attention to threat and difficulties initiating and maintaining sleep in an early adolescent anxious sample. We also explore the moderating role of development (age, puberty) and sex. Methods: Participants were 66 peripubertal youth (ages 9–14) with a primary anxiety disorder and 24 healthy control subjects. A dot-probe task was used to assess attentional bias to fearful relative to neutral face stimuli. Eye-tracking indexed selective attentional bias to threat, and reaction time bias indexed action readiness to threat. Sleep was assessed via actigraphy (e.g. sleep onset delay, wake after sleep onset, etc.), parent report (Children’s Sleep Habits Questionnaire), and child report (Sleep Self-Report). The Pediatric Anxiety Rating Scale assessed anxiety severity. Results: Eye-tracking initial threat fixation bias (β = .33, p = .001) and threat dwell time bias (β = .22, p = .041) were positively associated with sleep onset latency. Reaction time bias was positively associated with wake after sleep onset (β = .24, p = .026) and parent-reported sleep disturbance (β = .25, p = .019). Anxiety (severity, diagnosis) was not associated with these outcomes. Sex (β = −.32, p = .036) moderated the relation between initial threat fixation bias and sleep onset latency, with a positive association for males (p = .005), but not for females (p = .289). Age and pubertal status did not moderate effects. Conclusions: Vigilant attention to threat is related to longer sleep onset and reduced sleep maintenance. These associations are not stronger in early adolescents with anxiety. Implications for early intervention or prevention that targets vigilant attention to threat to impact sleep disturbance, and vice versa, are discussed. Keywords: Sleep; anxiety; adolescence.

Introduction

In basic science, sleep and wake are increasingly viewed as involving complementary neurobehavioral processes that work together over repeated 24-hr cycles to impact development and functioning (Bassetti et al., 2015; Schwartz & Roth, 2008). Accordingly, it is critical for models of developmental psychopathology to conceptualize symptoms and features of disorders as involving both sleep and wake-relevant processes across the 24-hr sleep-wake cycle (Alfano & Gamble, 2009; Meltzer, 2016). Here, we examine vigilant attention to threat, referring to a general state of increased orientation to and action readiness in response to environmental cues that may signal threat, and its associations with initiating and maintaining sleep during the sensitive period of early adolescence. We examine these questions in a sample of healthy and anxious youth to (a) dimensionally ascertain associations between vigilant attention to threat and sleep in a sample that covers a large swath of the continuum of severity for these key constructs, and (b) inform models of developmental psychopathology and treatment.

The scientific rationale for the focus on vigilant attention to threat in relation to sleep stems from literature bases showing sleep difficulties and attentional bias to threat are common in anxious youth (Bar-Haim, Lamy, Perbergam, Bakermans-Kranenburg, & Van Ijzendoorn, 2007; Dudeny, Sharpe, & Hunt, 2015; McMakin & Alfano, 2015). Sleep-related problems (e.g. difficulty initiating and/or maintaining sleep, decreased sleep duration, fear of sleeping alone, nightmares, daytime sleepiness) are present in up to 90% of children with anxiety disorders (Alfano, Pina, Zerr, & Villalta, 2010; Chase & Pincus, 2011). However, we know little regarding the mechanisms that may underlie specific aspects of sleep disturbance. A key feature of sleep is a necessary loss of awareness and responsiveness to environmental cues (Carskadon & Dement, 2011). Notably, this state is countered by vigilant attention to threat, which serves an adaptive purpose as part of the threat detection system – promoting alertness and mobilization in threatening environmental conditions (Boyer & Bergstrom, 2010; Neuberg, Kenrick, & Schaller, 2011). However, this process may become maladaptive when a propensity to display vigilant attention to threat leads to hypersensitivity to potential threats, disrupting functioning across multiple settings (Neuberg et al., 2011).

Broadly, attentional bias to anxiety-related threat is greater in anxious youth relative to healthy control

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youth (Dudeney et al., 2015). However, attentional bias to anxiety-related threat is not seen consistently in all samples of anxious youth, nor with all variants of relevant tasks (Dudeney et al., 2015; Price et al., 2013; Waters, Wharton, Zimmer-Gembeck, & Craske, 2008). This may be partially explained by findings showing age is predictive of attentional bias to threat, with differences in bias between anxious and healthy control youth widening from pre-to late adolescence (Dudeney et al., 2015) – suggesting anxiety and attentional bias to threat become more tightly linked over the course of development. With respect to links between attentional bias to anxiety-related threat and sleep difficulties, research is lacking. However, with respect to cognitive activation – also a facet of the threat detection system (Neuberg et al., 2011) – research shows associations between subjective report of cognitive-emotional arousal and both insomnia vulnerability (Fernández-Mendoza et al., 2010) and symptom severity among adult populations (Altena et al., 2017), suggesting cognitive-emotional arousal leaves one susceptible to development of insomnia. This factor has also been found in the children of parents displaying vulnerability to insomnia (Fernández-Mendoza et al., 2014), potentially implicating it in the etiology of insomnia.

Moreover, there is a sizeable literature showing increased attentional bias to sleep-related threat in adults with sleep difficulties (e.g. insomnia) relative to controls (Harris et al., 2015), suggesting that narrowing in on sleep-related threatening information is implicated in insomnia. Attentional bias to sleep-related threat has also been found in children of parents with insomnia relative to controls without parental history of insomnia (Ellis, Thomson, Gregory, & Sterr, 2013). However, research explicitly linking sleep-related attentional threat bias and objective measures of sleep disturbance is highly limited with one study actually showing associations between increased sleep-related attentional bias to threat and enhanced sleep maintenance (i.e. increased total sleep time, reduced number of awakenings) and depth (i.e. slow wave sleep) in an adult sample. This was contrary to expectations; however, the authors surmised that this surprising finding may be a function of increased sleep pressure related to the building of a sleep debt (Spiegelhalder et al., 2010). Nevertheless, presleep sleep-related cognitive arousal has been linked to objective difficulties with sleep initiation and maintenance in adults with sleep problems (Spiegelhalder et al., 2012). In addition, presleep cognitive arousal spanning general and sleep-related information is associated with objective difficulties with sleep initiation in adults with sleep problems (Wicklow & Espie, 2000) and with lower subjective sleep duration and overall problems in anxious youth (Alfano et al., 2010). In sum, subjective emotional and sleep-related arousal, and objectively measured sleep-related attentional bias to threat are linked with sleep patterns. However, these studies are few, and there are no studies examining objective measures of attentional bias to threat in relation to objective measures of sleep initiation and maintenance in youth.

This is of particular relevance for youth undergoing the pubertal transition, as neurodevelopmental changes beginning around pubertal onset (i.e. peripubertal) are associated with increased vigilant attention. This promotes the self-protection and survival needed for the adoption of expanded societal roles and responsibilities of adulthood (Spielberg, Olino, Forbes, & Dahl, 2014). Furthermore, physiological changes during pubertal onset (e.g. sex steroids; Mong et al., 2011) are associated with alterations in sleep depth (i.e. increases in lighter sleep stages 1 and 2, and reductions in deeper slow wave sleep stages), circadian rhythms (i.e. delayed phase sleep), and homeostatic drive to sleep (i.e. increased resistance to sleep pressure; Crowley, Acebo, & Carskadon, 2007) that make it more difficult to initiate and maintain sleep.

In addition, research has shown an association between advanced pubertal stage (controlling for age) and anxiety symptoms (Reardon, Leen-Feldner, & Hayward, 2009). These co-occurring maturational changes in vigilant attention to threat, sleep, and anxiety raise the possibility of an interplay among these systems across adolescence, which may contribute to adverse downstream effects on sleep and emotional functioning (Dahl, 1999). Furthermore, interactions between vigilant attention to threat and sleep may be heightened in vulnerable populations. For example, girls are at an increased risk for developing anxiety disorders relative to boys (Beesdo, Knappe, & Pine, 2009). No significant effect of sex on attentional bias to threat in youth has been shown in a meta-analysis. However, a limited number of studies were included, with varied samples and paradigms (Dudeney et al., 2015). Thus, it remains important to examine possible sex differences (Eliot, 2011).

In sum, youth with anxiety display attentional bias to threat and problems with sleep onset and maintenance. However, no prior studies have assessed the intersection of these constructs objectively in early adolescent youth with anxiety. Therefore, we examined attentional bias to unambiguous fearful faces as indexed by orientation to threat (i.e. eye tracking) and readiness for action (i.e. reaction time) during a dot-probe task (MacLeod, Mathews, & Tata, 1986), in relation to sleep onset and maintenance (i.e. actigraphy, subjective report) in a large sample of clinically anxious and nonanxious peripubertal youth. We hypothesized that measures of attentional bias to threat (i.e. reaction time bias and eye tracking) would be associated with actigraphy variables, including those pertaining to sleep onset (i.e. sleep onset latency) and maintenance (i.e. wake after sleep onset, sleep efficiency), in addition to other sleep indices that may be impacted by poor sleep onset and maintenance (i.e. total nocturnal sleep time, and overall sleep

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disturbance as reported by parent and child). We also expected associations between attentional bias to threat and specific subscale-derived indices of subjective sleep disturbance, included on an exploratory basis. Although, based on prior literature (Bar-Haim et al., 2007; Dudenev et al., 2015), we anticipated that the anxious group would differ from healthy controls with respect to attentional bias to threat, our prior research with this sample revealed no significant categorical differences between anxious and nonanxious youth (Price et al., 2013) such that we focused the present analytic strategy on anxiety severity as a dimensional construct. This heterogeneous sample provided variability in both attentional bias to threat and sleep patterns to facilitate the testing of our hypothesis that attentional bias to threat and sleep disturbance are associated, and also to examine whether these associations are stronger as anxiety severity increases.

Finally, given the lack of prior developmental research examining attentional bias to threat-sleep relations, an exploratory aim was to assess the moderating role of developmental factors (age, pubertal status), as well as sex differences in associations.

### Methods

#### Participants

Participants were 90 peripubertal adolescents (ages 9–14 years; M = 11.28; SD = 1.49) who completed attentional bias to threat and sleep monitoring assessments with raw data meeting specified quality metrics (as defined below). Participants were a subset of 176 youth who provided university institutional review board-approved assent with primary caregiver consent and completed a baseline assessment for the Child Anxiety Treatment Study (CATS; Silk et al., 2016), a clinical trial examining the clinical course, treatment response, and neurobehavioral (i.e. emotional functioning, sleep, threat biases, and anxiety) correlates of child anxiety. Study enrollment included 90 participants exhibited a primary anxiety disorder and 24 were healthy control subjects. The relatively larger anxiety sample reflects the goals of the overarching CATS study to delineate mechanisms of youth anxiety treatment. The anxious youth had a primary diagnosis (Diagnostisches and Statistical Manual of Mental Disorders, 4th ed.; American Psychiatric Association, 1994) of generalized anxiety disorder (GAD), social phobia (SP), and/or separation anxiety disorder (SAD) with no (a) current comorbid primary diagnosis of major depressive disorder, obsessive-compulsive disorder, posttraumatic stress disorder, conduct disorder, substance abuse or dependence, or attention deficit hyperactivity disorder combined or hyperactive-impulsive types; or (b) lifetime diagnoses of autism spectrum disorder, bipolar disorder, psychotic depression, schizophrenia, or schizoaffective disorder present per CATS inclusion/exclusion criteria. In addition, those on psychotropic medication were excluded. The comparison group was age and sex-matched with no current or past psychiatric diagnoses, or current or past parental anxiety or mood diagnoses. For a full description of CATS screening procedures, see Silk et al. (2016). The sample was 55.6% female and the racial structure was 83.3% Caucasian, 5.6% African-American, 1.1% Hispanic, and 10% bicultural, which is in line with demographic characteristics in the region in which the study was conducted. See Tables 1 and 2 for baseline characteristics and measurements.

#### Procedure

The 90 participants were selected based on the presence of CATS data from the dot-probe task (see Data Pre-processing section), and sleep monitoring (actigraphy and sleep diary) and exclusion criteria. This is in line with demographic characteristics in the sample. Actigraphy was performed directly following the baseline assessment. School nights were selected as: (a) our emphasis on relationships between attentional bias to threat and sleep in adolescence suggests the importance of capturing nights when youth may be worrying and ruminating about next day activities, which occurs more reliably on school nights; and (b) assessing school and weekend/vacation nights together leads to variable sleep schedules and can interfere with detection of reliable patterns.

Of the original sample (n = 176), 41 (Anxious = 24; Controls = 17) were excluded due to missing actigraphy (Anxious = 18; Controls = 13) or dot-probe data (Anxious = 10; Controls = 3). An additional 45 participants were excluded if they had fewer than a specified number of dot-probe task trials (outlined below in the Data Pre-processing section), leaving 90 participants. Using Chi-square and independent samples t-tests, no significant differences were found between excluded and included cases on clinician-rated anxiety (PARS), parent-reported sleep disturbance (CSHQ), child-reported sleep disturbance (SSR), pubertal status, sex, race, or diagnoses (p > .19). For further details of methodology and sample exclusion criteria, see the Measures section.

#### Measures

##### Dot-Probe Task

The Dot-Probe Task (Bradley, Mogg, Falla, & Hamilton, 1998; MacLeod et al., 1986) is a neurocognitive task assessing key components of attentional bias to emotional cues. In the present study, (a) behavioral reaction time (henceforth referred to as RT) to threat cues in the environment and (b) temporal course of orientation to threat cues in the environment were assessed to provide measures of attentional bias toward threat during wake. In the version used, participants responded as quickly as possible to a dot replacing either a fearful (congruent trials) or neutral (incongruent trials) face presented on either the top or bottom of the screen by pressing a key indicating up or down, yielding a measure of behavioral RT. Eye-tracking data were also collected to determine eye movement patterns in relation to the fearful face stimuli within the trial. Eye-tracking indices were a focus in the current analysis as they provide detailed information on the degree to which the participant is drawn to or disengages from the target stimulus, providing a measure of quality of bias. In addition, eye tracking reveals temporal patterns of visual attention (while RTs provide only a snapshot of attention at a single point in time) and was more reliable than RT indices obtained during this version of the dot-probe task (Price et al., 2015); however, RTs collected during concurrent trials were also assessed as they may provide unique information pertinent to arousal processes. For consistency within all analyses, data were restricted to the 32 trials per participant with a 2,000 ms fearful-neutral face pair presentation, as they provided sufficient time for meaningful eye-tracking analyses in addition to RTs. Additional randomly interspersed trials were presented but not analyzed. These contained fearful-neutral face pairs presented for a shorter (200 ms) interval (32 trials; excluded from present analyses, as 200 ms presentations do not reliably allow for completion of a single eye movement; Henderson & Hollingworth, 1999) and neutral-only trials (16 trials; excluded as they do not provide relevant information on vigilance to threat). Faces were modified from the NimStim battery (Tottenham et al., 2009) to be gray neutral-only trials (16 trials; excluded from present analyses, as 200 ms presentations do not reliably allow for completion of a single eye movement; Henderson & Hollingworth, 1999) and neutral-only trials (16 trials; excluded as they do not provide relevant information on vigilance to threat). Faces were modified from the NimStim battery (Tottenham et al., 2009) to be gray
was used to track eye movements. It features a video camera and infrared light beam directed toward the participant’s eyes (Price et al., 2013). For all present analyses, data from trials with long (2,000 ms) fearful-neutral face pair presentations were used exclusively, as they provided sufficiently long face presentations for meaningful eye-tracking analysis (given that a typical eye movement requires up to 300 ms to complete) and therefore allowed a consistent subset of trials to be used across all measures.

**Actigraphy.** The Ambulatory Monitoring Octagonal Basic Motionlogger actigraph was used as an objective estimate of sleep onset and maintenance. Actigraphy has been shown to have acceptable agreement with polysomnography and high agreement with subjective measures of sleep schedule (e.g., total sleep time and sleep duration; Sadeh, 2011). Participants wore the actigraph and kept a sleep diary (to inform actigraph scoring) from Thursday evening to Tuesday morning. As noted above, only data collected on school nights during the school year were used for this analysis in order to reduce variability that would be associated with vacations and summers and to capture evenings where higher vigilance and sleep disturbance were most likely to occur. Daily phone calls to participants verified school attendance. Data were aggregated to create sleep variable means for morning, afternoon, and nighttime transitions.
nights preceding a verified school day ($M = 2.36$; range 1–3; 14 participants [15.6%] had only one night of actigraphy; sensitivity analyses that removed 14 participants with one night of actigraphy were consistent with the current results and are provided in Appendix S1). Actigraphy variables were identified a priori based on the sleep literature (Buysse, 2014) and hypothesisized links between vigilance and sleep onset or maintenance problems (Dahl & Lewin, 2002) and included: sleep onset latency (minutes to fall asleep), wake after sleep onset (total duration of nighttime awakenings in minutes), and sleep efficiency [a percentage referring to total nocturnal sleep time/ (total time in bed – sleep onset latency)]. Total nocturnal sleep time (total nighttime minutes of sleep) was also included given that it could be readily impacted by difficulties initiating or maintaining sleep.

**Pediatric Anxiety Rating Scale (PARS).** The PARS (The Research Units On Pediatric Psychopathology Anxiety Study Group, 2002) is a psychometrically reliable and valid clinician-rated measure of anxiety severity, including a seven-item anxiety symptom checklist and rating scale. Items are rated from 0 to 5 with higher scores indicative of increased anxiety severity. Consistent with prior research methods (Compton et al., 2010; Silk et al., 2016), a total score was calculated using six items (anxiety symptom frequency, severity of worry, severity of somatic symptoms, avoidance, interference in family and home domains, and interference in social and performance domains), yielding a total score ranging from 0 to 30. Anxiety symptom number was excluded to avoid potential overlap with anxiety symptom frequency. A six-item PARS score greater than 10 is suggestive of clinically significant anxiety (Caporino et al., 2013).

**Children’s Sleep Habits Questionnaire (CSHQ).** The CSHQ (Owens, Spirito, & McGuinn, 2000) is a 33-item parent-report measure of child sleep patterns in the past week. Items are rated using the following scores: 1 (rarely/0–1 time per week), 2 (sometimes/2–4 times per week), and 3 (usually/5–7 times per week) yielding a total score ranging from 33 to 99, and subtotals for bedtime resistance, sleep onset delay, sleep maintenance, sleep quality, and daytime sleepiness. Higher scores reflect greater sleep disturbance. The CSHQ has satisfactory internal consistency, test-retest reliability, and discriminant validity. A cut-off score of 41 has been established as the cutoff for clinically significant sleep problems (Owens, Spirito, & McGuinn, 2000).

**Sleep Self-Report (SSR).** The SSR is a 26-item child-report measure of sleep-related disturbance in the past week, mirroring the Likert scaling and sleep problems assessed in the parent questionnaire, and yielding an overall sleep disturbance total score ranging from 33 to 78 (Owens, Spirito, McGuinn, & Nobile, 2000).

**The Pubertal Development Scale (PDS).** The PDS is a five-item self-report measure of pubertal maturation (physical growth, body hair growth, skin changes, facial hair growth, voice deepening, skin changes, menstruation, breast development) showing good validity and reliability. Items are traditionally rated on a 1 (development not started) to 4 (development completed) scale (Petersen, Crockett, Richards, & Boxer, 1988). However, the PDS was recoded into a 1–5-point total, roughly corresponding to Tanner physiological stage scoring (Shirtcliff, Dahl, & Pollak, 2009). The recoded PDS score differentially captures gonadal and adrenal hormonal signals of physical development (e.g. gonadal hormonal signals are related to growth spurt, breast development, and menarche in girls) thereby yielding a total score, and adrenal, and gonadal subtotals. PDS Total was used to form a dichotomized variable featuring pre/early (ratings 1–2) and mid/late (ratings 3–5) puberty ratings.

**Data preprocessing.** Dot-probe reaction time (RT) and eye-tracking data were preprocessed according to previously described procedures (see Price et al., 2013, 2015). Briefly, RT outliers (values more extreme than the median ± 2.5 x the interquartile range) were recoded using a Winsorizing procedure to improve psychometrics (Price et al., 2015) and used to calculate RT bias scores for each subject as: mean incongruent RT – mean congruent RT [with positive scores indicative of orientation toward threat (i.e., vigilance) and negative scores indicative of disengagement from threat (i.e. avoidance)]. Trials with incorrect responses (8.8% of all trials) were excluded prior to analysis. Eye fixations were defined as eye positions stable within 1° of visual angle for at least 100 ms and were used to calculate bias scores (i.e. percentage of trials/time with fixations to fearful face – percentage of trials/time with fixations to neutral face) for the following gaze patterns: percentage of trials with initial fixations falling within regions of interest defined by the fearful versus neutral face locations (an index of initial attentional capture; initial threat fixation bias); percentage of time spent fixating on fearful versus neutral faces (an index of overall attentional preference; threat dwell time bias); mean duration of individual fixations on fearful versus neutral faces (an index of attentional maintenance; threat fixation duration bias); disengagement latency to fearful versus neutral faces (a latency to fixation on the dot-probe, based on the subset of trials where the dot appeared in the opposite location from the subject’s point of fixation at the end of the face pair presentation – an index of disengagement difficulty; disengagement delay bias). Trials comprised ≥50% blinks and/or with no detectable fixations prior to RT were excluded from analysis (16.3% of trials). Participants (n = 45) were excluded from analyses if they had 10 or fewer trials with usable eye-tracking data or two or fewer trials of each type (congruent/ incongruent) for the disengagement bias. Using bivariate correlations, RT was not significantly associated with eye-tracking indices ($ps = .152–.943$).

**Analytic approach.** A dimensional approach was used to assess the spectra of attentional bias to threat and sleep disturbance across the full sample. This approach was justified given that distributions for the anxious and control samples were highly overlapping on variables of interest (Table 2 and Figure 1A–1E). Data were analyzed in SPSS 23.0 (IBM SPSS Statistics, Chicago, IL) using multiple regression analyses assessing relationships between attentional bias to threat and sleep disturbance. Sensitivity analyses examined associations with anxiety both dimensionally (i.e. scores on the PARS) and categorically (i.e. presence of an anxiety disorder), with attentional bias to threat entered at step 1, and anxiety (anxiety severity, or a coded categorical group variable, in separate analyses) entered at step 2. See Appendix S1 for tests validating appropriateness of covariates. Moderator analyses explored the extent to which developmental factors (age and puberty), and sex moderated the relationship between sleep and vigilance. Specifically, attentional bias to threat and the developmental factor of interest were each entered at step 1, and attentional bias to threat × the developmental factor was entered at step 2 in independent analyses using dichotomized variables.

**Type I error control.** Pairwise regressions were performed for each attentional bias to threat-sleep measure pairing, enabling a comprehensive exploration (Siegle, Condray, Thase, Keshavan, & Steinhauser, 2010) of the extent to which a critical number of attentional bias measures were associated with sleep variables, using two-tailed $p < .05$ for each individual

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test. We used a statistic that estimated the observed number of significant associations with respect to the number of significant associations that would be obtained under a null hypothesis. We ensured that Type I error was held at $p < .05$ across the entire family of regression analyses via permutation tests. To obtain a distribution of the expected number of significant associations under the null hypothesis, we considered simulations in which the attentional bias measures were randomly permuted, to preserve the observed values but create random distributions for their associations. This approach, widely used in psychophysiological and neuroimaging research, treats each pairwise test as a replication and quantifies how many such significant replications are needed in order for the overarching hypothesis (i.e. that sleep disturbance and vigilance are related) to be accepted. Measures were randomly permuted 1,000 times and then each sleep measure was regressed on permuted values to create simulated analyses of the exact same form as those performed in the real dataset. For each permutation, a tally was made of the number of individual attentional bias to threat parameter tests ‘significant’ at $p < .05$. The 95th percentile value generated across these permutations (in this case, four significant tests, each at $p < .05$) was then used as a critical value. The overall probability of obtaining a total of $>4$ significant individual tests (across all vigilance-sleep pairings) under the null hypothesis was, therefore, $p < .05$.

Results

Intercorrelations among sleep indices

Bivariate correlations were performed to assess relationships among sleep variables (Table 3). Results indicated wake after sleep onset was negatively correlated with total sleep time ($r = -.27$, $p = .01$) and positively correlated with overall parent-reported sleep disturbance (CSHQ Total; $r = .26$, $p = .02$) and sleep efficiency ($r = -.40$, $p < .001$). There was a significant correlation between parent- and child-report (Sleep Self-Report Total) sleep disturbance ($r = .51$, $p < .001$). Sleep efficiency was negatively correlated with parent-reported sleep disturbance ($r = -.25$, $p = .02$) and wake after sleep onset ($r = -.98$, $p < .001$). Due to the high correlation between sleep efficiency and wake after sleep onset, sleep efficiency was removed from further analyses.

Relationships among attentional bias to threat indices

One sample $t$-tests were conducted for anxious and healthy controls separately to establish whether RT and eye-tracking bias scores significantly differed from zero. Analyses by group (anxious vs. healthy control) showed that threat dwell time bias ($M = -0.03$, $SD = 0.07$) and threat fixation duration bias ($M = -1.19$, $SD = 4.59$) were significantly different from zero in the anxious group ($t = -2.83$; $p = .006$; $t = -2.11$; $p = .038$), and threat fixation duration bias ($M = -2.17$, $SD = 3.85$) was significantly different from zero in the healthy control group ($t = -2.77$; $p = .01$). Independent samples $t$-tests were used to compare attentional bias to threat variables between groups. There were no significant between group differences. (See Table 2 for means, standard deviations and between group comparisons. See Table 3 for intercorrelations among variables.)

Attentional bias to threat and sleep relationships

Linear regression analyses (Table 4) showed attentional bias to threat indices were associated with several aspects of sleep disturbance (determined a

Figure 1 Reaction time bias and eye-tracking bias indices predicting sleep disturbance measures
priori; see Figure 1A–1E). RT bias was significantly and positively associated with wake after sleep onset \((\beta = .24, t(88) = 2.26, p = .026)\), and parent-reported sleep disturbance \((\beta = .25, t(88) = 2.39, p = .019)\), with greater degree of vigilance associated with increased wake after sleep onset and overall parent-reported sleep disturbance. Eye-tracking indices were also associated with sleep disturbance (Table 5). Specifically, initial threat fixation bias \((\beta = .33, t(89) = 3.30, p = .001)\) and threat dwell time bias \((\beta = .22, t(89) = 2.08, p = .041)\) were significantly and positively associated with sleep onset latency.

### Table 3 Summary of intercorrelations for sleep and attentional bias to threat variables

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Sleep onset latency = minutes to fall asleep; Wake after sleep onset = total duration of nighttime awakenings in minutes; Total sleep time = total nighttime minutes of sleep; Sleep efficiency = a percentage referring to total nocturnal sleep time/(total time in bed - sleep onset latency); Reaction time bias = mean reaction time (ms) to probe replacing neutral face – mean reaction time to probe replacing fearful face; Eye-tracking bias scores = % of trials or % of time during a given window with fixations to fearful face – % of trials/time with fixations to neutral face; CSHQ, Children’s Sleep Habits Questionnaire; SSR, Sleep Self-Report.

*\(p < .05\); **\(p < .01\) (two-tailed significance).

### Table 4 Regression analyses of reaction time bias on sleep disturbance with and without anxiety covariates

#### Sleep efficiency

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Sleep efficiency = a percentage referring to total nocturnal sleep time/(total time in bed – sleep onset latency); Wake after sleep onset = total duration of nighttime awakenings in minutes; PARS, Pediatric Anxiety Rating Scale; Reaction time bias = mean reaction time (ms) to probe replacing neutral face – mean reaction time to probe replacing fearful face; Adj., adjusted; Sig., significance.

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with greater degree of vigilance associated with longer sleep onset latency. Collectively, these five significant findings surpassed the critical value of 4 required to hold family-wise Type I error at $p < .05$. Follow-up analyses explored the eight subscales of the CSHQ parent-reported measure and showed that RT bias was significantly and positively associated with parent-reported sleep onset delay ($\beta = .22, t(85) = 2.06, p = .042$), with greater degree of vigilance associated with increased sleep onset delay subscale scores. No other pairwise relationships between measures of attentional bias to threat (including threat fixation duration bias and disengagement delay bias) and sleep disturbance were found.

**Attentional bias to threat and sleep relationships in reference to anxiety severity and presence of an anxiety disorder**

Results showed the above associations between measures of attentional bias to threat and sleep did not interact with anxiety severity (PARS total; Table 4) diagnostic classification (Table 4; with a single exception, out of all pairwise tests), suggesting anxiety was a valid covariate for regression models and did not moderate the relationship between attentional bias to threat and sleep patterns (Appendix S1). For further analysis of anxiety diagnosis (i.e. sensitivity analyses examining associations with GAD diagnosis without psychiatric comorbidity), see Appendix S1.

**Exploratory moderator analyses for sex and developmental factors**

Sex was assessed as a moderator of attentional bias to threat and sleep patterns, with initial fixation bias and sex entered at step 1 and the initial fixation toward threat–sex interaction term entered at step 2. Results (Table 6 and Figure 2) showed sex was a significant moderator of the relationship between bias in initial threat fixation bias and sleep onset latency ($\beta = -.32, t(89) = -0.21, p = .036$). The model was statistically significant $R^2(2, 87) = 5.61$, $p = .001$, and accounted for an additional 4% of the variance in sleep onset latency ($\Delta R^2 = .04$). The association between initial threat fixation bias and sleep onset latency was significant for males ($\beta = .43, t(39) = 2.96, p = .005$) but not for females ($\beta = .15, t(49) = 1.07, p = .289$). Group comparisons revealed males ($M = 0.03, SD = 0.16$) showed greater initial threat fixation bias relative to females ($M = -0.04, SD = 0.16; t(88) = 2.17, p = .03$). No other attentional bias to threat indices and no sleep measures differed by sex. Sex was not found to be a significant moderator of the other original attentional bias to threat–sleep relationships ($ps = .298 – .604$). Age was also assessed as a moderator of the relationship between attentional bias to threat and sleep patterns. Age was not found to be a significant

| Predictors | $\beta$ | $t$ | $p$ | $R^2$ | Adj. $R^2$ | $F$ | Sig. | Cohen's $f^2$ $b$ | $t$ | $p$ | $R^2$ | Adj. $R^2$ | $F$ | Sig. | Cohen's $f^2$
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**Table 5** Regression analyses of eye tracking on sleep onset latency with and without anxiety covariates

**Sleep onset latency** = minutes to fall asleep; **Initial threat fixation bias** = % of trials with initial fixations falling within regions of interest defined by the fearful face – % of trials with initial fixations falling within regions of interest defined by the neutral face; **Threat dwell time bias** = % of time spent fixating on fearful face – % of time spent fixating on neutral face; **PARS** = Pediatric Anxiety Rating Scale; **Adj.**, adjusted; **Sig.**, significance.

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moderator of original findings ($p_s = .141 – .884$). Finally, pubertal status (pre/early and mid/late) was examined independently of age as a moderator of attentional bias to threat-sleep relationships. No moderating effect of puberty was found for the original findings ($p_s = .567 – .986$).

**Discussion**

The present study is the first to address whether attentional bias to anxiety-related threat (assessed using RT and eye-tracking indices during a dot-probe task) and sleep patterns (assessed by actigraphy and child and parent-report questionnaires) are associated in peripubertal youth. We tested these dimensional relationships in a sample spanning clinically anxious and nonanxious youth, which was expected to provide broad heterogeneity in both attentional bias to threat and sleep variables, while simultaneously allowing us to test whether attentional bias to threat-sleep relationships were linked with anxiety. Results showed indices of attentional bias to threat (biases in RT, initial visual fixation, and dwell time of fixation on threat) were associated with disturbances in sleep onset (i.e. longer sleep onset latency) and maintenance (i.e. increased wake after sleep onset), and increased general measures of sleep disturbance. It is noteworthy that the sample was slightly avoidant overall based on mean attentional bias variable values (Table 2). Nevertheless, our scatterplots indicate a relationship between attentional bias to threat and sleep patterns, whereby increasing vigilance is associated with greater sleep disturbance.

With respect to attentional bias to threat and sleep associations, Type I error control was group-wise allowing for interpretation of patterns of findings that apply to the entire family of tests. Accordingly, interpretation of specific attentional bias to threat and sleep patterns is exploratory. Taking this into consideration, findings revealed that eye-tracking measures (i.e. initial fixation on threat and overall dwell time on threat), which capture overt direction of visual gaze, were significant predictors of delayed sleep onset, while the RT measure, which combines influences from both overt and covert visual attention in addition to nonvisual aspects of attention (e.g. cognitive interference/distraction, response selection), was related to sleep measures that broadly reflect overall dysfunction throughout the night (i.e. wake after sleep onset and parent-reported sleep disturbance).

We also explored the moderating effects of key developmental variables (age and pubertal onset) and sex. Findings were significant above and beyond effects of anxiety severity, presence or absence of an anxiety disorder, developmental factors (i.e. age, puberty), and sex, suggesting that levels of vigilant attention are tightly coupled with sleep disturbance in ways that are distinct from levels of anxiety and certain developmental factors. Associations between degree of vigilance and sleep disturbance were similar whether youth presented with high (clinically elevated) or low overall levels of anxiety, suggesting a mechanistic link that transgresses significant diagnostic and clinical boundaries. This is clinically relevant as even in youth without clinically significant psychopathology, sleep disturbance is associated with poorer executive-functioning, and deficits in cognitive, behavioral and affective domains (Astill,
Van der Heijden, Van Ijzendoorn, & Van Someren, 2012; McMakin et al., 2016) and prospectively predicts increasing symptoms across adolescence (with several reviews of this in recent literature; Gregory & Sadeh, 2016; McMakin & Alfano, 2015). This lack of difference in attentional bias to threat between anxious youth and controls is also developmentally relevant, as the association between attentional bias to anxiety-related threat has been shown to strengthen with age in youth samples (Dudeney et al., 2015). Hence, early intervention may thwart strengthening of links between anxiety and both attentional bias to threat and sleep disturbance.

It is also possible that the lack of differences in outcomes between the anxious and healthy control groups is partially because the measures used here fail to capture some aspects of vigilant attention to threat. Specifically, RT bias and eye-tracking measures captured readiness for action or arousal and orientation to stimuli (i.e. fearful faces) predetermined as threatening. It is also possible that our methods did not capture the tendency to display attentional bias to threat stimuli of an even lower threshold or greater ambiguity than presently used. In addition, it is possible that limited reliability of the dot-probe task may have played a role (Brown et al., 2014). Furthermore, a recent meta-analysis revealed no significant evidence of anxiety-linked attentional bias when using the dot-probe task (Dudeney et al., 2015). The aforementioned factors may have differentiated anxious and healthy control groups, and should be addressed in future research.

With respect to moderation analyses, sex significantly moderated the relationship between bias in initial fixation on threat and sleep onset latency. Specifically, a greater initial fixation on threat was associated with greater sleep onset delay in males, but there was no association in females, suggesting a sex-specific pattern of driving influences. The reasons for this discrepancy are unclear. Despite the meta-analytic findings of no effect of sex on attentional bias to threat and sleep disturbance (McMakin & Alfano, 2016), studies using the dot-probe paradigm specifically provide preliminary support for the finding of greater attentional bias to threat in girls relative to boys (Vasey, El-Hag, & Daleiden, 1996; Waters, Lipp, & Spence, 2004). However, this may be complicated by hormonal changes, as pubertal increases in testosterone have been found to be associated with increased neural activation to threat (Spielberg et al., 2014). Clearly, more research is needed to understand this sex difference. With respect to developmental factors, no moderating effects of age or pubertal status were evident, implying that the magnitude of attentional bias to threat-sleep relationships is relatively consistent across this narrow developmental window (ages 9–14). The vast changes in sleep patterns, as well as neural, social, and physiological growth that begin to take shape during this peripubertal developmental window, do not appear to influence the degree of vigilance and sleep problems across this age range. However, based on previous findings (Dudeney et al., 2015), we would expect these associations to strengthen in later stages of adolescence.

Despite significant associations between several measures of attentional bias to threat and sleep onset/maintenance, there were several null findings that are worth noting. No significant relationships were found between sleep patterns and two eye-tracking measures related to disengagement from threat: fixation maintenance bias (i.e. bias in the length or ‘stickiness’ of individual fixations to fearful vs. neutral faces) and disengagement delay bias (i.e. latency in shifting attention away from threat after faces were replaced by the probe). Furthermore, no significant relationships were found between measures of attentional bias to threat (RT bias or eye-tracking indices) and child-reported sleep patterns and problems. Discrepancies could stem from measurement error, such as underreported sleep difficulties by children relative to parents, as has been previously demonstrated (Alfano et al., 2010), and/or the specific nature of vigilance-sleep relationships, for example, stronger links between sleep variables and relatively ‘early’ components of attention as compared with ‘late’ components like disengagement.

Furthermore, it should be noted that the reaction time and eye-tracking measures were not associated with one another. Prior research suggests that this is not uncommon (Isaacowitz, Wadlinger, Goren, & Wilson, 2006; Waechter, Nelson, Wright, Hyatt, & Oakman, 2014) as they measure distinct facets of attentional bias, with RT capturing a combination of overt and covert attentional processes, and eye-tracking measuring overt visual attention (Armsrong & Olatunji, 2012; Weierich & Treat, 2015). Despite the lack of an association between RT and eye tracking, baseline means for both indices were in the same direction and both measures were significantly associated with sleep measures.

**Developmental implications**

Our findings support positive associations between attentional bias to threat and sleep disturbance – during a sensitive period of peripubertal development. Although exploratory analyses of developmental variables (age, pubertal status) suggested relative stability in these relationships in the present age and pubertal status range, it is possible that youth in our sample who display increased attentional bias and sleep disturbances may be at higher risk for negative trajectories of symptoms and functioning later in adolescence. Developmental comparisons across a broader age range or pubertal stage are needed to address this question.
Clinical implications

Based on our hypothesis that vigilant attention to threat is associated with difficulties with sleep onset and maintenance, we would expect that interventions designed to minimize attentional bias toward threat either directly (attentional bias modification) or indirectly (i.e. cognitive behavioral therapy, exposure/response prevention) may lead to improved sleep onset and maintenance. In addition, interventions targeting relaxation, savoring positive feelings, or mindfulness may help reduce heightened sensitivity to threat and promote optimal sleep transitions and maintenance. Finally, improving sleep onset and continuity may prove to be beneficial in reducing vigilant attention to threat during waking hours. The prospective impact of persistent sleep disturbance on escalating symptoms of anxiety and depression in adolescence is well documented (for review, see McMakin & Alfano, 2015), suggesting that targeted sleep interventions could both ameliorate sleep disturbance and mollify trajectories of risk in adolescence. Furthermore, as findings are cross-sectional, it may be useful to examine the extent to which behavioral interventions for sleep disturbance are associated with reduced attentional bias to threatening cues.

Limitations

Several limitations are relevant to interpretations and conclusions. First, 14 participants (15.6%) had only one night of actigraphy, which is below the recommended 3-day threshold for obtaining a reliable estimate of sleep patterns (Littner et al., 2003). In addition, findings are cross-sectional; thus, directionality of influences among variables over time is unclear. Future longitudinal and experimental research is needed to parse these associations further. Also, anxiety severity was assessed using a measure of symptoms present within a 1-week period. It is possible that state anxiety assessed more proximally to dot-probe task administration may have altered the findings. As such, further research is needed to examine differences in the contributions of state relative to trait anxiety on relationships between outcome variables. In addition, sensitivity analyses (see Appendix S1) examining associations with GAD without psychiatric comorbidity produced results that were consistent with original findings with respect to directionality of effects. Future work should further explore the role of type of anxiety diagnosis in relationships among variables. Furthermore, findings may be specific to the dot-probe task design used here, reducing comparability with previous studies. For example, this version of the dot-probe task requires participants to indicate the location of the probe on the screen rather than the type of probe. Therefore, it is limited with respect to the ability of one to infer the location of visual attention through reaction times. However, this version is common in pediatric research (Vasey et al., 1996; Waters et al., 2004), and use of concurrent eye tracking in the present study does diminish this limitation as it allows for inferences to be drawn regarding the location of gaze. In addition, fearful rather than angry faces were used as threat stimuli. However, findings from Mogg, Garner, and Bradley (2007) showed that participants with anxiety showed similar patterns of attentional bias to fearful and angry faces, perhaps indicative of an overarching neural threat-processing system, and suggesting fearful face stimuli may ultimately prove to be sufficient for examining these outcomes of interest. Furthermore, it would be informative to investigate attentional bias to both anxiety-related stimuli and sleep-related stimuli in relation to sleep disturbance in anxious youth as these biases emerge over time as it is possible that anxiety-related attentional threat bias may transition to sleep-related attentional threat bias in those with chronic sleep disturbance. More broadly, future research should expand on the multimodal assessment of vigilant attention to further elucidate relevant biobehavioral mechanisms (e.g. neural substrates) involved in the relationship between attentional bias to threat and sleep. In addition, future longitudinal follow-up studies will be able to determine the importance of taking into account developmental changes.

Conclusions

In summary, results of the present study show that attentional bias to threat and sleep onset/maintenance are linked in peripubertal youth, and neither anxiety severity nor diagnostic classification are associated with these outcomes. Findings suggest both attentional bias to threat and sleep are dimensionally associated, with youth displaying greater degree of threat vigilance also showing increased sleep disturbance. Relationships between variables remained strong even after accounting for developmental factors (age and pubertal onset), suggesting the associations between attentional bias and sleep disturbance may be relatively stable within the target developmental range of the present study. The present findings call for further exploration of attentional bias to threat-sleep associations during various phases of development to deepen our understanding of how these relationships fluctuate during adolescence or as a function of specific developmental changes, and whether or not intervention strategies that target these dimensions can improve clinical outcomes.

Ethical considerations

We attest that the original trial from which this data originated was approved by a university institutional review board. All subjects provided written assent...
along with primary caregiver consent for participation following a thorough explanation of study procedures.

Supporting information
Additional supplemental material may be found online in the Supporting Information section at the end of the article:
Appendix S1. Sensitivity analyses of attentional bias to threat and sleep relationships in reference to nights of actigraphy.

Acknowledgements
The research reported in this publication was supported by the National Institute of Mental Health of the National Institutes of Health under Award Number MH080215.

Key points
- Vigilant attention to threat is common in anxious youth and undergoes developmental changes in adolescence. The current study proposed that vigilant attention to threat is associated with difficulty initiating and maintaining sleep in peripubertal youth.
- Findings showed associations between objective measures of attentional bias to threat and difficulties with sleep onset and maintenance.
- Associations between attentional bias to threat and sleep disturbance did not interact with anxiety severity or diagnostic classification.
- There was preliminary evidence for a moderating role of sex whereby boys evidenced associations between initial bias toward threat and delayed sleep onset.
- Targeting vigilant attention to threat may positively impact sleep (e.g. sleep onset and maintenance) and vice versa. This is especially relevant for youth within the sensitive period of peripuberty.

References

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