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Neurobiology of Early-onset Anxiety Disorders

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The past two decades of research on both the longitudinal course and familial distribution of anxiety disorders suggest that many forms of adult anxiety disorders can be conceptualized as developmental conditions. Family studies demonstrate strong associations between anxiety in parents and children (Battaglia et al. 1998; Weissman et al. 1997). Similarly, prospective epidemiological studies find strong associations between anxiety during adolescence and anxiety, as well as major depression, during adulthood (Pine 1999). Given the lifelong implications of anxiety disorders in childhood and adolescence, it is critical to understand the basic neurobiological mechanisms involved in these disorders early in their developmental trajectory.

Clinical characteristics of childhood anxiety disorders have made them particularly challenging to study, highlighting the importance of using neurobiological markers to eventually improve the nosological system and to develop targeted interventions. For example, childhood anxiety disorders exhibit high rates of comorbidity in both community- and clinic-based studies (Gurley et al. 1996; Verduin & Kendall 2003). Comorbidity appears particularly high for generalized anxiety disorder (GAD) in children, which shows strong associations with a range of conditions, including social anxiety disorder (SAD) and major depression (Kessler & Walters 1998; Pine 1999). Such high rates of comorbidity raise questions as to the validity of current nosological categories and suggest the need for alternative bodies of knowledge to validate conditions (Pine 2007). Additionally, there is an apparent contradiction between the transient nature of some childhood anxiety disorders and evidence suggesting that adult mood and anxiety disorders often begin in childhood. Longitudinal data suggest that this paradox may result from very high rates of childhood anxiety disorders. In most children, these disorders remit. However, those children who remain anxious may account for a large proportion of adult cases of both mood and anxiety disorders. Given this possibility, there is a pressing need to determine factors that distinguish anxious children at low versus high risk for continued anxiety or depression in adulthood. With advances in neurobiological understandings, new avenues for developing such insights are available. Specifically, children and adolescents who manifest clinically significant anxiety but then mature to become adults free of such anxiety might be viewed as having successfully extinguished their fears. Anxious children and adolescents who remain anxious as adults can be viewed as suffering from a deficiency in extinction (Pine et al. 2009). Monumental increases have occurred in understandings of fear extinction neurobiology. As such, the view of persistently anxious children as suffering from some form of "extinction deficit" generates novel insights for treatments that target anxiety by attempting to facilitate extinction.

The current chapter reviews the progress that has been made in the development of neurobiological models of pediatric anxiety disorders, as well as ongoing efforts to bridge basic science and clinical research. The first section briefly reviews the clinical characteristics of pediatric anxiety disorders. Post-traumatic stress disorder and obsessive-compulsive disorder will not be covered in the present chapter and can be found in Chapters X and Y. The second section reviews the basic science research of the neurobiology of fear responses that has provided the foundation for studies of youth with clinical anxiety. The third section will survey clinical research on the neurobiology of anxiety disorders in children and adolescents. Specific physiological responses associated with fear and panic, as well as genetic influences on clinical symptoms associated with anxiety disorders, will also be presented. The concluding section presents future directions for research on

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the neurobiology of anxiety disorders, with a focus on developmental approaches.

ANXIETY DISORDERS: SYMPTOMS AND PREVALENCE

Anxiety disorders are the most commonly occurring psychiatric conditions affecting children and adolescents. Although the prevalence of anxiety disorders varies by the manner in which they are operationalized and assessed, estimates range from 3% to 30% for youth under the age of 18 (Costello et al. 2005). Despite the fact that some children with anxiety disorders are effectively treated with empirically based treatment (e.g., exposure therapy for specific phobia) or experience a reduction in symptoms as they mature (e.g., SAD), even children with remitted anxiety remain vulnerable for later-life recurrences. Approximately 50% of children with anxiety disorders have some type of psychopathology in adulthood (Copeland et al. 2009; Gregory 2007; Pine et al. 1998). Therefore, significant gaps remain in the effective lifelong management of these disorders.

Fear is a normal response to danger or threat. Additionally, some fears are normative at certain developmental stages (i.e., separation anxiety in toddlers, social anxiety in adolescents). This presents a challenge to clinicians and researchers working with fearful youth. Broadly conceptualized, anxiety disorders occur when an individual experiences clinically excessive fear, either in the absence of a real threat or out of proportion to the threat, and this fear causes significant functional impairment in any one of various domains. This impairment in functioning is usually related to avoiding situations that will elicit fear or that are perceived as threatening. For example, a child with social phobia who fears rejection or embarrassment in social situations may avoid these situations and instead surround himself with individuals who will not reject him (e.g., parents, siblings). The following sections review the symptoms and prevalence of the five most common anxiety disorders in childhood: GAD, SAD, panic disorder, social phobia, and specific phobia.

Generalized Anxiety Disorder

According to the *Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition* (DSM-IV), GAD occurs when worry becomes pervasive in daily functioning. Individuals with GAD worry about a range of situations, including but not limited to those focused on approval, competence, future events, and new or unfamiliar situations. Before 1994, with the publication of DSM-IV, children presenting with symptoms of pervasive worry typically were classified as suffering from overanxious disorder (OAD), a condition that appeared in both DSM-III and DSM-III-R. With the 1994 publication of DSM-IV, the diagnosis of OAD was eliminated, and children presenting with pervasive worry were classified as suffering from GAD. Questions do remain about the advantages and disadvantages of this revision. On the one hand, there may be subtle differences in the longitudinal outcomes of children in the community presenting with OAD or GAD-typical symptoms, leading some to suggest that OAD and GAD are different conditions (Copeland et al. 2009). On the other hand, the overwhelming majority of children seen in the clinic with symptoms of either disorder meet criteria for both. As such, diagnosing such children with GAD utilizes a diagnostic approach consistent with that used among adults, thereby encouraging attempts to relate anxiety across the lifespan.

Children with GAD often seek reassurance to help allay fear, although this reassurance is typically ineffective. Somatic complaints and irritability are also characteristic of GAD. In terms of comorbidity, GAD or OAD in childhood is closely linked with other anxiety disorders, as well as to depression, and may hold similar genetic predispositions (Lau & Pine 2008; Pine 1999). This relationship may represent the broad fractures in social and emotional functioning common to both GAD and major depressive disorder.

The symptoms of GAD in youth are often difficult to differentiate from those of other anxiety disorders, such as social phobia and separation anxiety disorder. This difficulty of differential diagnosis is related to the fact that the type of pervasive worry found in children with GAD often centers around issues related to social interaction and academic performance, which can closely mirror these other disorders. For example, a child may worry about doing well at school, and it may be difficult to determine if the pervasiveness of the worry meets criteria for GAD or if it is better captured by socially phobic fears of evaluation. Additionally, these overlapping symptoms likely contribute to the significant comorbidity of GAD with other anxiety disorders. As a result, most studies of pediatric GAD include heterogeneous samples with significant comorbidity precluding specific conclusions about GAD.

High risk for adult psychopathology associated with childhood GAD has been demonstrated, although, as noted above, some questions do remain on the degree to which early risk is best exemplified by children with OAD as opposed to GAD (Copeland et al. 2009; Pine et al. 1998). Both retrospective and prospective data suggest that adolescent GAD strongly predicts adult major depression (Kessler & Walters 1998; Pine 1999). These data are consistent with data from family studies on the associations between childhood GAD or OAD and adult major depression (Copeland et al. 2009; Pine 1999). These associations may partially arise through the effects of stress, as adverse life events during adolescence show

the strongest and most consistent association with depression and GAD (Pine 2003).

Separation Anxiety Disorder

Separation anxiety disorder is characterized by an extreme fear of separation from the home or from an attachment figure (e.g., parent). Symptoms must begin before age 18, persist for at least 4 weeks, and be beyond what is expected for the individual's developmental level. As a result, it is the only anxiety disorder that is classified as a disorder first diagnosed in childhood in the DSM-IV. Separation anxiety disorder typically develops during middle childhood. Rates of the disorder show a relatively marked decline from childhood to adolescence, consistent with age-related changes in the related normal developmental phenomenon of separation anxiety. These data raise questions about factors that distinguish normal from pathological separation anxiety. Findings from longitudinal studies reveal a relationship between SAD and later panic attacks (Pine 1999). Moreover, family studies find that childhood separation anxiety disorder relates to adult panic disorder (Battaglia et al. 1998; Capps et al. 1996; Warner et al. 1995; Weissman et al. 1997). Other data suggest high rates of separation anxiety disorder in children of parents with other psychiatric disorders, such as major depression (Biederman et al. 2001).

Panic Disorder

Panic disorder is characterized by the presence of recurrent, unexpected panic attacks followed by at least 1 month of significant worry about the recurrence or consequences of another panic attack, as well as behavioral changes involving avoidance of triggers associated with panic attacks. Panic disorder is further qualified as occurring either in the presence or absence of *agoraphobia*, the fear of places or situations in which escape might be difficult or where embarrassment is likely if a panic attack were to occur. Panic attacks are a period of intense discomfort or fear that is accompanied by at least four somatic or cognitive symptoms such as nausea, chest pain, fear of dying, or chills. Panic attacks can be unexpected, situationally bound, or situationally predisposed, and each of these types is defined by a different set of relationships between the panic attack and the trigger. Panic disorder is very rare before adolescence (Costello et al. 1996), exhibiting rates that are generally below 1% for lifetime diagnosis (Pine 1999). In general, children tend to exhibit a relatively abrupt increase in rates of panic attacks around puberty (Hayward et al. 1992), although such panic attacks only infrequently progress to full-blown panic disorder (Pine 1999). In these children, panic attacks typically progress to panic disorder over a relatively lengthy period, into early adulthood.

Social Phobia

The terms *social phobia* or *social anxiety disorder* refer to a pattern of recurrent fear and apprehension in social situations or scenarios in which an individual believes he or she may be scrutinized. Individuals with social phobia experience an immediate fear response, and may even have a panic attack, when exposed to social or evaluative situations. Social situations are typically avoided or endured with distress, and this must cause functional impairment. Although adults and adolescents with this disorder often acknowledge that their fear is excessive, this is not typically the case with children. For individuals under the age of 18, symptoms must persist for at least 6 months before a diagnosis of social phobia can be made. Available data from epidemiological studies suggest that social phobia represents a relatively common primary diagnosis among adolescents (Pine 1999; Stein et al. 2000; Velting & Albano 2001; Wittchen et al. 2000).

Pediatric rates of social phobia peak in adolescence, mirroring normal developmental increases in social anxiety (Pine 1999; Stein et al. 2000). This raises questions about factors that distinguish adolescents with "normal" as opposed to "pathological" aspects of social anxiety. Specifically, the presence of moderate social anxiety may be viewed as developmentally appropriate and within the range of normal adaptive behavior, particularly during adolescence. For a sizable minority of children and adolescents, however, social anxiety becomes maladaptive, causing significant distress and impairment.

Early signs of potential risk for social phobia may be manifested in a temperamental profile, namely *behavioral inhibition*, recognizable early in life. This profile refers to a pattern of wariness in novel situations, particularly new social situations. Although children with behavioral inhibition face a high risk for various childhood anxiety disorders, by adolescence this risk is specific for social phobia (Schwartz et al. 1999). There is also evidence for a common neurobiological substrate underlying behavioral inhibition and social anxiety (Perez-Edgar et al. 2007).

Specific Phobia

Specific phobia refers to an unreasonable fear of a specific object or environmental condition that causes impairment in normal functioning, primarily due to avoidance of feared situations or objects. Diagnosis is appropriate only if the avoidance or fear interferes with functioning or if the fear is felt to be clinically significant in terms of overall magnitude. For diagnosis before

the age of 18, symptoms must persist for more than 6 months. Typical onset occurs during childhood, although it may arise in other developmental periods. As with social phobia, adults and adolescents with specific phobias typically realize that their fear is excessive, whereas children often do not. Data on developmental changes in phobias, as abnormal conditions, show parallels with data on developmental changes in normal specific fears, such as childhood fears of small animals or the dark. Prevalence of specific phobia in children and adolescents has been estimated at approximately 3%–4%, with the highest prevalence generally appearing between 10 and 13 years of age (Essau et al. 2000; Fyer et al. 1998).

Specific phobias in children generally cluster into three subtypes, similar to those found in adults. Children 7–19 years of age report animal phobias, blood injection–injury phobias, and environmental-situational phobias (Muris et al. 1999). Of these three subtypes, blood phobia has been shown to result in physiological symptomatology consisting of an initial rise in heart rate, followed by vasovagal bradycardia and, frequently, syncope (Marks 1988). This may indicate a uniquely different neurobiological substrate of blood/injection/injury phobia relative to other specific phobias.

NEUROBIOLOGY OF FEAR

Basic science research has provided a critical foundation upon which biological studies of anxiety disorders are based. Pioneering research by basic scientists such as Joseph LeDoux and Michael Davis provides essential evidence that the amygdala is a vital brain region for quickly processing threatening stimuli across species (Davis 1988; LeDoux 2000). The amygdala and related brain systems, including but not limited to the prefrontal cortex, are also central to effective functioning in threat perception, social response, and emotion processing (Phelps & LeDoux 2005; Pine 2003). Studies with animals have further demonstrated that the brain regions involved in fear responses are highly vulnerable to environmental influences and that early life influences can have lifelong implications (LeDoux 2000).

Current models of the neural basis of fear have emerged from experiments using fear conditioning paradigms across species. In these experiments, a neutral stimulus, such as a tone or a light, is paired with an aversive stimulus, such as a shock, a loud noise, or an aversive air blast. Following this experience, the formerly neutral stimulus becomes a conditioned stimulus (CS+) and acquires the ability to elicit behaviors and physiological responses formerly only associated with the aversive stimulus, the unconditioned stimulus (UCS). Enthusiasm for this work derives at least partly from the precise delineation of neural circuits, down to the level of the genome, engaged by environmental components that produce fear conditioning (LeDoux 1998). Additionally, this paradigm allows for the examination of neural circuits involved in extinction, a reduction in the learned fear response due to repeated presentation of the CS+. This is particularly relevant for the study of pediatric anxiety disorders, given recent theories that suggest that they result from a deficiency in extinction (Pine et al. 2009).

The amygdala plays a central role in mediating fear conditioning, and changes in genes within the amygdala are thought to reflect changes in behavior and physiology associated with this process. Further, results of human studies have tended to corroborate and build upon basic animal research regarding the role of the amygdaloid complex in fear conditioning as well as other emotional responses (Pine et al. 2001). Studies with nonhuman primates have demonstrated that the developmental timing of damage to the amygdala significantly impacts the way in which individuals perceive threats, particularly social threats (Phelps & LeDoux 2005; Pine 2003, 2007). These findings have raised questions about the impact of anxiety disorders on amygdala and prefrontal cortex development during adolescence, when key social-cognitive processes are forming.

The amygdala's role in fear responding has been further examined using socially salient stimuli such as faces. Early work in this area revealed that adults with bilateral amygdala damage have impaired recognition of fear expressions (Adolphs et al. 1995). Imaging studies of adults have shown amygdala activation in response to angry faces (Easter et al. 2005) representing direct threat, and fearful faces, which represent an ambiguous or unknown threat (Whalen et al. 1998). Similarly, amygdala activation during the viewing of fearful faces compared to control stimuli has been demonstrated in studies of children and adolescents (Guyer et al. 2008). Moreover, there is some evidence of differential amygdala sensitivity to fearful faces across development, although the directions of the findings, in terms of greater activation among younger relative to older individuals, varies with methodological aspects of the studies (Guyer et al. 2008). These findings, along with data showing age-related changes in face-emotion processing (Thomas et al. 2007), suggest developmental shifts in amygdala-based circuits that may contribute to the development of anxiety disorders in youth.

CLINICAL RESEARCH FINDINGS

Neuroimaging

Similarities in the neurobiological correlates of fear responses in rodents, nonhuman primates, and humans

implicate particular neural circuits in the pathophysiology of anxiety disorders (Pine 2007). However, even with advances in functional magnetic resonance imaging (fMRI) research, efforts to link clinical symptoms with neural circuitry are still in preliminary stages, particularly for children and adolescents. These developmental periods carry several challenges, including rapid neurobiological changes as well as the fact that nosology is often based upon adult presentation of disorders. Given these challenges, researchers have focused on neural circuits associated with known cognitive processes pertaining to fear in animals to establish a neural basis for anxiety disorder symptoms. This extensive body of research implicates the amygdala in fear processing across species.

In adults, patients with various anxiety disorders, particularly social phobia, show greater amygdala activity in response to feared or emotional stimuli than do nonanxious controls (see Etkin & Wager 2007, for a meta-analytic review). Similar results have been observed in pediatric samples and are summarized in Table 10.1. For example, functional neuroimaging studies demonstrate an increased amygdala response to negative face-emotion displays in pediatric anxiety disorders (McClure et al. 2007; Monk et al. 2008). Recent evidence suggests that amygdala hyper-responsivity to feared stimuli may underlie cognitive biases observed in children and adolescents with anxiety disorders. For example, youth with anxiety disorders show more activation in the amygdala than do nonanxious youth when they direct their attention to their internally experienced levels of fear (McClure et al. 2007). Additionally, the degree of amygdala activation has been found to correlate with the severity of anxiety symptoms and the degree of attentional biases toward a social threat (e.g., an angry face) (Monk et al. 2008). Structural studies have also shown differences in amygdala morphology between children with anxiety disorders and healthy controls, although the direction of these findings has been inconsistent (Milham et al. 2005 vs. De Bellis et al. 2002).

The amygdala is part of a larger emotion-regulation network that includes regions of prefrontal cortex that have also demonstrated altered function in pediatric anxiety disorders (see Table 10.1). For example, adolescents with GAD show increased ventrolateral prefrontal cortex (VLPFC) activation in response to angry faces presented for 500 msec (Monk et al. 2006). This result contrasts with a subsequent study that found increased amygdala activation to angry faces presented subliminally (17 msec). This difference suggests that threat is initially processed by the amygdala, and then prefrontal regions such as the VLPFC become involved in regulating that response. Further support for this is provided by functional connectivity analyses showing evidence of reduced negative coupling between amygdala and VLPFC in GAD youth, which is suggestive of reduced capacity to regulate emotion (Monk et al. 2006, 2008).

Recent studies have taken novel approaches to examine these functional alterations in the amygdala and prefrontal cortex using alternative paradigms designed to be more directly relevant for pediatric anxiety disorders (see Table 10.1). First, a recent study by Guyer and

TABLE 10.1 *Functional neuroimaging studies among children and adolescents with anxiety**

Author, Date of publication	Sample	Task	Targeted brain regions	Primary finding
Guyer et al. 2008	Case-control: Adolescent anxiety	Peer evaluation	Amygdala, VLPFC	ANX greater amygdale activation than HC. Significant positive amygdala-VLPFC relationship in ANX vs. HC.
Krain et al. 2008	Case-control: Adolescent anxiety	Decision-making	Amygdala, frontal regions	In ANX, greater intolerance of uncertainty associated with greater activation in amygdala and frontal regions.
Lau et al. 2009	Case Control: Adolescent anxiety, depression or both. 5-HTT Alleles	Face-emotion paradigm	Amygdala	HC with one short 5-HTT allele greater amygdala response to fearful faces than HC without short allele. ANX with two long 5-HTT alleles show greater amygdala activation than ANX with one long allele.
McClure et al. 2007	Case-control: Adolescent anxiety	Face-attention paradigm	Amygdala, VLPFC, ACC	ANX show greater activation amygdala, VLPFC, ACC than HC.
Monk et al. 2006	Case-control: Adolescent anxiety	Attention-orienting paradigm	VLPFC	ANX greater VLPFC activation than HC.
Monk et al. 2008	Case-control: Adolescent anxiety	Attention-orienting paradigm	Amygdala, VLPFC	ANX greater amygdale activation than HC. Amygdala and VLPFC functionally related during threat task.

Abbreviations: 5-HTT, serotonin transporter, ACC, anterior cingulate cortex, ANX, anxiety disorder group, HC, healthy control group, VLPFC, ventrolateral prefrontal cortex.

colleagues (2008) used a peer evaluation task to examine the impact of social anxiety on brain responses. Results indicated that adolescents with anxiety disorders showed amygdala and VLPFC dysfunction when anticipating negative social evaluation, a situation that occurs often for these teens. Second, a recent pediatric fMRI study found a significant association between intolerance of uncertainty and brain responses to uncertainty in adolescents with GAD and/or social phobia (Krain et al. 2008). Cognitive models propose a prominent role of intolerance of uncertainty, which is defined as "the tendency to react negatively on an emotional, cognitive, and behavioral level to uncertain situations and events" (Dugas et al. 2004) in anxiety disorders, particularly GAD. Therefore, understanding the neural basis of this cognitive bias can inform pathophysiological models of how anxiety disorders such as GAD develop. Within the anxious group, those who were more intolerant of uncertainty showed greater activation in frontal and limbic brain regions than did youth with low uncertainty tolerance. Although this study did not find a difference between anxiety-disordered and control youth, it represents an important step in understanding the relationship of specific traits or features of anxiety, such as intolerance of uncertainty, to underlying neural circuitry. Such findings call into question current nosological boundaries and demonstrate the need for further study of the neural basis of information processing and trait characteristics to inform diagnostic definitions.

Physiological Probes: Panic Disorder

Panic disorder is characterized by panic attacks, paroxysms of acute anxiety often accompanied by changes in respiration (Klein 1993; Pine et al. 2000). This observation has stimulated a series of studies examining respiratory function in adults with this disorder. Much of this work suggests that panic disorder is characterized by enhanced reactivity to innately dangerous situations that elicit changes in respiration. For example, panic attacks can be induced by exposure to substances that change respiratory patterns such as sodium lactate, doxapram, and carbon dioxide (CO_2). Studies in this area generate some of the strongest, most consistent evidence of biological correlates of clinical anxiety. Nevertheless, there remains considerable disagreement, fueled partially by the limited understanding of neural regulation of breathing and hypersensitivity to respiratory provocation, on the ultimate origin behind such enhanced reactivity.

In adults, the relationship of ventilatory physiology to anxiety disorders has been demonstrated through research on enhanced sensitivity to CO_2, manifested as both changes in subjective state and physiology, among panic disorder patients (Klein 1993; Perna et al. 1995). Klein (1993) suggests that these findings reflect an innate sensitivity of the central receptors to signals of possible respiratory compromise. At least among adults, such ventilatory abnormalities have also been identified in first-degree relatives of patients with panic disorder (Coryell 1997; Perna et al. 1995), whereas among children they manifest in patients with possible precursors for panic disorders, such as separation anxiety disorder (Pine et al. 2000) or isolated panic attacks (Perna et al. 1995). Additionally, studies have found family loading for panic disorder in the adult relatives of panic patients with respiratory abnormalities (Perna et al. 1996), suggesting that hypersensitivity to CO_2 inhalation may be a trait marker for panic disorder rather than a state marker. These data suggest that parents with panic disorder may transmit a diathesis for certain forms of anxiety that is observable in the respiratory system (Pine 1999), which may remain latent (Coryell 1997) or may be variably expressed across development as extreme separation distress during childhood or as panic attacks after puberty (Klein 1993; Panksepp 1998; Pine 1999, 2000). Nevertheless, the limited available work among childhood offspring of parents with panic disorder does not support these possibilities. Namely, healthy pediatric offspring of parents with panic disorder show normal responses to CO_2 inhalation (Pine et al. 2005).

These physiological investigations, along with evidence of strong familial associations, provide evidence that childhood SAD and adult panic disorder share a biological substrate (Pine 1999). Similarly, both SAD in children and panic disorder in adults are associated with abnormalities within the noradrenergic system that may contribute to risk for these conditions (Sallee et al. 2000). One major question concerns inconsistencies in the data for physiological measures. For example, innate hypersensitivity in chemoreceptors should be reflected in physiological responses to stimuli such as CO_2, but the data are more consistent for subjective than for ventilatory indices of CO_2 hypersensitivity. Ventilatory measures may be used to refine understandings of *endophenotypes*, categories that refer to more homogenous groups of individuals with a disorder or a latent risk for a disorder. Despite these limitations, research in this area has significantly advanced conceptualizations of biological markers in anxiety disorders.

Genetic Factors and Psychopharmacology

Given that early life processes can have long-term implications related to the pathophysiology of anxiety disorders, understanding genetic risk factors has become increasingly important. Lau and Pine (2008) explore mechanism through which genes are expressed in a review of gene–environment interactions on pediatric anxiety. Specifically, genes confer an increased vulnerability to environmental stressors that can contribute to

changes in neural circuitry. Individuals with greater genetic vulnerability can establish atypical neural responses when faced with stressors. Over time, atypical connections can contribute to psychological deficits in threat identification and appraisal, which can result in clinical symptoms of anxiety. Although several criteria have been explored to determine endophenotypes, the genes involved in pediatric anxiety disorders remain largely unknown. Still, the identification of genetic markers holds the promise of informing predictive models and disorder-specific nosology, as well as treatment protocols for pediatric anxiety disorders.

One candidate genetic marker extensively studied by Hariri and Weinberger (2003) is a functional polymorphism in the promoter region of the serotonin transporter gene (5-HTTLPR). Genetic variation in *5-HTTLPR*, namely having at least one copy of the short allele, has been found to impact many aspects of functioning including anxiety traits. Neuroimaging studies have found that healthy adult and pediatric subjects with at least one short allele have increased amygdala responses to fearful stimuli as compared to those with the typical alleles, similar to anxious youth. In one study, this difference accounted for roughly 20% of the total variance in the amygdala response to fear stimuli (Hariri et al. 2003). These findings provide evidence for a genetic basis of the brain's response to environmental stressors. Interestingly, some evidence suggests that different associations between *5-HTTLPR* and amygdala function manifest in healthy and anxious children and adolescents (Lau et al. 2009). Future studies are needed to determine the developmental impact of *5-HTTLPR* alleles by utilizing longitudinal models and exploring gene–environment relationships in pediatric populations, including both healthy and anxious individuals.

In light of these genetic findings, attention has focused on the role of serotonin in anxiety disorders. Although studies in adults reveal some evidence of serotonergic abnormalities, virtually no research in this area has been extended to pediatric populations (Charney & Bremner 2001; Pine 1999). Emerging data from therapeutic studies, however, do strongly implicate the serotonergic system in both adult and pediatric anxiety disorders. The selective serotonin reuptake inhibitors (SSRIs) effectively treat virtually all forms of adult anxiety. Studies of pediatric anxiety disorders have demonstrated positive effects of fluvoxamine for children and adolescents with social phobia, SAD, or GAD (Research Unit on Pediatric Psychopharmacology Anxiety Study Group 2001). More recently, a large multisite study compared the efficacy of sertraline, cognitive-behavioral therapy, the combination of sertraline and cognitive-behavioral therapy, and a pill placebo in 488 children (aged 7–17) with diagnoses of GAD, social phobia, and SAD (Walkup et al. 2008). Sertraline led to significant improvement in 54.9% of those treated with this medication versus placebo at 23.7%. The combination of cognitive-behavioral therapy and medication was the most effective at reducing symptoms (80.7% of those treated). These findings are consistent with those of adult studies and provide evidence of serotonin involvement in the pathophysiology of pediatric anxiety disorders.

Studies have also examined the putative role of norepinephrine (NE) systems. The evidence currently indicates a complex dysregulation of NE levels and locus coeruleus firing that may lead to increases or decreases in NE release coupled with altered sensitivities of the pre- and postsynaptic receptors (Ressler & Nemeroff 2000). Adults with GAD, panic disorder, obsessive-compulsive disorder, or social phobia exhibit a blunted growth hormone (GH) response to clonidine challenge, interpreted as a subsensitivity of central α_2-adrenergic postsynaptic receptors (Charney & Bremner 2001). Adults with panic disorder or post-traumatic stress disorder, in contrast, exhibit hypersensitivity to challenges with yohimbine, an α_2-adrenergic antagonist. Such hypersensitivity is manifest most consistently as increases in subjective anxiety, and it appears specific to these two conditions, as opposed to GAD or major depression. Interestingly, Sallee et al. (1998) reported *enhanced* GH secretion to clonidine challenge in children with anxiety disorders, findings that run counter to data from adults that document blunted GH response. More consistent with data in adult panic disorder, a subsequent study by Sallee et al. (2000) found enhanced anxiety responses to yohimbine, as well as blunted GH response in children with anxiety disorders, particularly among children with SAD.

CONCLUSION

Future Directions

Although recent studies have informed our understanding of the pathophysiology of pediatric anxiety disorders, this area of research remains in its infancy. The evolving development of neuroimaging tools and techniques to investigate neural substrates promises to advance this area of research. Similarly, advances in the basic neuroscience of anxiety and molecular genetics may offer unique opportunities to understand the transmission of these disorders across generations. Of paramount importance is the need to integrate research efforts to address issues of developmental vulnerability and to explore factors associated with resilience. For example, considerable work in rodents implicates the prefrontal cortex in regulation of fear responses. Perhaps the most extensive work in this area demonstrates the capacity of medial prefrontal regions to impact amygdala function in the service of fear extinction

(Quirk & Muell 2008). Development of prefrontal functions in humans may allow children and adolescents to undergo similar forms of fear extinction, accounting for the fact that most pediatric anxiety disorders remit. Similarly, persistent anxiety disorders may be those in which this normal developmental capacity fails to mature. As such, failure of prefrontal regions to effectively down-regulate amygdala hyper-responsivity, and interrupt patterns of chronic amygdala activation may account for the development of an anxiety disorder in children at risk. Novel therapies may be those that correct this underlying incapacity (Pine et al. 2009). Longitudinal studies combining neurobiological, neuroimaging, genetic, and behavioral measures are needed to test these pathophysiological models, with the ultimate goal of advancing early identification, preventive intervention, and effective treatment efforts.

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