Behavioral Inhibition: A Neurobiological Perspective

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Behavioral inhibition (BI) during early childhood has been associated with subsequent development of anxiety disorders. However, understanding of the neuroanatomical substrates of BI in humans generally has not kept pace with that of anxiety disorders. Recent interpretations and implementations of Gray's and Kagan's concepts of BI are examined from the perspective of current neurobiological models. Particular attention is given to evidence pointing to conceptual and operational limitations of self-report scales purported to measure trait BI in adults, and especially to inconsistent correlations between such behavioral inhibition system (BIS) scores and amygdala and autonomic responses to fear- or startle-inducing stimuli. Evidence showing a dissociation of both BI and trait anxiety from the amygdala is considered. Possible reasons for the poor association between BIS and trait anxiety self-report scale scores and predicted physiological outputs of the BIS are identified. Reasons to distinguish between the neural bases of BI as against trait anxiety also are discussed. The need to critically examine the role of the amygdala in BI and trait anxiety, as well as to consider other brain areas that appear to be involved in subserving these emotional traits, is emphasized.

Introduction

Behavioral inhibition (BI) has been found to be a risk factor for anxiety disorders in several studies [1•–3•,4••,5•]. Recent research into the functional anatomy of anxiety disorders points to abnormalities in a number of cortical and subcortical structures, as well as to possible abnormal interactions between these levels [2•]. Although there have been developments in understanding the functional anatomy of BI, this literature is fragmented in at least two important ways: First, animal and human studies are poorly integrated, and second, childhood BI is measured according to behavioral criteria, whereas adult BI is measured by a variety of fear-, anxiety-, and punishment-related self-report scales. This review examines existing models and recent findings regarding the neurobiology of BI against the backdrop of these "fault lines" in the literature.

What is BI?

BI is a temperament identifiable in early childhood and is characterized by a stable pattern of fearful feelings and inhibited behavioral responses to social and nonsocial stimuli. BI is measured in young children by observation under laboratory conditions designed "to discriminate between children who experience distress to novelty and those who do not" [6]. Based on a very limited number of studies, the incidence of BI measured in this way is reported to be between 10% and 20% [2•,7] and is considered to be "moderately" stable up to age 7.5 years [1•]. There is evidence for a genetic component to BI, especially in its extreme forms [1•], and behavioral and physiological [5•] similarities between childhood BI and anxiety may suggest a link between BI and anxiety disorders, especially social anxiety disorder (SAD) [2•,3•]. For example, BI is characterized by socially avoidant behavior, longer speech latencies, and high sympathetic nervous system activity, all features of SAD [3•].

Specific research, especially longitudinal research, into the association between BI and anxiety disorders is limited [3•]. Several studies have found an association, but a critical analysis of this literature by Turner et al. [7] identified several significant methodological and interpretive weaknesses and questioned the validity of drawing strong conclusions on these grounds. Subsequently, however, a unique longitudinal study involving subjects aged 21 to 31 months through adolescence by Schwartz et al. [8] found that the association with BI was significant for generalized social anxiety (stronger for girls than for boys) but not for specific fears, separation anxiety, or performance anxiety. Lastly, as not all BI develops into SAD and not all SAD follows BI, it has been suggested that BI constitutes a vulnerability factor for SAD [5•,9•].

There is also growing work on how internal and external factors interact with BI. Extrinsic variables such as peer, parenting, and attachment relationships and socioeconomic conditions in interaction with BI have a significant impact on emotional development [1•,3•,5•]. Equally, internal processes such as cognitive development combine with temperament to influence behavior. For example, Rothbart et al. [10] define "effortful control" (EC) as the ability to inhibit a dominant response in order to perform a subdominant response. EC, which emerges at age approximately 24 months and progressively develops in strength through late infancy and beyond, provides the developing BI child with self-regulatory powers to override inhibitory fear responses and approach or tolerate the fear-inducing stimulus. The authors reason that the outcome depends upon the strength of the dominant response, which is one likely explanation for the stability of extreme BI relative to less extreme BI. Nevertheless, EC draws attention to the early onset of the distinction between automatic unregulated responses to sensory stimuli mediated by subcortical reacting systems present from birth and self-willed, attention-dependent executive responses necessary for EC mediated by anterior cortices, which only begin to develop after 6 months.

Two theories of BI

Historically, BI has been defined from neurological and behavioral perspectives. As a neurological construct, BI can be traced back to Gray [11], who posited the existence of two orthogonal neural systems, the behavioral inhibition system (BIS) and behavioral activation system (BAS). Gray identified the BIS with anxiety and the BAS with impulsivity and proposed that stable unconditioned (ie, trait-like) individual differences in the fundamental sensitivity of the BIS and the BAS to emotionally salient conditioning stimuli independently bias emotional learning, thereby defining temperament. Gray originally identified the septo-hippocampal system (SHS) as the seat of the BIS, and inputs into the BIS were defined as (1) conditioned fear stimuli (which signal that a response will bring punishment), (2) novel stimuli (signaling possible punishment), and (3) frustrative nonreward stimuli (situations in which ongoing goal-oriented behavior is not bringing success) [12]. In an updated model, inputs are any stimuli that generate conflicting response tendencies, such as a simultaneous motivation to approach and avoid a stimulus [12,13•]. In response to such conflicts, the outputs of the BIS are (1) stopping of current behavior (ie, BI); (2) reallocation of attention; and (3) in the case of threatening stimuli, increased arousal (which is associated with increased startle response [SR]—see below).

According to the original theory, the SHS is the neural substrate of the BIS, and experiments with rats showed that this system is activated by theta input originating from the brainstem and septal area [12]. Theta input is modulated by permissive gating pathways that, when open, lead to the BIS outputs mentioned above. A more recent addition to this model has been the amygdala. Amygdala activation in response to perceived threat increases theta and permissive theta gating, thereby increasing SHS processing, which in turn further stimulates the amygdala and increases arousal and attention [12]. The latter causes increased perception of threat and thus increased theta, making for a positive feedback loop with a gain determined by the degree of threat.

Evidence in support of the SHS-amygdala model of the BIS has been derived mainly from studying the effects of drugs and brain lesions on theta activity and behavior in rodents. Only clinically effective anxiolytic drugs such as barbiturates, benzodiazepines, buspirone, imipramine, and fluoxetine inhibit theta input into the SHS, whereas nonanxiolytic drugs do not [14 and McNaughton, Personal communication]. During certain behaviors, the supramammillary nucleus regulates the frequency of theta activity, and Aranda et al. [15] recently found that lesions of the supramammillary nucleus in rats led to reduced BI. Very few direct tests of the updated Gray-McNaughton BIS model in humans exist, but a recent electroencephalogram (EEG) study by Moore et al. [16] did find increased theta power and coherence at points of conflict, as well as during response in a go–no-go type of task.

Somewhat later, BI was identified as a psychological–behavioral construct by Garcia-Koll et al. [17] as "BI to the unfamiliar," which refers to "the child's initial behavioral reactions to unfamiliar people, objects, and contexts or challenging situations" [18]. BI children are characterized as being either avoidant of unfamiliar situations, objects, and people, or, when exposed to such stimuli, they stop playing, become watchful, and tend to retreat toward their mothers [1•–3•,4••,5•]. Childhood BI earned construct validity as a temperament type by virtue of evidence of its longitudinal stability and heritability, as well as its identification as a possible risk factor for psychopathology later in life. Drawing on advances in understanding the role of the amygdala in fear conditioning in rats, largely attributable to Ledoux [19] and Davis [20], during the 1980s [1•], Kagan and Snidman [21] proposed that individual differences in BI reflect differences in amygdala reactivity to novel stimuli.

Measuring BI

As research into the neurobiology of BI in humans has, with very few exceptions [1•], all been done in adults, some issues and recent developments regarding the operationalization of the BI construct in adults needs to be examined. Kagan's childhood BI is measured under laboratory conditions. In these experiments, the child is exposed to a standardized set of novel social (eg, a stranger entering the room in which the child and mother are otherwise alone) and nonsocial stimuli (eg, being requested to dip his or her finger into small cups containing either water or red or black liquid). A "fear score" of the child's behaviorally inhibited states

Table 1. Factor analysis loadings of "anxiety" items for 5 different self-report scales

Scale/subscale	Loading
SP(SR)Q	0.81
CW-BIS	0.72
STAI-T	0.88
TCI-HA	0.83
Eysenck Personality Scale (neuroticism subscale)	0.84

CW-BIS—Carver and White Behavioral Inhibition System subscale; SP(SR)Q—Sensitivity to Punishment, Sensitivity to Reward Questionnaire; STAI-T—Spielberger State Trait Anxiety Inventory-Trait Version; TCI-HA—Tridimensional Character Inventory-Harm Avoidance subscale.
(*Data from* Caseras et al. [23].)

(ie, the amount he or she fails to engage with the social or physical stimuli) then is taken as a measure of the child's trait BI. BI in adults, on the other hand, uses self-report questionnaires to measure trait fear/anxiety/punishment sensitivity and does not include any measurement of state BI. In short, childhood BI is a measure of behavior interpreted as a "fear score," and in adults, self-report measures of fear/anxiety are interpreted as a measure of trait behavioral inhibition. Gray's SHS-amygdala BIS, Kagan's BI construct, and self-report BIS scores in adults are therefore conceptually related but operationally three very different entities. Indeed, there is evidence in the recent literature (1) pointing to a neuroanatomical dissociation between trait anxiety and the amygdala and (2) questioning the validity of the adult "trait BI" construct as measured by self-report scales. This evidence is discussed in the following section.

Recent Developments

Measuring BI

Carver and White (CW) [22] specifically developed a BIS/BAS scale to measure these dimensions of Gray's theory, and the CW-BIS subscale has been widely and for the most part uncritically used as a measure of Gray's BIS sensitivity in adults. However, there is evidence of limitations of the CW-BIS/BAS scale, the most pertinent being (1) the BIS and BAS components are not orthogonal [23] and (2) in at least some samples, a failure of factor analysis to obtain a significant fit to the data [24]. More recently, Torrubia et al. [25] developed the Sensitivity to Punishment, Sensitivity to Reward Questionnaire (SPSRQ) in an attempt to improve upon the CW-BIS/BAS scales. Caseras et al. [23] used factor analysis to compare several personality scales, and the scales in Table 1 scored highly on a single "anxiety" factor.

SP(SR)Q [25,26], CW-BIS, Spielberger State Trait Anxiety Inventory (STAI-T) [27], and Harm Avoidance (HA) [28] subscales have all been used as a measure of BIS sensitivity and trait anxiety in very similar ways. Yet despite high common loadings, correlation coefficients between these anxiety scales are typically only approximately 50% [23], and the factors accounting for the similarities and differences between these scales are not well defined. Two studies found the SP(SR)Q to suffer from limitations similar to those for the CW-BIS [24,29]. Yet aside from these problems with self-report scales, for the purposes of this review, it is pertinent to ask to what extent self-report instruments purporting to measure BIS sensitivity and trait anxiety in general are supported by neurobiological evidence, and how these two entities exist in relation to each other. These issues are addressed in the following section.

Neurobiology

Skin conductance level (SCL) and eye-blink SR are autonomic responses that have been investigated as outputs of the BIS. A full discussion of these topics is beyond the present scope, but a few recent articles are relevant insofar as they suggest conceptual and operational limitations of self-report measures of the BIS.

Hofmann and Kim [30] studied changes in SCL from baseline during an impromptu speech task in 55 males who had previously scored greater than 65% on a public-speaking anxiety scale. Scores for this sample on the Social Avoidance and Distress Scale (SADS) and Personal Report of Confidence as a Speaker (PRCS) were similar to those from SAD samples, and STAI-T scores fell within the ranges reported for panic disorder and SAD. In contrast, CW-BIS scores were within the normal range. Results revealed a significant correlation between SCL and STAI-T scores, but not between SCL and CW-BIS scores, possibly suggesting a fundamental difference between the self-report trait BIS construct and general trait anxiety (as measured by the STAI-T) with regard to a physiologic component of anxious arousal. The electrodermal system is regulated by a diffuse system of cortical, subcortical, and brainstem areas [31]. Among these, the amygdala exerts an excitatory influence on the SCL, which raises theoretical questions about its involvement with SCL changes, trait anxiety (STAI-T), social anxiety (SADS), and public speaking fear (PRCS) on the one hand, but not with CW-BIS scores on the other. The authors suggest that their discrepant results may be due to the presence of items in the STAI-T but not in the CW-BIS scale, which measure general affective tone, and that these items are possibly sensitive to physiologic aspects of the BIS. This possibility is returned to further below.

The eye-blink component of the startle reflex is a physiologic variable modulated by emotional stimuli. Negative/aversive stimuli cause an increase in eye-blink magnitude, whereas positive/appetitive stimuli attenuate it. This "affect-modulated SR" is absolutely dependent upon the integrity of the amygdala [32] and has been investigated in studies of trait emotion in both normal and psychiatric populations [32,33]. Recently, Fullana et al.

[26] predicted but did not find that individuals at opposite extremes in terms of BIS scores (as measured by SP[SR]Q) would show significant differences in magnitude and time course of fear-potentiated startle that involved the threat of a shock. Previously, Grillon et al. [34] found the same results. As for the SCL results discussed above [30], but much more specifically so, these SR findings question the relationship between amygdala reactivity and BIS sensitivity originally proposed by Kagan and Snidman [21].

There have been several investigations of affect-modulated SR using emotionally negative images as stimuli rather than fear of shock. Corr et al. [28] found that SR potentiation was only evident for individuals with high HA scores. However, Hawk and Kowmas [35] found no SR potentiation differences between individuals with high/low CW-BIS scores. Caseras et al. [36] hypothesize that such inconsistencies may be a result of mixed stimulus content (ie, insufficient care with regard to image valence by not ensuring that fear images were not also provoking feelings of disgust). These authors therefore compared SR potential in subjects preselected for extreme high and low SP(SR)Q using distinct fear and blood-disgust stimulus valences. Results showed that fear caused SR modulation in the high SP(SR)Q group, but not the low SP(SR)Q group, and blood-disgust caused SR modulation in both personality groups. Blood-disgust also elicited greater SR magnitudes than fear. These results indicate a personality–valence interaction effect on SR, thereby possibly explaining some previous inconsistent findings.

Cornwell et al. [37], using a Virtual Reality Public Speaking paradigm, found SR to be linearly related to Fear of Negative Evaluation (FNE) and Self-Statements during Public Speaking (SSPS) scores (both assess predisposition to experience anxiety in social-evaluative conditions) but not associated with general trait anxiety as measured by STAI-T. Thus, there may be a functional distinction in amygdala sensitivity to social and nonsocial anxiety stimuli/contexts.

No or inconsistent correlations between CW-BIS, SP(SR)Q, and STAI-T and hypothesized neural substrates of the BIS (ie, amygdala) as indexed by SCL and SR is therefore a relatively consistent finding (see also [38,39]) and may expose a lack of sensitivity/specificity in these self-report measures of BIS temperament. The suggestion that STAI-T is more sensitive to affective tone and physiologic output of the BIS than the CW-BIS scale [30] is not consistent with the data of Cornwell et al. [37], which showed that STAI-T did not correlate with SCL or SR at baseline or during speech performance. On the other hand, both of these public speaking studies found significant autonomic nervous system correlations with measures of social anxiety, which suggests that the STAI-T may be more sensitive than the CW-BIS to social fears.

Alternatively, evidence of a dissociation between general trait anxiety (whether measured by CW-BIS, SP[SR]Q, or STAI-T) and physiologic indices of BIS activity suggests that the BIS is independent of both the amygdala and other neural pathways involved in the modulation of SR and SCL. In other words, insofar as the BIS is a neural entity hypothesized to account for individual differences in anxious temperament through biases in reinforcement sensitivity to aversive stimuli, it does so without involving the amygdala, which is known to be essential for fear conditioning [18,40]. Nowhere is this more evident than in the failure of Fullana et al. [26] and Grillon et al. [34] to find a correlation between measures of self-report BIS temperament (SP[SR]Q and STAI-T, respectively) and "fear of shock"-potentiated SR, a paradigm that depends upon fear conditioning. At the very least, these results appear to be pointing to a different or more complex relationship between the BIS and the amygdala than originally proposed.

A failure to control for a trait anxiety–image valence interaction as shown by Caseras et al. [36] may be an important insight into resolving some of the inconsistencies between BIS temperament and BIS autonomic response in the emotional image-modulated SR literature. However, it does not apply to "fear of shock"-potentiated SR, in which no images are involved and the lack of an association is particularly stark [26,34]. Can the importance of social anxiety over general anxiety help to explain a dissociation between self-report BIS temperament and autonomic reactivity? Of possible interest in this regard, Cornwell et al. [37] found that subjective state anxiety (measured by STAI-State) correlated with FNE, SSPS, and STAI-T (but not SR) during speech, suggesting that neither "general" trait nor "general" state anxiety mediates the correlation between trait social anxiety and SR found by these authors. Phan et al. [38] also recently reported on a functional MRI (fMRI) study that found significant amygdala hyperactivation to aversive (anger/disgust/fear) facial expressions in 10 subjects with generalized social phobia (GSP), whereas 10 healthy controls subjects showed slight deactivation. The results confirmed several previous studies but also revealed a significant correlation between the blood oxygen level-dependent signal change and symptom severity in the GSP group as measured by the Liebowitz Social Anxiety Scale. However, no correlations with general state or trait anxiety (STAI) measures were found. Taken together, these studies suggest a link between social anxiety as distinct from general trait anxiety and physiologic responsiveness. In addition, the failure to find an association between general trait anxiety and SR in a speech task [37], as well as the observation of amygdala deactivation to aversive stimuli in normal controls [38], is yet more evidence of a dissociation between these kinds of anxiogenic stimuli, which theoretically should activate the BIS and this brain structure.

Questions about the validity of self-report measures of BIS temperament are echoed by Cogswell et al. [24], who caution against the unqualified use of self-report scales, emphasizing both the need to improve their validity and

investigate their behavioral correlates in order to properly examine their theoretical bases.

Davidson [41] also emphasizes the importance of supplementing self-report scores with physiologic observations, and Jackson et al. [42•] used this approach to further investigate the role of the cortex in emotion. In addition to measuring SR *during* the stimulus, which is a measure of emotional reactivity, they also focused on the poststimulus period in order to quantify the SR *following* an affective stimulus, which reflects regulation of the initial emotional reaction. Subject's resting frontal alpha EEG asymmetry, an index of underlying prefrontal cortex (PFC) activity, also was recorded. No significant correlations between SR magnitude *during* stimulus and EEG were found, but SR magnitude *following* picture offset was significantly inversely correlated with increased left frontal cortical activity. Together with other findings, the authors propose that the left PFC inhibits the amygdala both tonically and phasically [42•,43]. These findings support the role of the PFC, a region not originally considered part of the BIS, in the regulation of emotion. However, extensive interaction between the cortex and the SHS is an integral part of the updated version of the Gray-McNaughton theory [12,13•], and in the anxiety literature, the idea that anxiety disorders may be the result of cortical-subcortical dysregulation is emerging [2•,44•].

The only investigations of PFC function in specific relation to BI have been those examining frontal EEG asymmetry. An initial model [45] associated greater right frontal brain activity as indexed by alpha EEG activity with greater self-report BIS sensitivity. However, Harmon-Jones and Allen [46] and Coan and Allen [47] both found no significant association between self-report BIS and greater right frontal cortical activity. Hewig et al. [48] also recently found no evidence for an association between right frontal activity and self-report BIS scores but did find self-report BAS to be correlated with both left- and right-sided activity. On the other hand, there is good evidence for an association between trait right frontal cortex activity and SAD in humans [49] and anxious temperament in nonhuman primates [50••].

There is another possible explanation for the negative results for correlations between BIS/trait anxiety self-report scales and BIS physiologic arousal. Experimenting with rhesus monkeys, Kalin et al. [50••] found positive evidence indicating that differences in amygdala response to fear stimuli do not mediate differences in anxious temperament. In these investigations, selective fiber-sparing ibotenic acid lesions of the amygdala blunted unconditioned fear responses to snakes but spared unconditioned trait-like anxiety/fear responses. These studies deserve careful attention because of the following:

- A validated ethological definition of a trait-like anxious phenotype was used: Chronically fearful/anxious monkeys have exaggerated unconditioned fear responses present from age 3 months. This phenotype also is associated with extreme right-sided EEG asymmetry.

- Anxious temperament is assessed by observing the monkey's response to a human intruder, which closely resembles the methods used to assess BI in children.

- The results stand in contrast to the effects of earlier "classical" results of non–fiber-sparing amygdala ablation experiments in nonhuman primates, which reported obvious changes in personality and a "taming effect." In this study, monkeys that showed marked freezing and hostile behavior presurgery behaved identically postsurgery. EEG asymmetry scores also did not differ pre- and postsurgery.

- Two independent studies that found very similar results using two different fiber-sparing methods of destroying the amygdala are cited.

Interpreting their results, the authors first note the difference between the sparing in the monkeys of *unconditioned* trait-like anxiety and amygdala lesion experiments in rodents that block *conditioned* fear responses. In addition, they draw attention to other rodent studies that indicate that although the amygdala mediates immediate responses to fear stimuli, other areas, such as the bed nucleus of stria terminalis (BNST), have been implicated in mediating *longer-term*, *nonspecific* anxiety. The authors conclude that the observed anxious temperament is not mediated by the amygdala [44•,50••], although they cite evidence supporting an important developmental role for the amygdala in the acquisition and expression of anxious responses.

It is noteworthy in this respect that a recent study by Schwerdtfeger [51] found significant correlations between STAI-T scores and autonomic responses (ie, heart rate [HR] and SCL) in a fear-conditioning paradigm in which happy and sad smileys (CS) were consistently followed by nonthreatening and threatening pictures (UCS), respectively. High-anxiety individuals showed significantly higher HR accelerations in response to nonthreatening CS and significantly greater HR decelerations in response to threatening CS than low-anxiety subjects. High-anxiety individuals had significantly higher SCL magnitudes to the CS irrespective of threat valence, as well as to the UCS, but only when it was threatening.

As previously mentioned, fear conditioning inherently implies amygdala involvement [18,40]. Although at first glance, these results may appear to forge a link between trait anxiety and fear-conditioning sensitivity, upon closer inspection, the HR and SCL differences between the high- and low-anxiety groups were consistently observed

right from the beginning of the tasks. Hence, although the results show in an *apparent* fear-conditioning experiment clear autonomic differences that correlate with trait anxiety differences, the differences appear to have nothing to do with fear conditioning itself. Therefore, the correlation between STAI-T scores and autonomic responsiveness to fear stimuli does not appear to depend upon differences in sensitivity of the "amygdala as BIS" to fear conditioning. Instead, these findings are consistent with the sparing of unconditioned trait anxiety in amygdala-lesioned monkeys [50••].

Another noteworthy feature of this study is that in order to ensure that subjects were paying attention, they were required to locate a small target hidden equally in some of the threatening and nonthreatening images. High-anxiety subjects found fewer targets in threatening but not in nonthreatening images than did low-anxiety subjects. Overall, the author's preferred interpretation of the data is that the heightened autonomic indices seen in high-anxiety subjects reflect increased allocation of attentional resources (for target spotting) to forthcoming threatening stimuli because these subjects' attention is more readily distracted from this goal by the threatening content of the image. As arousal and attention are outputs of the BIS [13•], Schwerdtfeger's interpretation is supported by theory.

Second, based on the correlation between right-sided EEG asymmetry and observed anxious temperament, which both survived amygdala destruction, Kalin et al. [50••] propose a mediating role for PFC in trait anxiety, and a recent prospective study by Blackhart et al. [52] did, indeed, find a significant association between right-sided EEG asymmetry and trait anxiety (STAI-T).

In a more recent study of rhesus monkeys, Kalin et al. [53••] utilized positron emission tomography and ^{18}fluoro-deoxyglucose to measure brain activity across two conditions that elicited different degrees of freezing behavior (BI). Notably, activity in the BNST, as opposed to the amygdala, correlated with duration of freezing in both conditions. However, subtraction of brain activity between conditions did not show a correlation between BNST activity and freezing duration. Instead, differences in cingulate cortex, thalamic areas, and the dorsal raphe nucleus were found to correlate with differences in freezing duration between conditions—further evidence dissociating the amygdala from, but implicating PFC in, observed trait BI. It is noteworthy that whereas SAD in humans and anxious temperament in rhesus monkeys are associated with extreme right-sided EEG asymmetry [49,50••], no correlation between activity in this region and BI was found. This is logical because left- and right-sided EEG asymmetry are (1) both associated with the BAS [48] and (2) associated with approach and withdrawal, respectively [48], whereas BI by definition is *not* active and, depending upon the situation, may reflect passive approach *or* passive withdrawal/avoidance.

fMRI and BI

In keeping with the concerns about self-report scales previously discussed, the small number of recent studies concerning individual differences in the biological substrates of BI temperament all are notable for the complete absence of BIS self-report scales (relying instead on laboratory observations) [4••,6,39,54]. Two of these appear to be the only fMRI studies investigating amygdala responsivity in relation to BI. Pursuing Kagan's overreactive amygdala idea, Schwartz et al. [4••] compared fMRI amygdala activity in 22 adults who had been assessed as BI (13) or uninhibited (9) in the second year of life. They found that amygdala activity elicited by novel stimuli but not by familiar stimuli was significantly greater in the BI group. Among the inhibited group, amygdala activity in two subjects with a diagnosis of generalized SAD did not differ from that of the rest of the group, suggesting that although this amygdala response may be an endophenotype [55] for BI, it is not specific to BI–SAD versus BI–non-SAD. In the other fMRI study, Bertolino et al. [39] investigated amygdala and SCL responses of normal individuals to a matching task involving angry and fearful face images in relationship to 5-HTTLPR polymorphisms and a measure of trait anxiety called "phobic prone." Phobic proneness was independently assessed by two investigators highly experienced in using a validated semistructured interview instrument. Results showed genotype and personality type could independently predict amygdala activity during the emotional faces matching task. Once again, no correlations between trait anxiety and SCL during the task were found.

Genetic factors

Smoller et al. [54] found significant associations between childhood BI in children of parents diagnosed with panic disorder and a haplotype comprising three single-nucleotide polymorphisms in and around the corticotropin-releasing hormone (CRH) gene. Moehler et al. [6] found blond hair pigmentation to be significantly associated with childhood BI in a sample of 101 German toddlers. Proopiomelanocortin is co-produced with CRH in the skin, and because amelanocytic hair follicles produce higher levels of CRH, it is possible that the elevated CRH levels found in childhood BI cause the brain changes underlying BI (rather than elevated CRH being the result of increased brain-pituitary-adrenal activation in the BI brain).

Finally, Fox et al. [9•] found a gene–environment interaction between the short 5-HTTLPR allele gene and social support in relation to the risk of childhood BI, but Hirschfield-Becker et al. [56] found that psychosocial adversity did not explain the risk for BI in children of parents with panic disorder. These studies are no doubt among the first steps toward understanding the complex gene–environment interactions that bear upon the phenotypic expression of temperament, and in turn upon

temperament as a diathesis for illness. None of these recent genetic studies utilized self-report scales.

Conclusions

A number of recent studies can be singled out for having added credibility to the notion of BI as a biologically mediated temperament type [4••,6,9•,39,53••,54,56] that constitutes a risk factor for anxiety disorders [8]. What makes these studies particularly strong is the fact that they did not depend upon self-report scales to measure BI. In all these studies, apart from one case in which a structured interview was conducted [39], BI was measured using behavioral criteria.

Although some of these studies found an association between BI and amygdala activity in humans [4••,39], one study of rhesus monkeys did not, implicating brainstem, thalamic, and cortical areas instead [49]. A separate study of rhesus monkeys provides compelling evidence against the amygdala as the substrate of trait anxiety [44•,50••].

Several recent studies investigated the biological correlates of the BIS and trait anxiety as measured by a variety of self-report scales [26,30,35–39,48,51]. On the whole, these studies failed to find a consistent association between BIS/trait anxiety scores and autonomic responses to stimuli or tasks designed to activate the BIS [26,30,35–39,48,51]. In addition to the evidence of dissociation between BI and trait anxiety on the one hand and the amygdala on the other, there is evidence of neural dissociation between BI and trait anxiety. Right-sided EEG asymmetry is associated with trait anxiety [50••,52] and SAD [49] in humans and with trait anxiety in rhesus monkeys [50••] (but not with BI) in either species [46–48,53••]. Therefore, from this perspective, there appears to be a dissociation between the BIS and trait anxiety at the cortical level, which suggests a neuroanatomical distinction between the BIS and the neural substrates that activate the BIS, which is a feature of the Gray-McNaughton BIS model [11,12,13•].

Taken together, the results of all the studies discussed here reveal a poor understanding of the neural basis of BI in humans, as well as its relationship with trait anxiety. This may stem from a failure in the conceptual and operational translation of Gray's BIS in rats and Kagan's BI in children to adult humans, in whom most functional neurophysiologic and neuroimaging investigations have been performed. Conceptually, Gray's original hypothesis that individual differences in the degree to which classical conditioning stimuli elicit BI from the BIS has frequently, implicitly or explicitly, been taken to mean that the core substrate of fear conditioning, the amygdala, is the BIS. From this, it is frequently assumed that questionnaires and tasks designed to tap fear/anxiety sensitivity are tapping the CNS substrates of the BIS and therefore of BIS sensitivity. Operationally, this has entailed the use of self-report "BIS" scales possibly lacking in specificity and sensitivity, as well as the use of experimental paradigms and fear-inducing stimuli that appear to be too nonspecific. For example, factors such as social versus nonsocial anxiety [30,37,38], temperament–valence interactions [36], or attentional demands [51] appear to be exerting significant confounding effects. There also has been, with few exceptions [50••,53••], a failure to consider cortical or subcortical substrates of BI and trait anxiety other than the amygdala.

In conclusion, recent investigations into the biological correlates of supposed measures of BIS sensitivity in adults such as the CW-BIS, SPSRQ, and other measures of trait anxiety are more or less consistently returning negative results, thereby bringing the biological meaning of these scales into focus. Three possibly important confounding factors are identified: social versus nonsocial anxiety, subject–condition interactions, and attentional demand. Nevertheless, perhaps in recognition of these problems, there appears to be a trend away from the use of self-report scales in studies investigating neural, genetic, and other biological correlates of the BIS [4••,6,9•,39,54]. Future studies aimed at characterizing the neural correlates of BI thus should either not rely upon self-report scales or else strive to control for these and other possible confounding factors [57•]. In addition, evidence pointing toward BI and trait anxiety being distinct entities with distinct neural substrates should be considered. Last, there should be an effort to formulate and test hypotheses about the neuroanatomical substrates of the BIS in humans that may or may not include, but certainly should extend beyond, the amygdala.

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References and Recommended Reading

Papers of particular interest, published recently, have been highlighted as:

- • Of importance
- •• Of major importance

1.• Fox NA, Henderson HA, Marshall PJ, et al.: **Behavioral inhibition: linking biology and behavior in a developmental framework.** *Annu Rev Psychol* 2005, **56:**235–262.

The most recent substantial review of BI from a biological perspective. It includes an intriguing discussion on research into event-related potentials and BI in children.

2.• Tillfors M: **Why do some individuals develop social phobia? A review with emphasis on the neurobiological influences.** *Nord J Psychiatry* 2004, **58:**267–276.

A brief but very clear review of current knowledge regarding cortical and subcortical substrates, roles, and mechanisms underlying fear and anxiety.

3.• Neal JA, Edelman RJ: **The etiology of social phobia: toward a developmental profile.** *Clin Psychol Rev* 2003, **23:**761–786.

Includes a brief review of BI that is outstanding for its fresh and critical perspective of the literature.

4.•• Schwartz CE, Wright CI, Shin LM, et al.: **Inhibited and uninhibited infants "grown up": adult amygdalar response to novelty.** *Science* 2003, **300:**1952–1953.

An fMRI study entailing the follow-up of subjects with childhood BI to adulthood makes this a unique paper in the neurobiology of BI literature.

5.• Marshall J, Stevenson-Hinde J: **Behavioral inhibition: physiological correlates.** In *International Handbook of Social Anxiety: Concepts, Research and Interventions Relating to the Self and Shyness*. Edited by Crozier WR, Alden LE. Chichester, England: John Wiley & Sons; 2001:53–76.

A thorough and insightful biological review of BI.

6. Moehler E, Kagan J, Brunner R, et al.: **Association of behavioral inhibition with hair pigmentation in a European sample.** *Biol Psychol* 2006 [Epub ahead of print].

7. Turner SM, Biedel DC, Wolff PL: **Is behavioral inhibition related to the anxiety disorders?** *Clin Psychol Rev* 1996, **16:**157–172.

8. Schwartz CE, Snidman NS, Kagan J: **Adolescent social anxiety as an outcome of inhibited temperament in childhood.** *J Am Acad Child Adolesc Psychiatry* 1999, **53:**1008.

9.• Fox NA, Nichols KE, Henderson HA, et al.: **Evidence for a gene-environment interaction in predicting behavioral inhibition in middle childhood.** *Psychol Sci* 2005, **16:**921–926.

A recent study examining the interaction effect between the short and long 5-HTT polymorphisms and maternal support on risk for childhood BI.

10. Rothbart MK, Ellis LK, Posner MI: **Temperament and self-regulation.** In *Handbook of Self-Regulation: Research, Theory, and Applications*. New York: Guilford Press; 2004:357–370.

11. Gray JA: *The Neuropsychology of Anxiety: an Enquiry into the Functions of the Septo-Hippocampal System,* edn 1. Oxford: Oxford University Press; 1982.

12. Gray JA, McNaughton N: *The Neuropsychology of Anxiety,* edn 2. Oxford: Oxford University Press; 2000.

13.• McNaughton N, Gray JA: **The neurobiology of behavioral inhibition.** In *Cognitive and Affective Neuroscience of Psychopathology*. Edited by Barch D. Oxford: Oxford University Press; 2006, In press.

A detailed and up-to-date presentation of the Gray-McNaughton neuropharmacologic theory of the BIS and its relation to anxiety disorders.

14. McNaughton N, Gray JA: **Anxiolytic action on the behavioral inhibition system implies multiple types of arousal contribute to anxiety.** *J Affect Disord* 2000, **61:**161–176.

15. Aranda L, Santin LJ, Begega A, et al.: **Supramammillary and adjacent nuclei lesions impair spatial working memory and induce anxiolytic behavior.** *Behav Brain Res* 2006, **167:**156–164.

16. Moore R, Gale A, Morris PH, Forrester D: **Theta phase locking across the neocortex reflects cortico-hippocampal recursive communication during goal conflict resolution.** *Int J Psychophysiol* 2005 [Epub ahead of print].

17. Garcia-Koll C, Kagan J, Reznick JS: **Behavioral inhibition in young children.** *Child Dev* 1984, **50:**915–922.

18. Kagan J, Reznick JS, Snidman N: **Temperamental inhibition in early childhood.** In *The Study of Temperament: Changes, Continuities, and Challenges*. Edited by Plomin R, Dunn J. Hillsdale, NJ: Earlbaum; 1985:53–65.

19. Ledoux JE: *The Emotional Brain*. New York: Simon & Schuster; 1996.

20. Davis M: **Neural systems involved in fear-potentiated startle.** *Ann N Y Acad Sci* 1989, **563:**165–183.

21. Kagan J, Snidman N: **Temperamental factors in human development.** *Am Psychol* 1991, **46:**856–862.

22. Carver CS, White TL: **Behavioural inhibition, behavioural activation, and affective responses to impending reward and punishment: the BIS/BAS scales.** *J Pers Soc Psychol* 1994, **67:**319–333.

23. Caseras X, Avila C, Torrubia R: **The measurement of individual differences in behavioral inhibition and behavioral activation systems: a comparison of personality scales.** *Pers Individ Dif* 2003, **34:**999–1013.

24. Cogswell A, Alloy LB, van Dulmen MHM, Fresco DM: **A psychometric evaluation of behavioral inhibition and approach self-report measures.** *Pers Individ Dif* 2006, In press.

25. Torrubia R, Avila C, Molto J, Caseras X: **The Sensitivity to Punishment and Sensitivity to Reward Questionnaire (SPSRQ) as a measure of Gray's anxiety and impulsivity dimensions.** *Pers Individ Dif* 2001, **31:**837–862.

26. Fullana MA, Caseras X, Riba J, et al.: **Influence of individual differences in Behavioral Inhibition System on the magnitude and time course of the fear-potentiated startle.** *Int J Psychophysiol* 2006, In press.

27. Zinbarg RE, Mohlman J: **Individual differences in the acquisition of affectively valenced associations.** *J Pers Soc Psychol* 1998, **74:**1024–1040.

28. Corr PJ, Wilson GD, Fotiadou M, et al.: **Personality and affective modulation of the startle reflex.** *Pers Individ Dif* 1995, **19:**543–553.

29. O'Connor RM, Colder CR, Hawk LW, Jr: **Confirmatory factor analysis of the Sensitivity to Punishment, Sensitivity to Reward Questionnaire.** *Pers Individ Dif* 2004, **37:**985–1002.

30. Hofmann SG, Kim HJ: **Anxiety goes under the skin: behavioral inhibition, anxiety, and autonomic arousal in speech anxious males.** *Pers Individ Dif* 2006, In press.

31. Dawson ME, Schell AM, Filion DL: **The electrodermal system.** In *Handbook of Psychophysiology,* edn 2. Edited by Cacciopo JT, Tassinary LG, Bernston GG. New York: Cambridge University Press; 2000:200–223.

32. Bradley MM: **Emotion and motivation.** In *Handbook of Psychophysiology,* edn 2. Edited by Cacciopo JT, Tassinary LG, Bernston GG. New York: Cambridge University Press; 2000:602–642.

33. Grillon C, Baas J: **A review of the modulation of the startle reflex by affective states and its application in psychiatry.** *Clin Neurophysiol* 2003, **114:**1557–1579.

34. Grillon C, Ameli R, Foot M, Davis M: **Fear-potentiated startle: relationship to the level of state/trait anxiety in healthy subjects.** *Biol Psychiatry* 1993, **33:**566–574.

35. Hawk LW, Kowmas AD: **Affective modulation and prepulse inhibition of startle among undergraduates high and low in behavioral inhibition and approach.** *Psychophysiology* 2003, **40:**131–138.

36. Caseras X, Fullana MA, Riba J, et al.: **Influence of individual differences in Behavioral Inhibition System and stimulus content (fear versus blood-disgust) on affective startle reflex modulation.** *Biol Psychol* 2006 [Epub ahead of print].

37. Cornwell BR, Johnson L, Berardi L, Grillon C: **Anticipation of public speaking in virtual reality reveals a relationship between trait social anxiety and startle reactivity.** *Biol Psychiatry* 2006 [Epub ahead of print].

38. Phan KL, Fitzgerald DA, Nathan PJ, Tancer ME: **Association between amygdala hyperactivity to harsh faces and severity of social anxiety in generalized social phobia.** *Biol Psychiatry* 2006, **59:**424–429.

39. Bertolino A, Arciero G, Rubino V, et al.: **Variation of human amygdala response during threatening stimuli as a function of 5'HTTLPR genotype and personality style.** *Biol Psychiatry* 2005, **57:**1517–1525.

40. Ledoux JE: **Emotion circuits in the brain.** *Annu Rev Neurosci* 2000, **23:**155–184.

41. Davidson RJ: **What does prefrontal cortex "do" in affect: perspectives on frontal EEG asymmetry research.** *Biol Psychol* 2004, **67:**219–233.

42.• Jackson D, Mueller CJ, Dolski I, et al.: **Now you feel it now you don't: frontal brain electrical asymmetry and individual differences in emotion regulation.** *Psychol Sci* 2003, **14:**612–617.

An investigation of the eye-blink SR in relation to frontal EEG asymmetry notable for the sophisticated use of autonomic responses as opposed to self-report scales in the investigation of individual differences in emotional regulation. One of several studies led by Richard Davidson implicating the PFC in trait emotion.

43. Davidson RJ, Jackson D, Kalin NH: **Emotion, plasticity, context and regulation: perspectives from affective neuroscience.** *Psychol Bull* 2000, **126:**890–909.

44.• van Honk J, Peper JS, Schutter DJ: **Testosterone reduces unconscious fear but not consciously experienced anxiety: implications for the disorders of fear and anxiety.** *Biol Psychiatry* 2005, **58:**218–225.

One of the major brain targets of testosterone is the amygdala. In this study, testosterone administration in normal healthy females reduced unconscious fear as measured by a masked emotional stroop task. However, no differences in cognitive anxiety, tension anxiety, and CW-BIS between placebo and testosterone conditions were found.

45. Sutton SK, Davidson RJ: **Prefrontal brain asymmetry: a biological substrate of the behavioural approach and inhibition systems.** *Psychol Sci* 1997, **8:**204–210.

46. Harmon-Jones E, Allen JJB: **Behavioural activation sensitivity and resting frontal EEG asymmetry: covariation of putative indicators related to risk of mood disorders.** *J Abnorm Psychol* 1997, **106:**159–163.

47. Coan JA, Allen JJB: **Frontal EEG asymmetry and the behavioral activation and inhibition systems.** *Psychophysiology* 2003, **40:**106–114.

48. Hewig J, Hagemann D, Seifert J, et al.: **The relation of cortical activity and BIS/BAS on the trait level.** *Biol Psychol* 2006, **71:**42–53.

49. Davidson RJ, Marshall JR, Tomarken AJ, Henriques JB: **While a phobic waits: regional brain electrical and autonomic activity in social phobics during anticipation of public speaking.** *Biol Psychiatry* 2000, **47:**85–95.

50.•• Kalin NH, Shelton SE, Davidson RJ, Kelley AE: **The primate amygdala mediates acute fear but not the behavioral and physiological components of anxious temperament.** *J Neurosci* 2001, **21:**2067–2074.

An elegant and meticulously performed ethological brain lesion study on rhesus monkeys providing compelling evidence against amygdala as a substrate for trait anxiety.

51. Schwerdtfeger A: **Trait anxiety and autonomic indicators of the processing of threatening information: a cued S1-S2 paradigm.** *Biol Psychol* 2006, **72:**59–66.

52. Blackhart GC, Minnix JA, Kline JP: **Can EEG asymmetry patterns predict future development of anxiety and depression? A preliminary study.** *Biol Psychol* 2006, **72:**46–50.

53.•• Kalin NH, Shelton SE, Fox AS, et al.: **Brain regions associated with the expression and contextual regulation of anxiety in primates.** *Biol Psychiatry* 2005, **58:**796–804.

A unique study showing an association between brainstem, thalamic, and PFC regions, but not the amygdala, and the regulation of freezing behavior (BI) in rhesus monkeys.

54. Smoller JW, Yamaki LH, Fagerness JA, et al.: **The corticotropin-releasing hormone gene and behavioral inhibition in children at risk for panic disorder.** *Biol Psychol* 2005, **57:**1485–1492.

55. Hariri AR, Weinberger DR: **Imaging genomics.** *Br Med Bull* 2003, **65:**259–270.

56. Hirschfield-Becker DR, Biederman J, Faraone SV, et al.: **Lack of association between behavioral inhibition and psychosocial adversity factors in children at risk for anxiety disorders.** *Am J Psychiatry* 2004, **161:**547–555.

57.• Coan JA, Allen JJ, McKnight PE: **A capability model of individual differences in frontal EEG asymmetry.** *Biol Psychol* 2006, **72:**198–207.

This recent semitheoretical paper presents evidence and argues in favor of a "capability" model that takes individual–condition interactions into account and against the general trait or "dispositional" model that doesn't.