

Epistasis between 5-HTTLPR and ADRA2B polymorphisms influences attentional bias for emotional information in healthy volunteers

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1 Epistasis between 5-HTTLPR and ADRA2B 2 polymorphisms influences attentional bias for 3 emotional information in healthy volunteers

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11 Abstract

12 Individual differences in emotional processing are likely to contribute to vulnerability and resilience to
13 emotional disorders such as depression and anxiety. Genetic variation is known to contribute to these
14 differences but they remain incompletely understood. The serotonin transporter (5-HTTLPR) and α_2B -
15 adrenergic autoreceptor (ADRA2B) insertion/deletion polymorphisms impact on two separate but inter-
16 acting monaminergic signalling mechanisms that have been implicated in both emotional processing and
17 emotional disorders. Recent studies suggest that the 5-HTTLPR *s* allele is associated with a negative
18 attentional bias and an increased risk of emotional disorders. However, such complex behavioural traits
19 are likely to exhibit polygenicity, including epistasis. This study examined the contribution of the
20 5-HTTLPR and ADRA2B insertion/deletion polymorphisms to attentional biases for aversive information
21 in 94 healthy male volunteers and found evidence of a significant epistatic effect ($p < 0.001$). Specifically, in
22 the presence of the 5-HTTLPR *s* allele, the attentional bias for aversive information was attenuated by
23 possession of the ADRA2B deletion variant whereas in the absence of the *s* allele, the bias was enhanced.
24 These data identify a cognitive mechanism linking genotype-dependent serotonergic and noradrenergic
25 signalling that is likely to have implications for the development of cognitive markers for depression/
26 anxiety as well as therapeutic drug effects and personalized approaches to treatment.

27 Received 10 June 2011; Reviewed 7 July 2011; Revised 14 July 2011; Accepted 20 July 2011

28 **Key words:** ADRA2B, emotional processing, 5-HTTLPR.

29 Introduction

30 Enhanced processing of emotionally salient information in relation
31 to neutral information is normally considered to be an
32 adaptive process enabling threat detection and in-
33 creasing the probability of survival (Vuilleumier,
34 2005). However, there is considerable evidence that
35 biased processing of emotional information also plays
36 a role in the aetiology and maintenance of emotional
37 disorders such as depression and anxiety (Leppanen,
38 2006). The monoamine neurotransmitters (serotonin,
39 dopamine, noradrenaline) are known to play a

40 significant role in emotional processing and although
41 they are generally considered to act synergistically,
42 few studies have specifically investigated interactions
43 between these neurotransmitters.

44 Serotonin and noradrenaline are also heavily im-
45 plicated in the aetiology of emotional disorders and
46 the majority of therapeutic agents increase synaptic
47 levels of these neurotransmitters (Nutt, 2002). Despite
48 their use for over half a century, it remains unclear
49 how antidepressants exert their therapeutic effects.
50 More recently, evidence has emerged suggesting that
51 serotonergic and noradrenergic antidepressant drugs
52 may act by modifying emotional processing biases
53 (Harmer *et al.* 2009). Yet in spite of this increasing in-
54 sight, the fact that up to 50% of patients treated with
55 these medications fail to respond adequately remains

AQ1

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56 a significant challenge in the management of these
57 disorders (Souery *et al.* 1999). Inter-individual dif-
58 ferences in responses to emotional stimuli may con-
59 tribute to differences in vulnerability to emotional
60 disorders as well as response to therapeutic agents
61 (Hamann & Canli, 2004). It is increasingly accepted
62 that genetic factors explain small but significant
63 amounts of this variability (Todd *et al.* 2011). Thus,
64 polymorphisms in genes involved in serotonergic and
65 noradrenergic signalling represent apposite candi-
66 dates for further investigation.

67 One of the genetic variants that have been most ex-
68 tensively investigated in relation to human emotional
69 processing and emotional disorders is the gene
AQ2 70 encoding the serotonin transporter (5-HTT or
71 SLC6A4). An insertion/deletion polymorphism in the
72 promoter region of this gene (5-HTTLPR) results in two
73 common allelic variants: short (*s*) and long (*l*). The
74 former has been associated with reduced transporter
75 transcription, resulting in approximately 50% re-
76 duction in transporter availability *in vitro* and pre-
77 sumed increased synaptic serotonin availability (Heils
78 *et al.* 1996). More recently, an additional A/G single
79 nucleotide polymorphism (SNP) in the *l* allele (rs25531)
80 has been found to further influence transcriptional
81 activity. The G variant of the *l* allele is considered to
82 result in a reduction in transcriptional efficiency to a
83 level similar to that of the *s* allele (Hu *et al.* 2005;
84 Wendland *et al.* 2006). However, the frequency of this
85 G allele varies with ethnicity and it is relatively un-
86 common in white European ethnic groups (Hu *et al.*
87 2006). Early seminal studies linked the *s* allele to in-
88 creased neurotic personality traits (Lesch *et al.* 1996)
89 and an increased risk of depression in the context of
90 adverse life events (Caspi *et al.* 2003). Subsequent
91 studies examining the *in-vivo* effects of this genetic
92 variation on the phenotypic expression of 5-HTT in the
93 human brain have produced inconsistent results
94 (Praschak-Rieder *et al.* 2007; Reimold *et al.* 2007; van
95 Dyck *et al.* 2004; Willeit *et al.* 2000). However, consist-
96 ent with a number of previous studies, a relatively
97 large recent positron emission tomography (PET)
98 study in healthy volunteers found that polymorphic
99 variation in 5-HTTLPR did not alter expression of
100 5-HTT (Murthy *et al.* 2010). It has, however, been sug-
101 gested that this genetic variation instead contributes to
102 early neurodevelopmental changes that may impact
103 on brain structure and function in later life (Lesch &
104 Gutknecht, 2005). This would be consistent with the
105 further body of functional magnetic resonance
106 imaging (fMRI) literature that has more consistently
107 documented that *s* allele carriers demonstrate signifi-
108 cantly greater amygdala activation in response to

109 aversive, relative to neutral, stimuli in a variety of
110 emotional processing tasks (Bertolino *et al.* 2005; Canli
111 *et al.* 2005; Hariri *et al.* 2002, 2005; Heinz *et al.* 2004;
112 Pezawas *et al.* 2005; for a meta-analysis see Munafò *et al.*
113 2008). Yet, the behavioural implications of these neural
114 differences remained unclear. More recently a number
115 of studies have focused on ‘behavioural endo-
116 phenotypes’ such as selective attentional biases for
117 emotional information. To date, these studies have
118 demonstrated an association between the 5-HTTLPR *s*
119 allele and preferential attention to aversive stimuli
(Beevers *et al.* 2007, 2010, 2011; Fox *et al.* 2009; Osinsky
120 *et al.* 2008), although these reports have not been en-
121 tirely consistent (Caspi *et al.* 2010). One important
122 potential source of inconsistency and non-replication
123 in genetics studies of complex quantitative traits is the
124 issue of polygenicity, including biological epistasis
125 (Moore, 2008). However, none of these neuroimaging
126 or behavioural genetics studies has examined the ef-
127 fects of the other major neurotransmitter system im-
128 plicated in affective spectrum disorders and emotional
129 processing, i.e. the noradrenergic system. 130

131 Noradrenaline has an established role in modulat-
132 ing memory enhancement for emotionally arousing
133 information (McGaugh, 2004) and recent pharmaco-
134 logical challenge studies indicate that it is also in-
135 volved in modulating attentional biases for emotional
136 information in healthy human volunteers (De Martino
137 *et al.* 2008). However, the contribution of genetically
138 influenced differences in noradrenergic tone to inter-
139 individual differences in human emotional processing
140 has been largely unexplored. An insertion/deletion
141 polymorphism in the α_{2B} -adrenergic (auto)receptor
142 gene (ADRA2B) has recently been found to contribute
143 to individual differences in emotionally influenced
144 memory processes. The deletion variant (Del301-303)
145 is associated with decreased agonist-promoted phos-
146 phorylation and receptor desensitization *in vitro*
147 (Small *et al.* 2001), presumed to be associated with in-
148 creased noradrenergic tone *in vivo*. In two seminal
149 studies, de Quervain and colleagues demonstrated
150 an association between this polymorphic variant, in-
151 creased amygdala reactivity and an increased memory
152 bias for emotional stimuli (de Quervain *et al.* 2007;
153 Rasch *et al.* 2009). However, the contribution of
154 ADRA2B to emotionally enhanced attentional pro-
155 cesses was not explored. It therefore remains possible
156 that the observed memory bias arises due to an atten-
157 tional advantage contributing to enhanced encoding of
158 emotional information (Todd *et al.* 2011). The purpose
159 of this study was therefore to test the hypothesis that
160 an increased attentional bias for emotionally arousing
161 information is associated with the deletion variants of

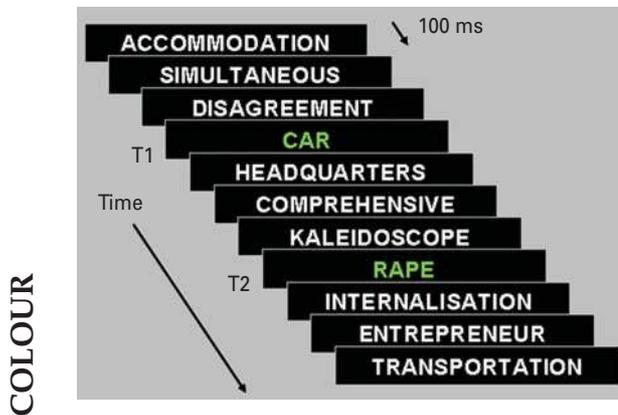


Fig. 1. Schematic representation of attentional blink task.

ADRA2B and 5-HTTLPR and examine whether these effects are subject to additive or non-additive interactions.

Materials and methods

Participants

One hundred and seven healthy white British male volunteers between the ages of 18 and 35 yr (mean \pm s.d. = 24.0 \pm 4.8) were recruited from the university and local community. They had no lifetime history of psychiatric or neurological disorder. Estimates of verbal IQ were derived from the National Adult Reading Test (NART; Nelson, 1982). The study was approved by the local research ethics committee. Following complete description of the study to the participants, written informed consent was obtained.

Behavioural task

We used an emotional attention blink (AB) task based on dual-target rapid serial visual presentation (RSVP) methodology (Raymond *et al.* 1992). Identification of a first target (T1) in a rapid stream of stimuli leads to transient impairment in identification of a second target (T2) – an effect, known as the *attentional blink*. It has previously been used by us and others to demonstrate a bias towards accurate detection of aversive T2 targets compared to neutral (Anderson, 2005; Anderson & Phelps, 2001; De Martino *et al.* 2008; Gibbs *et al.* 2007; Keil & Ihssen, 2004). The task comprised 168 trials, each trial consisting of 13 white distracter words and 2 green target words (T1 and T2) presented sequentially in the centre of a laptop computer screen (see Fig. 1). T1 stimuli were all neutral words averaging 4.8 letters in length. T2 words were derived from the Affective Norms for English Words (Bradley & Lawson, 1999) and half

were aversive-arousing (mean valence and arousal ratings of 2.5 and 7.0, respectively) and half were neutral (mean valence and arousal ratings of 5.1 and 3.5, respectively). Aversive and neutral T2 stimuli did not differ significantly in letter length (mean = 5.1 vs. 4.8, respectively, $p = 0.21$) or written word frequency (mean = 67.1 vs. 87.9, respectively, $p = 0.50$) (Kucera & Francis, 1967). Distracter items were 92 words of longer length (mean letters = 12.5) to facilitate masking of the targets. Each item was presented for 100 ms and was immediately followed by the subsequent item. The lag between the T1 and T2 targets was varied to contain one, three, or five intervening distracters (lag 2, lag 4 or lag 6) with corresponding stimulus onset asynchronies (SOAs) of 200 ms, 400 ms or 600 ms. Participants were instructed to ignore the words in white (distracters) and identify the two green target words (T1 and T2). Responses were made by participants writing down the two targets in any order immediately after each trial on sheets that were subsequently scored. Exact correct spelling was not necessary for a correct response. Vowel and consonant omissions, insertions or replacements were allowed provided the word was recognizable and the spelling was phonologically accurate.

Genotyping

DNA extraction and genotyping for the ADRA2B insertion/deletion polymorphism was performed by KBioscience, Hertfordshire, UK as previously reported (Gibbs *et al.* 2010). For the 5-HTTLPR insertion/deletion, polymerase chain reaction (PCR) was also performed using KBioscience's in-house SNP genotyping system (KASPar[®]) using fluorescently labelled primers (pF1: Cy5.5-CCCAGCGTGCTCCAGAAAC; pR: GGACCTGGGCAGTTGTGC). For technical reasons, we were unable to complete further tri-allelic genotyping of the A/G SNP (rs25531) in the 5-HTTLPR insertion allele.

Statistical analysis

Hardy-Weinberg equilibrium (HWE) of genetic data was assessed by χ^2 analysis. Possible genotype-dependent differences in demographic variables between genotype groups were assessed in a multivariate analysis of variance (ANOVA) with age and IQ as dependent variables and 5-HTTLPR and ADRA2B genotypes as between-subjects factors. Genotype effects on T1 detection in the AB task were examined in a univariate ANOVA with percent correct T1 report as the dependent measure and 5-HTTLPR and ADRA2B genotypes as the between-subjects factors. Affective

Table 1. Demographic characteristics for 5-HTTLPR and ADRA2B genotypes

| | 5-HTTLPR | | | | ADRA2B | | | |
|------------------------|-------------|-------------|----------|----------|-------------|-------------|----------|----------|
| | S | L | F | <i>p</i> | Del | Ins | F | <i>p</i> |
| Number of participants | 34 | 60 | – | – | 51 | 43 | – | – |
| Age, yr mean (s.d.) | 23.9 (4.9) | 23.9 (4.3) | 0.02 (–) | 0.88 (–) | 24.7 (4.9) | 23.0 (4.3) | 1.18 (–) | 0.31 (–) |
| IQ mean (s.d.) | 105.9 (6.5) | 104.4 (7.0) | 2.28 (–) | 0.14 (–) | 106.7 (5.7) | 104.7 (6.4) | 3.01 (–) | 0.06 (–) |

246 modulation of the AB effect was examined in a
 247 repeated-measures ANOVA with the same between-
 248 subjects factors, valence (aversive, neutral) and lag
 249 (2, 4, 6) as within-subject factors and percent correct T2
 250 report (contingent on the correct identification of T1)†
 251 as the dependent measure. Significant interactions
 252 were explored using *post-hoc t* tests. A Greenhouse-
 253 Geisser correction was applied where sphericity as-
 254 sumptions were violated.

255 Results

256 Genotypes

257 Of the 107 participants, ADRA2B genotypes were un-
 258 available for two participants, 11 were homozygous
 259 carriers of the ADRA2B deletion, 48 were hetero-
 260 zygotes and 46 were non-carriers, consistent with
 261 HWE ($\chi^2=0.09$, $p=0.77$). Due to the small number of
 262 homozygous carriers, they were combined with the
 263 heterozygotes, giving two genotype groups of deletion
 264 carriers (Del) and non-carriers (Ins) as previously done
 265 by us and others (de Quervain *et al.* 2007; Gibbs *et al.*
 266 2010). For 5-HTTLPR, 12 genotypes were unavailable,
 267 35 were homozygous *l/l*, 41 were heterozygous *s/l*
 268 and 19 were homozygous *s/s*, consistent with HWE
 269 ($\chi^2=1.18$, $p=0.28$). Given that the *s* allele is considered
 270 to have a dominant effect (Lesch *et al.* 1996), partici-
 271 pants were divided into two groups: homozygous or
 272 heterozygous *s* allele carriers (S group) and non-
 273 carriers (L group) consistent with previous studies
 274 (Canli *et al.* 2005). Participants for whom genetic data
 275 were not available for both polymorphisms were ex-
 276 cluded from further analysis, leaving a total sample of
 277 94. Demographic characteristics (age and IQ) are given
 278 in Table 1. There were no genotype effects on these
 279 variables.

† This is to guarantee that proper attention has been devoted to T1 to ensure an AB effect.

Behavioural data

280
 281 There was a significant main effect of valence on T2
 282 detection accuracy [$F(1, 90)=11.0$, $p=0.001$], with
 283 greater detection of aversive ($92.7 \pm 1.6\%$) compared to
 284 neutral ($81.2 \pm 1.2\%$) words. T2 detection accuracy also
 285 increased significantly [$F(1.2, 109.4)=183.3$, $p<0.001$]
 286 as the temporal lag between the targets increased [lag
 287 2 = $64.4 \pm 22.2\%$; lag 4 = $83.8 \pm 15.7\%$; lag 6 = $91.1 \pm$
 288 12.3%] and there was a significant lag \times valence inter-
 289 action [$F(2, 180)=28.5$, $p<0.001$] such that the
 290 emotional attentional bias was most pronounced at lag
 291 4 (9%) compared to lag 6 (3%) and lag 2 (–3%) where
 292 it was absent. There were no main effects of the indi-
 293 vidual genes; however, there was a highly significant
 294 ADRA2B \times 5-HTTLPR \times valence interaction [$F(1, 90)=$
 295 15.0 , $p<0.001$]. In order to clarify this interaction we
 296 first conducted a separate repeated-measures ANOVA
 297 in the 5-HTTLPR S and L groups with T2 detection as
 298 the dependent variable, valence (aversive, neutral) as
 299 the between-subjects variable and ADRA2B genotype
 300 as the between-subjects variable. We found significant
 301 ADRA2B \times valence interactions in both 5-HTTLPR S
 302 [$F(1, 58)=8.2$, $p=0.006$] and L [$F(1, 32)=6.06$, $p=0.02$]
 303 groups. *Post-hoc* paired *t* tests demonstrated that in the
 304 5-HTTLPR L group, there was a significant attentional
 305 bias for aversive *vs.* neutral T2 words in ADRA2B
 306 deletion carriers [$t(20)=3.0$, $p=0.007$, $d=0.7$] that was
 307 absent in non-carriers [$t(12)=-0.68$, $p=0.504$].
 308 Conversely, in the 5-HTTLPR S group, the significant
 309 emotional attentional bias was absent in ADRA2B de-
 310 letion carriers [$t(29)=0.477$, $p=0.637$] but present in
 311 non-carriers [$t(29)=4.9$, $p<0.001$, $d=1$] (see Fig. 2).
 312 There were no other significant main genotype effects
 313 or interactions in relation to T1 or T2 detection.

314 Discussion

315 The purpose of this study was to examine the
 316 effects of serotonergic (5-HTTLPR) and noradrenergic
 317 (ADRA2B) genetic variants on attentional biases
 318 for aversive stimuli using an attentional blink (AB)

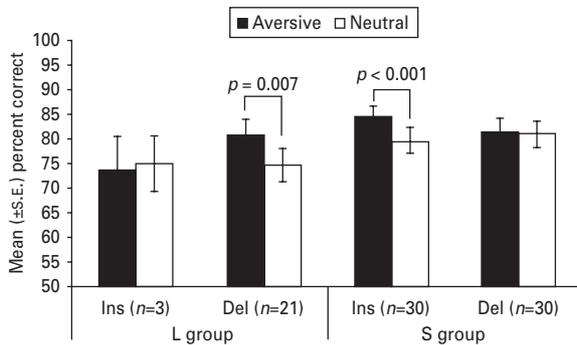


Fig. 2. Accuracy (percent correct) of T2 detection in 5-HTTLPR and ADRA2B groups.

319 paradigm. To our knowledge, this is the first study to
 320 examine the contribution of the ADRA2B insertion/
 321 deletion polymorphism to individual differences in
 322 emotional attentional biases and only the second study
 323 to explore the genetic basis of the emotional AB effect
 324 [Munafò and colleagues previously found an associ-
 325 ation between 5-HTTLPR genotype, smoking status
 326 and detection of smoking-related stimuli in an AB task
 327 (Munafò *et al.* 2005)]. The significant novel finding
 328 from this study is that the affective modulation of T2
 329 detection is influenced by a non-additive (epistatic)
 330 interaction between the ADRA2B and 5-HTTLPR inser-
 331 tion/deletion polymorphisms.

332 Specifically, we found that a significant attentional
 333 bias for aversive compared to neutral information was
 334 present in individuals possessing at least one copy of
 335 the short (*s*) allele of 5-HTTLPR, but only if they did
 336 not carry the ADRA2B deletion. Conversely, the at-
 337 tentional bias for emotional information was only
 338 present in 5-HTTLPR long (*l*) allele homozygotes if
 339 they were ADRA2B deletion carriers. This suggests
 340 that in the presence of the 5-HTTLPR *s* allele which is a
 341 putative risk allele for depressive and anxiety dis-
 342 orders, the negative attentional bias is attenuated by
 343 the ADRA2B deletion variant whereas in the absence
 344 of the *s* allele, the bias is enhanced. Both of these effects
 345 may be related to adaptive processes. For instance,
 346 dependent on 5-HTTLPR genotype, the effect of the
 347 ADRA2B deletion variant may be to either exert a
 348 protective effect against affective spectrum disorders
 349 or facilitate enhanced detection of threat, in both cases
 350 contributing to increased probability of survival.

351 Behavioural genetics implications

352 We did not find a main effect of the serotonin trans-
 353 AQ4 353 porter polymorphism on emotional attention as a
 354 number of previous studies have done (Beevers *et al.*
 355 2007; Fox *et al.* 2009; Munafò *et al.* 2005; Osinsky *et al.*

2008). However, with the exception of Munafò *et al.* 356
 (2005), all of these studies used a variation of the dot 357
 probe task to evaluate emotional biases in selective 358
 attention, rather than the AB task. Although both of 359
 these tasks evaluate selective attention when cognitive 360
 resources are limited, the latter measures deployment 361
 of attention resources under temporal constrains 362
 while the former typically utilizes spatial limitations. 363
 Notably, Munafò *et al.* (2005) also used an alternate 364
 variant of the AB task that indexes attention by estab- 365
 lishing whether the detection of a neutral T2 target is 366
 impaired when preceding an emotionally salient or 367
 neutral T1 target. It is possible that these task-related 368
 differences may account for the difference in findings. 369
 Yet the studies reporting positive associations using 370
 the dot probe task are not without inconsistencies. For 371
 example some have linked the *s* allele to biases towards 372
 aversive stimuli (Beevers *et al.* 2007; Osinsky *et al.* 373
 2008) while others suggest that the *l* allele results 374
 in biases away from negative stimuli (Fox *et al.* 2009; 375
 Kwang *et al.* 2010). Other discrepancies include 376
 5-HTTLPR associations found only with long (Osinsky 377
et al. 2008) or short (Beevers *et al.* 2007) stimulus pres- 378
 entation durations. In spite of using shared dot probe 379
 methodology, there are still significant differences 380
 between these studies in terms of subjects (healthy 381
 volunteers *vs.* psychiatric patients; men *vs.* women), 382
 stimuli (words *vs.* spiders *vs.* pictorial scenes) and 383
 duration of stimulus presentation (<500 ms *vs.* 384
 >500 ms). These differences highlight the need for 385
 task consistency in future studies in order to facilitate 386
 replication (NCI-NHGRI Working Group on 387
 Replication in Association Studies, 2007). 388

The effect of the ADRA2B insertion/deletion 389
 polymorphism on attentional biases for emotional 390
 information has not been previously investigated. 391
 However, it has been suggested that it might contrib- 392
 ute to the emotional memory bias observed in 393
 ADRA2B deletion carriers (Todd & Anderson, 2009). 394
 We did not find any main effect of ADRA2B on 395
 emotional attention in this study suggesting that the 396
 ADRA2B deletion variant does not independently bias 397
 attention towards emotional stimuli but may interact 398
 with other monoaminergic gene systems to contribute 399
 to such bias. 400

401 Behavioural pharmacogenetics implications

402 A number of studies have begun to examine possible 403
 interactions between 5-HTTLPR polymorphisms, 404
 emotional attentional biases and the effects of sero- 405
 tonergic manipulation by acute tryptophan depletion 406
 (ATD) but have thus far failed to produce consistent

findings (Firk & Markus, 2009; Markus & De Raedt, 2011; Markus & Firk, 2009; Roiser et al. 2007). This may in part be due to the fact that ATD in healthy volunteers, independent of genotype, has not produced entirely consistent effects on emotional processing (Hayward et al. 2005; Murphy et al. 2002; Rubinsztein et al. 2001). No studies have as yet examined the possible contribution of genetic variation to the effects of serotonergic and noradrenergic drugs on emotional attention. However, a number of pharmacological studies have examined the effects of serotonergic and noradrenergic drugs on emotional processing in healthy subjects (Arce et al. 2008; Arnone et al. 2009; Brühl et al. 2009; Harmer et al. 2008, 2003, 2004; Murphy et al. 2009a; Norbury et al. 2007; Rawlings et al. 2010). This work may be relevant to understanding the present findings; however, it is difficult to make direct comparisons between genetically and pharmacologically mediated effects on emotional processing as highlighted in the Clinical implications subsection below. Additionally, only three of these studies have specifically examined attentional biases and these have produced relatively inconsistent findings (Browning et al. 2007; De Martino et al. 2008; Murphy et al. 2009b).

Using a dot probe task, Browning et al. (2007) found that the administration of a single dose of the selective serotonin reuptake inhibitor (SSRI) antidepressant citalopram to healthy volunteers resulted in an attentional bias towards positive words (Browning et al. 2007). This is consistent with some of the behavioural findings in relation to the 5-HTTLPR *l* allele described above. On an AB task, De Martino and colleagues found that a single dose of the noradrenaline reuptake inhibitor (NRI) reboxetine boosted detection of emotionally arousing compared to neutral words in healthy volunteers (De Martino et al. 2008). Also on a dot probe task in healthy volunteers, Murphy et al. (2009a,b) found that repeated citalopram administration reduced the attentional bias towards emotional faces, independently of valence, while reboxetine had no effect (Murphy et al. 2009b). These apparently conflicting findings could again be related to differences in methodology (dot probe vs. AB, words vs. faces, single vs. repeated dosing). In fact, there is evidence from both animal and human studies that acute and chronic citalopram administration may differentially influence emotional processing such that acute doses result in an initial increase in the processing of negative information that is attenuated with repeated dosing (Burghardt et al. 2004; Harmer et al. 2003, 2004). However genotype-dependent drug effects may also contribute to these differences. For

example, pharmacological enhancement or attenuation of emotional attentional biases may be less prominent in individuals with genotype-related emotional processing biases or in whom such biases are absent, respectively. If emotional attentional bias is to function as an effective cognitive marker, future pharmacological challenge studies will also need to consider the contribution of genetic variations in the neurotransmitter systems under investigation. There is also an increasing need to evaluate the effects of genetic epistasis between these systems as this may have important clinical implications for the pharmacogenetics of depression and anxiety.

Clinical implications

That such biological epistasis exists between serotonergic and noradrenergic genes, is consistent with the fact that these neurotransmitter systems are intimately connected in the central nervous system (de Boer, 1995). Noradrenergic neurotransmission is modulated by presynaptic inhibitory α_2 -adrenergic (auto)receptors and their blockade increases synaptic levels of noradrenaline. However, there is evidence that serotonergic neurotransmission is also modulated by presynaptic α_2 -adrenergic (hetero)receptors (Clement et al. 1992; De Boer et al. 1994; Mongeau et al. 1993). Yet precisely how these systems may interact to produce the intermediate phenotypes and the clinical disorders themselves remains unclear. The fact that the majority of drugs used to treat affective spectrum disorders act by inhibiting the 5-HTT seems at odds with the fact that individuals with genetically influenced reductions in 5-HTT function have greater risks of developing these disorders, as well as poorer treatment response rates (Lesch & Gutknecht, 2005). This ostensible contradiction is increasingly understood in terms of the complex autoregulatory processes governing serotonergic function (Routledge & Middlemiss, 1996) and the potentially deleterious neurodevelopmental effects of excessive intra-synaptic accumulation of serotonin (Lesch & Gutknecht, 2005). Via its intimate relationship with serotonergic signalling, the ADRA2B polymorphism may also exert its epistatic effects via these autoregulatory and neurodevelopmental mechanisms.

While this hypothesis warrants further investigation, the biological epistasis suggested in the present study may have important implications for individual responses to serotonergic and noradrenergic antidepressant drugs. Most pharmacogenetics studies in depression have focused on variations in 5-HTT (Schosser & Kasper, 2009; Serretti et al. 2007).

511 However, two large recent projects (STAR*D and
 512 GENDEP) have found associations between anti-
 513 depressant response and a number of candidate genes
 514 involved in both serotonin and noradrenaline signal-
 515 ling (Hu *et al.* 2007; McMahon *et al.* 2006; Paddock *et al.*
 516 2007; Uher *et al.* 2009), although none of these studies
 517 included the ADRA2B polymorphism. The GENDEP
 518 project did examine a polymorphism in the related
 519 ADRA2A gene encoding α_{2A} -adrenoceptor subtype
 520 but failed to find any significant effect despite a
 521 previously reported association with the response
 522 to the serotonin-noradrenaline reuptake inhibitor
 523 milnacipran (Wakeno *et al.* 2008). Of note, neither of
 524 the two antidepressants evaluated in GENDEP was a
 525 molecular target of the α_2 -adrenoceptor group.
 526 Thus, the role of polymorphic variation in α_2 -
 527 adrenergic receptors (and their interaction with sero-
 528 tonergic targets) in the therapeutic response of
 529 patients with affective spectrum disorders warrants
 530 further investigation.

531 *Study limitations*

532 The purpose of this study was to investigate epistatic
 533 effects of serotonergic and noradrenergic genes on
 534 emotional attentional biases. However, the fact that we
 535 measured only two polymorphisms out of a number
 536 that might contribute to the behavioural effect of in-
 537 terest represents a limitation to this study. Most sig-
 538 nificantly, we were unable to genotype the additional
 539 rs25531 SNP in the long allele of the 5-HTT gene.
 540 However, given that the prevalence of the L_G allele
 541 is low (~10%), this is unlikely to have significantly
 542 biased our findings. A further limitation is that al-
 543 though the overall sample size was reasonable, there
 544 were relatively few individuals in some genotype
 545 combinations. This is a particular difficulty inherent in
 546 measuring epistatic gene effects (Moore, 2008). Future
 547 studies will need to use large sample sizes and evol-
 548 ving methodologies to effectively evaluate the likely
 549 effects on emotional processing of multiple gene in-
 550 teractions (Cordell, 2009). The final limitation is that
 551 we only used aversive stimuli and male volunteers.
 552 While the latter eliminated possible biases associated
 553 with gender differences in emotional processing, it
 554 limits the generalizability of our findings. It is there-
 555 fore unclear whether the observed biases are valence
 556 and/or gender specific. Additionally the aversive
 557 stimuli used included range of negative emotions
 558 (disgust, fear, sadness) rather than specifically dys-
 559 phoric or threat-related emotions. It is therefore un-
 560 clear how these processing biases might map onto
 561 those considered to relate to depression and anxiety

disorders. Further studies with larger sample sizes 562
 including men and women will be required to repli- 563
 cate and extend our findings. 564

Conclusions 565

In spite of these limitations, this study begins to con- 566
 tribute to the understanding of multiple gene effects 567
 and interactions in an established cognitive marker for 568
 affective spectrum disorders – the negative attentional 569
 bias. It further underlines the potential utility of 570
 adopting the ‘endophenotype’ approach in pharma- 571
 cogenetics studies, i.e. examining the genetic factors 572
 underlying not only the clinical response but also res- 573
 sponses in cognitive and neural markers. One signifi- 574
 cant challenge for such studies will be to delineate 575
 the interaction between potential neurodevelopmental 576
 effects of genetic polymorphisms influencing brain 577
 neurotransmitter systems and the acute/sub-acute 578
 effects of drug administration. Knockout and trans- 579
 genic mouse models are likely to be useful in 580
 appreciating the dynamics of the behavioural– 581
 psychopharmacogenetic–neurodevelopmental inter- 582
 face. 583

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Statement of Interest 590

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References 592

- Anderson AK (2005). Affective influences on the attentional 593
 dynamics supporting awareness. *Journal of Experimental* 594
Psychology: General **134**, 258–281. 595
- Anderson AK, Phelps EA (2001). Lesions of the human 596
 amygdala impair enhanced perception of emotionally 597
 salient events. *Nature* **411**, 305–309. 598
- Arce E, Simmons A, Lovero K, Stein M, *et al.* (2008). 599
 Escitalopram effects on insula and amygdala BOLD 600
 activation during emotional processing. 601
Psychopharmacology **196**, 661–672. 602
- Arnone D, Horder J, Cowen P, Harmer C (2009). Early effects 603
 of mirtazapine on emotional processing. 604
Psychopharmacology **203**, 685–691. 605
- Beevers CG, Ellis AJ, Wells TT, McGeary JE (2010). 606
 Serotonin transporter gene promoter region 607

- 608 polymorphism and selective processing of emotional
609 images. *Biological Psychology* **83**, 260–265.
- 610 **Beevers CG, Gibb BE, McGeary JE, Miller IW** (2007).
611 Serotonin transporter genetic variation and biased
612 attention for emotional word stimuli among psychiatric
613 inpatients. *Journal of Abnormal Psychology* **116**, 208–212.
- 614 **Beevers CG, Marti CN, Lee HJ, Stote DL, et al.** (2011).
615 Associations between serotonin transporter gene promoter
616 region (5-HTTLPR) polymorphism and gaze bias for
617 emotional information. *Journal of Abnormal Psychology* **120**,
618 187–197.
- 619 **Bertolino A, Arciero G, Rubino V, Latorre V, et al.** (2005).
620 Variation of human amygdala response during threatening
621 stimuli as a function of 5-HTTLPR genotype and
622 personality style. *Biological Psychiatry* **57**, 1517–1525.
- 623 **Bradley MM, Lawson ML** (1999). *Affective Norms for English*
624 *Words (ANEW)*. Gainesville, FL: NIMH Center for the Study
625 of Emotion and Attention.
- 626 **Browning M, Reid C, Cowen PJ, Goodwin GM, et al.** (2007).
627 A single dose of citalopram increases fear recognition in
628 healthy subjects. *Journal of Psychopharmacology* **21**, 684–690.
- 629 **Brühl AB, Kaffenberger T, Herwig U** (2009). Serotonergic
630 and noradrenergic modulation of emotion processing by
631 single dose antidepressants. *Neuropsychopharmacology* **35**,
632 521–533.
- 633 **Burghardt NS, Sullivan GM, McEwen BS, Gorman JM, et al.**
634 (2004). The selective serotonin reuptake inhibitor
635 citalopram increases fear after acute treatment but reduces
636 fear with chronic treatment: a comparison with tianeptine.
637 *Biological Psychiatry* **55**, 1171–1178.
- 638 **Canli T, Omura K, Haas B, Fallgatter A, et al.** (2005). Beyond
639 affect: a role for genetic variation of the serotonin
640 transporter in neural activation during a cognitive
641 attention task. *Proceedings of the National Academy of Sciences*
642 *USA* **102**, 12224–12229.
- 643 **Caspi A, Hariri AR, Holmes A, Uher R, et al.** (2010). Genetic
644 sensitivity to the environment: the case of the serotonin
645 transporter gene and its implications for studying complex
646 diseases and traits. *American Journal of Psychiatry* **167**,
647 509–527.
- 648 **Caspi A, Sugden K, Moffitt TE, Taylor A, et al.** (2003).
649 Influence of life stress on depression: moderation by a
650 polymorphism in the 5-HTT gene. *Science* **301**, 386–389.
- 651 **Clement HW, Gamsa D, Wesemann W** (1992). The effect of
652 adrenergic drugs on serotonin metabolism in the nucleus
653 raphe dorsalis of the rat, studied by *in vivo* voltammetry.
654 *European Journal of Pharmacology* **217**, 43–48.
- 655 **Cordell HJ** (2009). Detecting gene–gene interactions that
656 underlie human diseases. *Nature Reviews Genetics* **10**,
657 392–404.
- 658 **de Boer T** (1995). The effects of mirtazapine on central
659 noradrenergic and serotonergic neurotransmission.
660 *International Clinical Psychopharmacology* **10** (Suppl. 4),
661 19–23.
- 662 **De Boer T, Nefkens F, Van Helvoirt A** (1994). The α_2 -
663 adrenoceptor antagonist org 3770 enhances serotonin
664 transmission *in vivo*. *European Journal of Pharmacology* **253**,
665 R5–R6.
- De Martino B, Strange B, Dolan R** (2008). Noradrenergic
666 neuromodulation of human attention for emotional and
667 neutral stimuli. *Psychopharmacology* **197**, 127–136. 668
- de Quervain DJF, Kolassa IT, Ertl V, Onyut PL, et al.** (2007).
669 A deletion variant of the α_2b -adrenoceptor is related to
670 emotional memory in Europeans and Africans. *Nature*
671 *Neuroscience* **10**, 1137–1139. 672
- Firk C, Markus C** (2009). Differential effects of 5-HTTLPR
673 genotypes on mood, memory, and attention bias following
674 acute tryptophan depletion and stress exposure. 675
Psychopharmacology **203**, 805–818. 676
- Fox E, Ridgewell A, Ashwin C** (2009). Looking on the bright
677 side: biased attention and the human serotonin transporter
678 gene. *Proceedings of the Royal Society of London, Series B:*
679 *Biological Sciences* **276**, 1747–1751. 680
- Gibbs A, Naudts K, Spencer E, David A** (2007). The role of
681 dopamine in attentional and memory biases for emotional
682 information. *American Journal of Psychiatry* **164**, 1603–1609. 683
- Gibbs AA, Naudts KH, Azevedo RT, David AS** (2010).
684 Deletion variant of α_2b -adrenergic receptor gene
685 moderates the effect of COMT val158met polymorphism
686 on episodic memory performance. *European*
687 *Neuropsychopharmacology* **20**, 272–275. 688
- Hamann S, Canli T** (2004). Individual differences in emotion
689 processing. *Current Opinion in Neurobiology* **14**, 233–238. 690
- Hariri AR, Drabant EM, Munoz KE, Kolachana BS, et al.**
691 (2005). A susceptibility gene for affective disorders and the
692 response of the human amygdala. *Archives of General*
693 *Psychiatry* **62**, 146–152. 694
- Hariri AR, Mattay VS, Tessitore A, Kolachana B, et al.**
695 (2002). Serotonin transporter genetic variation and the
696 response of the human amygdala. *Science* **297**, 400–403. 697
- Harmer C, Heinzen J, O’Sullivan U, Ayres R, et al.** (2008).
698 Dissociable effects of acute antidepressant drug
699 administration on subjective and emotional processing
700 measures in healthy volunteers. *Psychopharmacology* **199**,
701 495–502. 702
- Harmer CJ, Bhagwagar Z, Perrett DI, Völlm BA, et al.** (2003).
703 Acute SSRI administration affects the processing of social
704 cues in healthy volunteers. *Neuropsychopharmacology* **28**,
705 152. 706
- Harmer CJ, Goodwin GM, Cowen PJ** (2009). Why do
707 antidepressants take so long to work? A cognitive
708 neuropsychological model of antidepressant drug action. 709
British Journal of Psychiatry **195**, 102–108. 710
- Harmer CJ, Shelley NC, Cowen PJ, Goodwin GM** (2004).
711 Increased positive *vs.* negative affective perception and
712 memory in healthy volunteers following selective
713 serotonin and norepinephrine reuptake inhibition. 714
American Journal of Psychiatry **161**, 1256–1263. 715
- Hayward G, Goodwin GM, Cowen PJ, Harmer CJ** (2005).
716 Low-dose tryptophan depletion in recovered depressed
717 patients induces changes in cognitive processing
718 without depressive symptoms. *Biological Psychiatry* **57**,
719 517–524. 720
- Heils A, Teufel A, Petri S, Stöber G, et al.** (1996). Allelic
721 variation of human serotonin transporter gene expression. 722
Journal of Neurochemistry **66**, 2621–2624. 723

- 724 **Heinz A, Braus DF, Smolka MN, Wrase J, et al.** (2004).
725 Amygdala-prefrontal coupling depends on a genetic
726 variation of the serotonin transporter. *Nature Neuroscience*
727 **8**, 20–21.
- 728 **Hu X, Oroszi G, Chun J, Smith TL, et al.** (2005). An
729 expanded evaluation of the relationship of four alleles to
730 the level of response to alcohol and the alcoholism risk.
731 *Alcoholism: Clinical and Experimental Research* **29**, 8–16.
- 732 **Hu XZ, Lipsky RH, Zhu G, Akhtar LA, et al.** (2006).
733 Serotonin transporter promoter gain-of-function
734 genotypes are linked to obsessive-compulsive disorder.
735 *American Journal of Human Genetics* **78**, 815–826.
- 736 **Hu XZ, Rush AJ, Charney D, Wilson AF, et al.** (2007).
737 Association between a functional serotonin transporter
738 promoter polymorphism and citalopram treatment in
739 adult outpatients with major depression. *Archives of*
740 *General Psychiatry* **64**, 783–792.
- 741 **Keil A, Ihssen N** (2004). Identification facilitation for
742 emotionally arousing verbs during the attentional blink.
743 *Emotion* **4**, 23–35.
- 744 **Kucera H, Francis W** (1967). *Computational Analysis of Present-*
745 *Day American English*. Providence: Brown University Press.
- 746 **Kwang T, Wells TT, McGeary JE, Swann WB, et al.** (2010).
747 Association of the serotonin transporter promoter region
748 polymorphism with biased attention for negative word
749 stimuli. *Depression and Anxiety* **27**, 746–751.
- 750 **Leppanen JM** (2006). Emotional information processing in
751 mood disorders: a review of behavioral and neuroimaging
752 findings. *Current Opinion in Psychiatry* **19**, 34–39.
- 753 **Lesch K-P, Bengel D, Heils A, Sabol S, et al.** (1996).
754 Association of anxiety-related traits with a polymorphism
755 in the serotonin transporter gene regulatory region. *Science*
756 **274**, 1527–1531.
- 757 **Lesch KP, Gutknecht L** (2005). Pharmacogenetics of the
758 serotonin transporter. *Progress in Neuro-Psychopharmacology*
759 *and Biological Psychiatry* **29**, 1062–1073.
- 760 **Markus CR, De Raedt R** (2011). Differential effects of
761 5-HTTLPR genotypes on inhibition of negative emotional
762 information following acute stress exposure and
763 tryptophan challenge. *Neuropsychopharmacology* **36**,
764 819–826.
- 765 **Markus CR, Firk C** (2009). Differential effects of tri-allelic
766 5-HTTLPR polymorphisms in healthy subjects on mood
767 and stress performance after tryptophan challenge.
768 *Neuropsychopharmacology* **34**, 2667–2674.
- 769 **McGaugh JL** (2004). The amygdala modulates the
770 consolidation of memories of emotionally arousing
771 experiences. *Annual Review of Neuroscience* **27**, 1–28.
- 772 **McMahon FJ, Buervenich S, Charney D, Lipsky R, et al.**
773 (2006). Variation in the gene encoding the serotonin 2A
774 receptor is associated with outcome of antidepressant
775 treatment. *American Journal of Human Genetics* **78**, 804–814.
- 776 **Mongeau R, Blier P, Montigny C** (1993). *In vivo*
777 electrophysiological evidence for tonic activation by
778 endogenous noradrenaline of α 2-adrenoceptors on
779 5-hydroxytryptamine terminals in the rat hippocampus.
780 *Naunyn-Schmiedeberg's Archives of Pharmacology* **347**,
781 266–272.
- Moore JH** (2008). Analysis of gene–gene interactions. *Current*
782 *Protocols in Human Genetics* (Suppl. 59), ■, 1–10. 783 AQ5
- Munafò MR, Brown SM, Hariri AR** (2008). Serotonin
784 transporter (5-HTTLPR) genotype and amygdala
785 activation: a meta-analysis. *Biological Psychiatry* **63**,
786 852–857. 787
- Munafò MR, Johnstone EC, Mackintosh B** (2005).
788 Association of serotonin transporter genotype with
789 selective processing of smoking-related stimuli in current
790 smokers and ex-smokers. *Nicotine and Tobacco Research* **7**,
791 773–778. 792
- Murphy F, Smith B, Cowen P, Robbins T, et al.** (2002). The
793 effects of tryptophan depletion on cognitive and affective
794 processing in healthy volunteers. *Psychopharmacology* **163**,
795 42–53. 796
- Murphy SE, Norbury R, O'Sullivan U, Cowen PJ, et al.**
797 (2009a). Effect of a single dose of citalopram on amygdala
798 response to emotional faces. *British Journal of Psychiatry*
799 **194**, 535–540. 800
- Murphy SE, Yiend J, Lester KJ, Cowen PJ, et al.** (2009b).
801 Short-term serotonergic but not noradrenergic
802 antidepressant administration reduces attentional
803 vigilance to threat in healthy volunteers. *International*
804 *Journal of Neuropsychopharmacology* **12**, 169–179. 805
- Murthy NV, Selvaraj S, Cowen PJ, Bhagwagar Z, et al.**
806 (2010). Serotonin transporter polymorphisms (SLC6A4
807 insertion/deletion and rs25531) do not affect the
808 availability of 5-HTT to [11C] DASB binding in the living
809 human brain. *NeuroImage* **52**, 50–54. 810
- NCI-NHGRI Working Group on Replication in Association**
811 **Studies** (2007). Replicating genotype-phenotype
812 associations. *Nature* **447**, 655–660. 813
- Nelson H** (1982). *The National Adult Reading Test (NART)*.
814 Windsor: NEFR-Nelson. 815
- Norbury R, Mackay CE, Cowen PJ, Goodwin GM, et al.**
816 (2007). Short-term antidepressant treatment and facial
817 processing: functional magnetic resonance imaging study.
818 *British Journal of Psychiatry* **190**, 531–532. 819
- Nutt DJ** (2002). The neuropharmacology of serotonin and
820 noradrenaline in depression. *International Clinical*
821 *Psychopharmacology* **17**, S1–S12. 822
- Osinsky R, Reuter M, Kupper Y, Schmitz A, et al.** (2008).
823 Variation in the serotonin transporter gene modulates
824 selective attention to threat. *Emotion* **8**, 584–588. 825
- Paddock S, Laje G, Charney D, Rush AJ, et al.** (2007).
826 Association of GRIK4 with outcome of antidepressant
827 treatment in the STAR*D cohort. *American Journal of*
828 *Psychiatry* **164**, 1181–1188. 829
- Pezawas L, Meyer-Lindenberg A, Drabant EM,**
830 **Verchinski B, et al.** (2005). 5-HTTLPR polymorphism
831 impacts human cingulate–amygdala interactions: a genetic
832 susceptibility mechanism for depression. *Nature*
833 *Neuroscience* **8**, 828–834. 834
- Praschak-Rieder N, Kennedy J, Wilson AA, Hussey D, et al.**
835 (2007). Novel 5-HTTLPR allele associates with higher
836 serotonin transporter binding in putamen: a [¹¹C]DASB
837 positron emission tomography study. *Biological Psychiatry*
838 **62**, 327–331. 839

- 840 **Rasch B, Spalek K, Buholzer S, Luechinger R, et al.** (2009).
841 A genetic variation of the noradrenergic system is related
842 to differential amygdala activation during encoding of
843 emotional memories. *Proceedings of the National Academy of*
844 *Sciences USA* **106**, 19191–19196.
- 845 **Rawlings N, Norbury R, Cowen P, Harmer C** (2010). A single
846 dose of mirtazapine modulates neural responses to
847 emotional faces in healthy people. *Psychopharmacology* **212**,
848 625–634.
- 849 **Raymond JE, Shapiro KL, Arnell KM** (1992). Temporary
850 suppression of visual processing in an RSVP task: an
851 attentional blink? *Journal of Experimental Psychology:*
852 *Human Perception and Performance* **18**, 849–860.
- 853 **Reimold M, Smolka MN, Schumann G, Zimmer A, et al.**
854 (2007). Midbrain serotonin transporter binding potential
855 measured with [¹²⁵I]DASB is affected by serotonin
856 transporter genotype. *Journal of Neural Transmission* **114**,
857 635–639.
- 858 **Roiser JP, Müller U, Clark L, Sahakian BJ** (2007). The effects
859 of acute tryptophan depletion and serotonin transporter
860 polymorphism on emotional processing in memory and
861 attention. *International Journal of Neuropsychopharmacology*
862 **10**, 449–461.
- 863 **Routledge C, Middlemiss D** (1996). The 5HT hypothesis of
864 depression revisited. *Molecular Psychiatry* **1**, 437.
- 865 **Rubinsztein J, Rogers R, Riedel WJ, Mehta MA, et al.** (2001).
866 Acute dietary tryptophan depletion impairs maintenance
867 of ‘affective set’ and delayed visual recognition in healthy
868 volunteers. *Psychopharmacology* **154**, 319–326.
- 869 **Schosser A, Kasper S** (2009). The role of pharmacogenetics in
870 the treatment of depression and anxiety disorders.
AQ6 871 *International Clinical Psychopharmacology* **24**.
- 872 **Serretti A, Kato M, De Ronchi D, Kinoshita T** (2007). Meta-
873 analysis of serotonin transporter gene promoter
874 polymorphism (5-HTTLPR) association with selective
875 serotonin reuptake inhibitor efficacy in depressed patients.
876 *Molecular Psychiatry* **12**, 247–257.
- 877 **Small KM, Brown KM, Forbes SL, Liggett SB** (2001).
878 Polymorphic deletion of three intracellular acidic residues
of the alpha 2B-adrenergic receptor decreases g protein- 879
coupled receptor kinase-mediated phosphorylation and 880
desensitization. *Journal of Biological Chemistry* **276**, 881
4917–4922. 882
- Souery D, Amsterdam J, de Montigny C, Lecrubier Y, et al.** 883
(1999). Treatment resistant depression: methodological 884
overview and operational criteria. *European* 885
Neuropsychopharmacology **9**, 83–91. 886
- Todd RM, Anderson AK** (2009). The neurogenetics of 887
remembering emotions past. *Proceedings of the National* 888
Academy of Sciences USA **106**, 18881–18882. 889
- Todd RM, Palombo DJ, Levine B, Anderson AK** (2011). 890
Genetic differences in emotionally enhanced memory. 891
Neuropsychologia **49**, 734–744. 892
- Uher R, Huezio-Diaz P, Perroud N, Smith R, et al.** 893
(2009). Genetic predictors of response to antidepressants 894
in the GENDEP project. *Pharmacogenomics Journal* **9**, 895
225–233. 896
- van Dyck CH, Malison RT, Staley JK, Jacobsen LK, et al.** 897
(2004). Central serotonin transporter availability measured 898
with [¹²³I]β-CIT SPECT in relation to serotonin transporter 899
genotype. *American Journal of Psychiatry* **161**, 525–531. 900
- Vuilleumier P** (2005). How brains beware: neural 901
mechanisms of emotional attention. *Trends in Cognitive* 902
Sciences **9**, 585–594. 903
- Wakeno MM, Kato MM, Okugawa GM, Fukuda TP, et al.** 904
(2008). The alpha 2A-adrenergic receptor gene 905
polymorphism modifies antidepressant responses to 906
milnacipran. *Journal of Clinical Psychopharmacology* **28**, 907
518–524. 908
- Wendland JR, Martin BJ, Kruse MR, Lesch KP, et al.** (2006). 909
Simultaneous genotyping of four functional loci of human 910
SLC6A4, with a reappraisal of 5-HTTLPR and rs25531. 911
Molecular Psychiatry **11**, 224–226. 912
- Willeit M, Praschak-Rieder N, Neumeister A, Pirker W,** 913
et al. (2000). [¹²³I]-[beta]-CIT SPECT imaging shows 914
reduced brain serotonin transporter availability in 915
drug-free depressed patients with seasonal affective 916
disorder. *Biological Psychiatry* **47**, 482–489. 917