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Differential modulations of response control processes by 5-HT1A gene variation

Christian Beste [a,*], Katharina Domschke [b], Michael Falkenstein [c], Carsten Konrad [b,d,e]

[a] Institute for Cognitive Neuroscience, Department of Biopsychology, Ruhr-Universität Bochum, Germany
[b] Department of Psychiatry and Psychotherapy, University of Münster, Germany
[c] IFADO, Leibniz Institute, Dortmund, Germany
[d] Interdisciplinary Center for Clinical Research (IZKF), University of Münster, Germany
[e] Department of Psychiatry and Psychotherapy, University of Marburg, Germany

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ABSTRACT

Response selection and control are supposed to reflect important basal ganglia functions. Recently, we showed that the dopaminergic system may be especially important for response selection in compatible, but not in incompatible stimulus–response (S–R) relations. Research indicates that the dopaminergic system is influenced by the serotonergic system, but little is known about the involvement of the serotonergic system in response selection. Analyzing event-related potentials (ERPs) in a sample of healthy probands ($N = 74$), we show the 5-HT1A C(-1019)G polymorphism modulating response-related processes, as reflected in the N2 component, in compatible, but not incompatible, S–R relations. This modulation was a function of the number of -1019 G alleles. Decreasing numbers of −1019 G alleles were stepwise related to increases in the N2 on compatible trials and concomitant increases in response times. The functional effect of the 5-HT1A C(-1019)G polymorphism has previously been shown to be specific for serotonergic 1 A autoreceptors of serotonergic neurons in the dorsal raphe nucleus (DRN). Due to this close relation of genotype effects to neuroanatomically dissociable structures, the results suggest that DRN serotonin 1 A autoreceptors are important for compatible S–R relations, i.e., response selection, but not for incompatible S–R relations, i.e. response conflict or inhibition. The results extend previous findings on the dopaminergic system to the serotonergic system. The examined functions are precisely regulated on a neuronal level, since neurophysiological and behavioural effects are driven in an allele–dose fashion. Because of this, the results are of importance for future clinical applications.

Introduction

The selection and control of behavioural responses is important for normal cognitive functioning (Botvinick et al., 2004). Disturbances of response monitoring may contribute to impulsive behaviour, both as a personality trait or as a symptom of psychiatric disorders (Munro et al., 2007; Potts et al., 2006; Sprink et al., 2008). Response selection and control can be differentially demanding, depending on the amount of conflict involved in response selection and hence the ease to relate the appropriate response to a stimulus (stimulus–response mapping) (Wild-Wall et al., 2008). Electrophysiological studies suggested that these processes can be examined by the N2, an event-related potential (ERP) component (e.g. Beste et al., 2008; Gajewski et al., 2008; Van Veen and Carter, 2002). Several functional interpretations of this component have been put forward. Wild-Wall et al. (2008) showed that the N2 is usually small when stimulus–responses mapping is easy, while it is enhanced when conflict between responses occurs and needs to be resolved or controlled. The N2 in non-conflict trials most likely reflects response selection, i.e. the unimpaired assignment of a specific response to a specific stimulus. In conflict trials the N2 may simply be enhanced because response selection is intensified due to conflict processing; alternatively the enhancement may be due to an additional process reflecting conflict processing, or the N2 in conflict trials may exclusively reflect conflict processing or response control (e.g. Beste et al., 2008; Gajewski et al., 2008; Folstein and Van Petten 2008). Response selection is also reflected in latency modulations of the parietal P3 (P3b), with longer P3 latencies in complex than in easy tasks and in incompatible than in compatible S–R relations (e.g. Falkenstein et al. 1994; Doucet and Stelmack, 1999; Leuthold and Sommer, 1998).

Several brain systems have been shown to be important assuring these functions, including the anterior cingulate cortex (ACC) and the mesocortico-limbic system (for rev. Chudasama and Robbins, 2006; Botvinick et al., 2004). Similarly, electrophysiological studies as well as theoretical neurocomputational simulations indicate a pivotal role of the basal ganglia in response selection and control (Gurney et al., 2004; Bar-Gad et al., 2003; Redgrave et al., 1999), which seems likely because striatal and prefrontal areas are highly interconnected. Striatal areas, as well as the anterior cingulate cortex (ACC) are massively modulated by the serotonergic system (Hensler, 2006). Projections from the dorsal raphe nucleus (DRN) heavily innervate

* Corresponding author. Fax: +49 234 321 4377.
E-mail address: christian.beste@rub.de (C. Beste).

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the prefrontal cortex and the striatum (Molliver, 1987). Especially serotonin 1 A receptors (5-HT1A) are densely localized in the DRN, and in limbic, paralimbic and prefrontal cortical regions (Varnäs et al., 2004). Within frontal cortical regions serotonin 1A receptor binding has recently been found to be strong in the ACC (Frey et al., 2008). This is of particular importance for the N2 component, which is supposed to be generated in the ACC (Van Veen and Carter, 2002). Moreover, response selection and control functions are impaired in mood and anxiety disorders (Fossati et al., 2002; Holmes and Pizzagalli, 2008; Munro et al., 2007; Potts et al., 2006; Sprink et al., 2008), for which a functional serotonin 1A receptor polymorphism (5-HT1A C(-1019)G) (Huang et al., 2004) has been implied in association studies (see Drago et al., 2008). The functional 5-HT1A C(-1019)G (Huang et al., 2004) influences serotonergic neurotransmission (Albert and Lemonde, 2004). The presence of a -1019 G allele is accompanied by a de-repression of 5-HT1A autoreceptor expression by disrupting an inhibitory transcription factor-binding site. This leads to a reduced serotonergic neurotransmission (Lemonde et al., 2003). Because of well-validated functional relevance of this single-nucleotide polymorphism (SNP), this SNP seems well suited to investigate the contribution of the serotonergic system to cognitive control processes. We examined the N2 and P3 against the functional serotonin 1A receptor polymorphism (5-HT1A C(-1019)G) in compatible and incompatible stimulus–response relations. It has recently been shown that compatible and incompatible stimulus–response relations may not be mediated by the same neurotransmitter system (Willemssen et al., 2009). A similar dissociated pattern between compatible and incompatible S–R relations would imply that the serotonin 1 A receptor system is selectively important for different kinds of response-related processes. Interestingly, the functional effect of the 5-HT1A C(-1019)G polymorphism seems to be specific for 1A autoreceptors of serotonergic neurons in the dorsal raphe nucleus (DRN) (Czesak et al., 2006; Parsey et al., 2006a; 2006b). If the effects of the 5-HT1A C(-1019)G polymorphism are restricted to compatible or incompatible response selection processes, this would suggest a specific influence of the DRN 5-HT1A autoreceptors on response selection per se (compatible S–R relation) or rather conflict processing/response control (incompatible S–R relation).

Concerning the P3, our hypothesis is based on evidence suggesting that the P3 does not depend on the serotonergic system (e.g. Wienberg et al., 2009; Oranje et al., 2008). Hence this ERP should not be modulated by the 5-HT1A C(-1019)G polymorphism. Such a dissociating pattern between two ERP-components would increase the specificity of results.

Materials and methods

Subjects

A sample of $N=74$ genetically unrelated subjects of Caucasian descent was recruited by newspaper announcements. The mean age of the subjects was 25.1 (5.6). The sample consisted of 24 males and 50 females. Calculating a Kruskal–Wallis Test (*H*-Test) it is shown that the sexes were comparably distributed across the different 5-HT1A C (-1019)G genotype groups ($chi^2=0.03$; $df=1$; $p>0.8$). As the functional 5-HT1A C(-1019)G polymorphism was found to be associated with mood and anxiety disorders (for review: Drago et al., 2008; Albert and Lemonde, 2004), we examined the Beck Depression inventory (BDI) (Beck et al., 1961) and anxiety sensitivity (ASI) (Reiss et al., 1986) to account for these factors. The mean depression score as measured by the BDI was 3.71 (SD = 2.99), indicating a non-depressed study population. There was only one subject revealing a BDI score of 13. A univariate ANOVA shows that the BDI scores did not differ between the genotype groups ($F(2,70)=1.12$; $p>0.2$). The mean ASI score was 20.3 (11.1). Genotype groups differed from each other with respect to this score ($F(2,70)=5.45$; $p=0.005$), reflecting the relevance of this polymorphism for anxiety sensitivity. Bonferroni-corrected pair-wise comparisons revealed that the CG genotype group showed a higher score (25.6 ± 9.3) than the other genotype groups (CC: 15.8 ± 9.8; GG: 18.6 ± 12.6). The CC and GG genotype groups did not differ from each other ($p>0.6$).

The Hardy–Weinberg equilibrium was examined using the program De Finetti provided as an online source (http://ihg.gsf.de/cgi-bin/hw/hwa1.pl; Wienker TF and Strom TM). The distribution of 5-HT1A C(-1019)G genotypes did not significantly differ from the expected numbers calculated on the basis of observed allele frequencies according to Hardy–Weinberg equilibrium (CC = 20, CG = 33, GG = 21; $p=0.353$). Volunteers were paid 8 Euros per hour as compensation. The study was approved by decision of the ethics committee of the University of Münster. Demographical data and basic behavioural data are given in Table 1.

Genotyping

Genomic DNA was extracted from a 10 ml EDTA venous blood sample with the Qiagen FlexiGene DNA kit (Qiagen, Hilden, Germany). The 5-HT1A C(-1019)G (rs6295) polymorphism was genotyped by means of a polymerase chain reaction (PCR)-based restriction fragment length polymorphism assay. Primers were designed to amplify a 296-bp DNA fragment containing the forward primer 122-F

5′-AGTTTTGTTCTTCATTTCGAGAT-3′ and reverse mutagenic primer 122-R; 5′-GAAGAAGACCGAGTGTGTCTA**A**C-3′.

The mutagenic primer was constructed in order to introduce an artificial polymorphic restriction site. By using a Biometra T-Gradient thermocycler (Whatman, Göttingen, Germany) standard PCR was carried out in a total volume of 20 µl containing 60 ng of genomic DNA, 1× PCR buffer, 8 pmol of each primer, 8 mM dNTPs and 0.4 U of Taq polymerase (5Prime, Hamburg, Germany). After an initial step of denaturation at 94 °C for 5 min, 35 cycles were carried out consisting of 94 °C for 30 s, 54 °C (annealing temperature) for 30 s, 72 °C for 60 s and a final extension step of 10 min at 72 °C. Subsequent digestion overnight for 16 h at 65 °C of an 8 µl sample of the PCR product was accomplished with 3 U of TaiI (Fermentas, St. Leon-Rot, Germany) in a total volume of 20 µl resulting in two patterns of fragments consisting of 203 + 57 + 36 bp for the G-allele and 183 + 57 + 36 + 20 bp for the C-allele. Digestion products were visualized by silver staining after separation on a 15% polyacrylamide gel in 1× TBE buffer (Tris-Borate, EDTA) at 220 V for 3 h. Genotypes were determined independently by two investigators.

Experimental paradigm

To assess response selection and control for conflict and non-conflict conditions, we used a modified flanker task (Kopp et al., 1996).

Table 1
Demographical and basic behavioural data across 5-HT1A C(-1019)G genotype groups. The mean and standard deviation (SD) are given.

	CC	CG	GG
N	20	33	21
Age	24.5 (6)	24.9 (5.1)	25.9 (5.7)
Sex	6 males: 14 females	10 males: 23 females	8 males: 13 females
BDI	2.6 (2.5)	3.6 (2.6)	4.14 (3.5)
ASI	15.8 (9.8)	25.6 (9.3)	18.6 (12.6)
Reaction time (RT)			
Compatible	412 (29)	391 (91)	374 (87)
Incompatible	415 (31)	417 (92)	415 (90)
Error rate			
Compatible	2.35 (1.46)	2.2 (2.1)	2.1 (1.1)
Incompatible	9.1 (2.8)	10.2 (3.8)	8.2 (2.4)

The task consisted of vertical arrays of arrowheads or circles. The central part of the stimulus was defined as target. When the target was an arrowhead, the subjects had to press a button on the side the target pointed to; when the target was a circle, no response had to be given (Nogo trials). Above and below each target, a flanker was presented which pointed either to the same side (congruent trials) or to the opposite side (incompatible trials) of the target. Nogo and incongruent trials had a probability of 20% each, congruent trials had a probability of 60%. By making the incongruent stimuli relatively rare, we aimed at increasing interference and hence on response control processes. Right and left pointing flankers were equiprobable. The flankers preceded the targets by 100 ms (Stimulus Onset Asynchrony, SOA = 100 ms) to further strengthen their influence, and consequently further increase the demands on response control. Flankers and targets were switched off 100 ms after target onset. The next flanker was presented 800 to 1200 ms (interval randomised) after the response of the subjects, or 1900 to 2300 ms after a Nogo target. Altogether 420 stimuli were presented in four blocks of 105 stimuli each, which were interrupted by short breaks. The subjects were asked to react as fast as possible to the arrowhead targets. For the analysis, all Nogo trials were excluded.

A response was given with a response panel. Buttons were mounted at the top and had to be operated with the right and left thumb. Time pressure was administered by an individual deadline method; the deadline reaction time (RT) was determined for each subject by the mean individual RT and error rate in the flanker task in the training session. A feedback tone (1000 Hz) was presented 500 ms after the response, if the RT was slower than the deadline RT. The subjects were asked to respond fast enough to avoid the feedback tone.

Data processing

During task performance the electroencephalogram (EEG) was recorded from 26 electrodes: Fp1, Fpz, Fp2; F7, F3, Fz, F4, F8; FC5, FC3, FCz, FC4, FC6; C3, Cz, C4; P7, P3, Pz, P4, P8; M1, M2; O1, Oz, O2. The vertical EOG was recorded from 4 electrodes above and below both eyes, and the horizontal EOG from 2 electrodes at the outer canthi of the eyes. The forehead was used as ground. The primary reference was Cz. EEG and EOG data were sampled with 500 Hz (Acquire, Neuroscan Inc.) and stored continuously on a PC hard-disk, together with stimulus and response markers. The data were analysed off-line. The data was filtered using a band-pass filter from 0.5 to 16 Hz. EEG segments beginning 200 ms before and ending 1000 ms after the stimulus were cut out. The baseline was set at 200 ms till stimulus presentation. These segments were checked offline for artefacts (zero-lines, fast shifts, or drifts). For the final quantification of the data, only participants were included for whom less than 15 trials were excluded in each condition during the artefact rejection procedure. The number of excluded trials in each condition was not different across genotype groups. Trials with horizontal eye movements (saccades) preceding the latency of the components of interest were excluded by manual inspection. The influence of remaining eye movements upon electrocortical activity was corrected by the algorithm proposed by Gratton, Coles and Donchin (1983). The ERP data were re-referenced to average reference to make them independent on any specific reference such as the mastoid. To avoid baseline effects, the N2 was measured against the amplitude of the preceding P2, which was determined as the largest positive peak from 190 ms after target onset until the N2 peak. The N2 was quantified at electrode Fz and FCz, as these electrodes revealed the maximum of the N2, as can be seen in the scalp topography plots (Fig. 1). The P3 was quantified at electrodes Cz and Pz and defined as the most positive peak within the time window of 300 to 600 ms. These electrodes were chosen, because potentials were strongest at these sites. Only trials with correct reactions were used for data analyses. The whole quantification procedure is comparable to Willemssen et al. (2009).

Statistical analysis

Behavioural parameters (reaction times RT, error rates) were analyzed in separate repeated measures ANOVAs with the within-subject factor "compatibility" (compatible vs. incompatible) and the between subject factor "5-HT1A C(-1019)G genotype group". Amplitude and latency parameter of the N2 were analyzed in separate repeated measures ANOVAs with the within-subject factors "electrode" (Fz, FCz), "compatibility" (compatible vs. incompatible) and the between subject factor "5-HT1A C(-1019)G genotype group". The P3 was quantified at electrodes Cz and Pz und subjected to a similar ANOVA. All performed *post-hoc* tests were Bonferroni-corrected and Greenhouse–Geisser correction was applied, where appropriate. All variables included into the analyses were normally distributed (all $z<0.9$; $p>0.2$; one-tailed). The mean and standard error of the mean are given (M ± SEM). As the 5-HT1A C(-1019)G genotype groups differed in their ASI scores, all the analyses described above were repeated using the ASI scores as covariate in an analysis of covariance (ANCOVA) to examine, whether the effects are robust or further modulated by this factor.

Results

Behavioural data

For the reaction times (RTs) the ANOVA revealed a main effect "compatibility" ($F(1,71) = 197.42$; $p<0.001$), denoting that RTs were

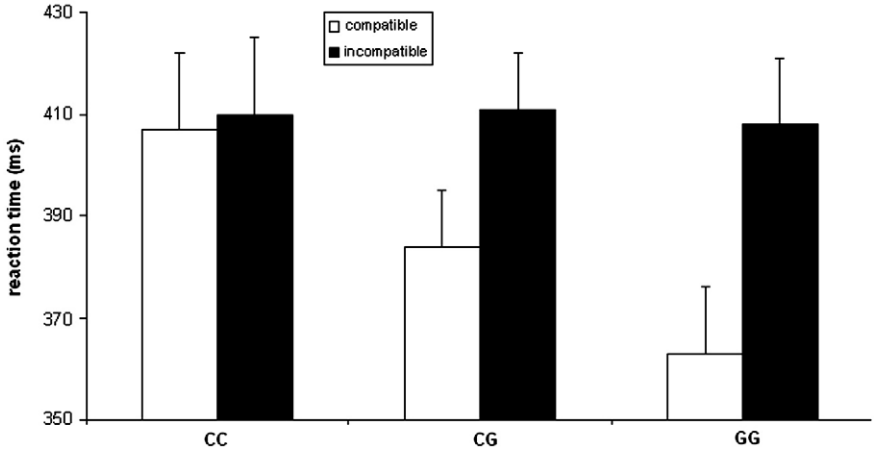

Fig. 1. Reaction times (RTs) for compatible and incompatible trials, separated for the different 5-HT1A C(-1019)G genotype groups. Differences in RTs between compatible and incompatible trials were evident in the CG and GG genotype groups.

faster on compatible (373 ± 8) compared to incompatible trials (401 ± 9). While there was no main effect of "group" ($F(2,71) = 0.52$; $p > 0.5$), there was a strong interaction "compatibility × group" ($F(2,71) = 40.30$; $p < 0.001$). Subsequent univariate ANOVAs performed as post-hoc tests revealed that, on incompatible trials, there was no difference in RTs between groups ($F(2,71) = 0.21$; $p > 0.9$), while there was a difference on compatible trials $F(2,71) = 6.85$; $p < 0.001$): in this condition RT was shortest for the GG group (363 ± 4), longer for the CG group (384 ± 5), and longest for the CC group (407 ± 6). All groups differed from each other ($p < 0.001$). Error rates were lower in compatible (2.12 ± 0.2), compared to incompatible S–R relations (9.21 ± 0.3) ($F(1,71) = 252.65$; $p < 0.001$; $\eta = 0.718$). There was no main or interaction effect with group (all Fs < 1.1; $p > 0.2$). These results are illustrated in Fig. 1. The ANCOVA used to control for possible effects of the ASI-score revealed that the results are unbiased with respect to this factor (all Fs < 0.3; $p > 0.6$). Similarly, "sex" did not affect the pattern of results (all Fs < 0.4; $p > 0.6$).

Neurophysiological data

N2

Stimulus-locked averages across the groups are given in Fig. 2.

For the N2, the repeated measures ANOVA revealed that the N2 was larger at electrode Fz (−5.8 ± 0.1), compared to FCz (−4.02 ± 0.2) ($F(1,71) = 447.5$; $p < 0.001$; $\eta = 0.85$) and was also generally larger for incompatible (−6.33 ± 0.3), compared to compatible trials (−3.71 ± 0.2) ($F(1,71) = 300.3$; $p < 0.001$; $\eta = 0.58$). Across trial types (compatible and incompatible) the N2 was largest for the CC genotype group (−5.54 ± 0.1) and decreased over the CG (−5.04 ± 0.1) to the GG genotype group (−4.48 ± 0.1) (main effect genotype group: $F(2,91) = 25.97$; $p < 0.001$). Moreover, there was an interaction "electrode × compatibility × group" ($F(2,71) = 5.33$; $p = 0.006$). This interaction is given in Fig. 3.

As can be seen in Fig. 3, the difference in N2 amplitude between compatible and incompatible trials was lacking (at electrode Fz)

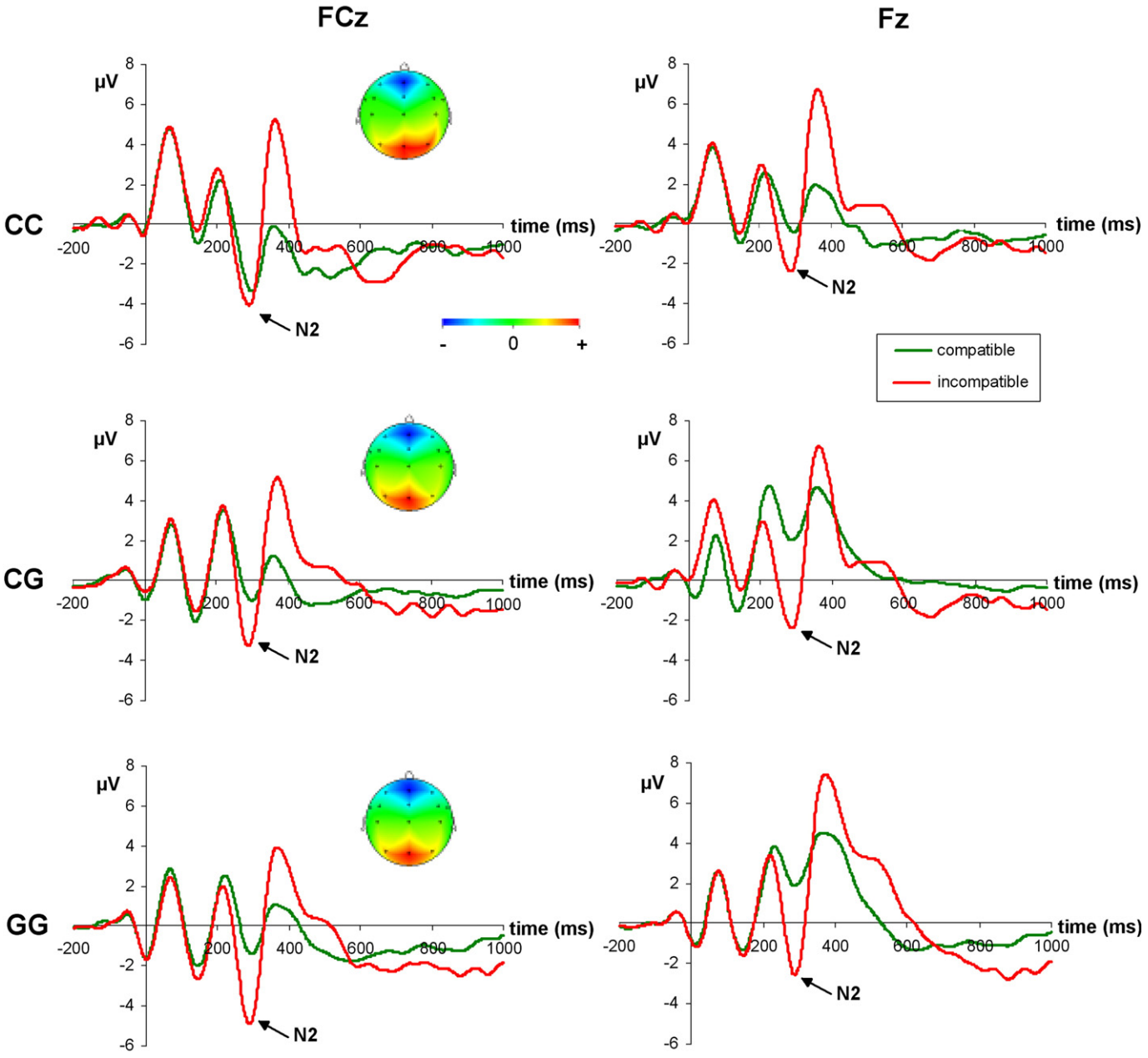

Fig. 2. Stimulus-locked ERPs on compatible and incompatible trials at electrodes FCz (left) and Fz (right), separated for the different 5-HT1A C(-1019)G genotype groups. Time point 0 denotes the point of target presentation. The N2 is clearly seen at all electrodes for each genotype group. Furthermore, a clear N2-topography can be seen in all genotype groups.

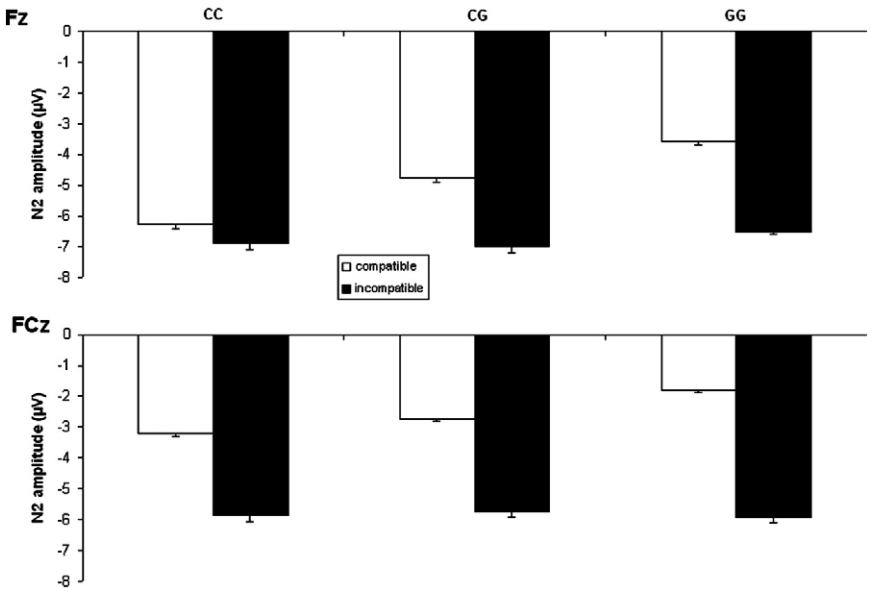

Fig. 3. Amplitudes of the N2 on compatible and incompatible trials, separated for the different 5-HT1A C(-1019)G genotype groups. The top row denotes electrode Fz, the bottom row denotes electrode FCz. As can be seen, there is a lack of difference between compatible and incompatible trials at electrode Fz for the CC genotype group, only. This lack of difference is due to an increase in amplitude for compatible trials.

selectively for the CC genotype group. For all other groups, there was a difference between the trial types. This is underlined by the statistical analysis, where the electrodes were analyzed separately. These analyses revealed that for electrode Fz the effect-size of the interaction effect "compatibility × group" was larger ($F(2,71) = 23.22$; $p<0.001$; $\eta = 0.58$) than for electrode FCz ($F(2,71) = 9.66$; $p<0.001$; $\eta = 0.17$). Hence, only electrode Fz was further analyzed. While there was a difference between compatible and incompatible trials for the CG ($F(1,30) = 162.16$; $p<0.001$; $\eta = 0.80$) and GG genotype groups ($F(1,18) = 163.69$; $p<0.001$; $\eta = 0.84$), this effect was absent for the CC genotype group ($F(1,17) = 0.59$; $p>0.4$ $\eta = 0.02$) (cf. Fig. 3a). Calculating a univariate ANOVA across the difference in N2 amplitudes between compatible and incompatible trials it is shown that these differences increase from the CC over the CG to the GG genotype group ($F(2,71) = 31.51$; $p<0.001$) (see Fig. 3), with each group differing from each other. It is also shown that the groups did not differ in N2 amplitude on incompatible trials ($F(2,71) = 0.15$; $p>0.8$), but on compatible trials ($F(2,71) = 108.6$; $p<0.001$; $\eta = 0.70$). The GG group showed the smallest N2 (-2.70 ± 0.3); followed by the CG (-3.72 ± 0.3) and CC genotype group (-4.91 ± 0.2) ($p<0.001$).

Analyzing the latencies of the N2 revealed no significant effects (all F's<1.2; $p>0.2$). Similar to the behavioural data, the ASI score did not further modulate the effects obtained for amplitudes and latencies, as revealed by an ANCOVA (all F's<0.5; $p>0.5$). The same holds true for the factor "sex" (all F's<0.5; $p>0.4$). The specificity of results is underlined analyzing the P2-amplitudes against baseline. The analysis shows that neither the amplitudes, nor the latencies differed between genotype groups (all F's<0.07; $p>0.3$).[1]

P3

For the amplitudes the ANOVA revealed a main effect "compatibility" ($F(1,71) = 54.62$; $p<0.001$; $\eta = 0.266$) showing the P3 being larger for compatible (8.03 ± 0.2) than for incompatible trials (6.52 ± 0.2). Potentials were also larger at electrode Pz (8.27 ± 0.2), compared to Cz (6.27 ± 0.3) ($F(1,71) = 33.02$; $p<0.001$). However, there was no main effect "group" ($F(2,71) = 0.17$; $p>0.8$). All interaction effects were not significant (all. F's<1.7; $p>0.2$). Concerning the latencies, the ANOVA revealed that these were shorter at electrode Pz (377 ± 7), compared to Cz (430 ± 8) ($F(1,71) = 57.93$; $p<0.001$; $\eta = 0.38$). They were also prolonged for incompatible (437 ± 8), compared to compatible trials (371 ± 7) ($F(1,71) = 86.18$; $p<0.001$; $\eta = 0.48$). There was an interaction "electrode × compatibility" ($F(1,71) = 19.64$; $p<0.001$) showing that compatibility effects were stronger at electrode Pz, than at Cz (latency difference Cz: 34 ± 9; latency difference Pz: 99 ± 9). All other main or interaction effects were not significant (all F's<0.96; $p>0.3$). Also, the ASI-score (all F's<0.6; $p>0.5$) and sex (all F's<0.4; $p>0.6$) did not further modulate the observed effects.

Discussion

In the current study, we examined the effects of the functional serotonin 1A receptor polymorphism C(-1019)G on response selection and control. The behavioural data indicates that a difference between compatible and incompatible S–R relations was almost absent in the CC genotype group. The CG genotype group showed a stronger difference than the CC group. The difference between compatible and incompatible trials was most prominent in the GG genotype group. Hence, the gradual difference in performance between compatible and incompatible trials appears to be a function of the gradual increase in the number of 5-HT1A -1019 G alleles. This modulation is restricted to compatible trials only, since only RTs of compatible trials differed between the genotype groups, but not RTs on incompatible trials. This behavioural pattern is paralleled by the neurophysiological data (i.e. N2-data): the longer the RTs the larger the N2 in compatible trials. The data also show a genotype-specific modulation, again selectively for compatible trials. The specificity of these results is underlined by the P3-data, which showed no modulatory effects of the functional 5-HT1A C(-1019)G polymorphism. Yet, we observed well-known P3 effects of compatibility (Leuthold and Sommer, 1998), which shows the validity of our data. The observation that the P3 was not modulated by the serotonin 1A receptor system is in line with other studies reporting no influence of the serotonergic system on the P3 (e.g. Wienberg et al., 2009; Oranje et al., 2008). The results are further unbiased with respect to mood and anxiety scores of the subjects. The current results extend the

[1] The N2 on Nogo trials (Nogo-N2) did not show any modulations by genotype (all F's < 0.2; $p > 0.7$). This underlines the specificity of results obtained.

differential effect of the dopaminergic system shown by Willemssen et al. (2009) to the serotonin 1A receptor system.

Because of the relatively moderate sample size, the results should be treated cautiously. However, despite of the moderate sample size, the results are in line with our hypothesis. Phenotypes not overtly observable, the so-called endophenotypes or intermediate phenotypes (Flint and Munafo, 2007, Gallinat et al. 2008), here the functional measures applied (EEG/ERPs), may require considerably fewer subjects to identify significant gene effects on the response characteristics of the brain (Hariri and Weinberger, 2003). The moderate sample size may be a reason for the incongruent results obtained for the ASI score, which was highest in the CG and not in the GG genotype group. According to the concept of endophenotypes, we would expect that functional polymorphisms in genes are only weakly related to behaviour, but more strongly related to electrophysiologcial phenotypes (Hariri and Weinberger, 2003, Flint and Munafo, 2007, Gallinat et al. 2008).

Our data suggest that even very slight changes of serotonergic activity, e.g. as conferred by 5-HT1A C(-1019)G genotype, are able to induce a gradual modulation of the N2 and behavioural outcome in the compatible condition. Functionally altered serotonergic neural transmission based upon the 5-HT1A C(-1019)G genotype may lead to more demanding response-selection processes during compatible S–R relations, which is underlined by the behavioural data. Response selection processes seem to become fairly demanding in the compatible condition in the CC genotype, and gradually less in CG and GG genotype groups, as can be seen in an increased N2 and prolonged RTs in compatible trials in the CC genotype groups, relative to the other genotype groups. Hence, the CC genotype group seems to encounter problems in response selection even in easy, compatible S–R relations.

The results allow two important implications concerning the functional meaning of the N2 in different conditions. Since the N2 on congruent trials was modulated by a serotonergic genotype, while the N2 on incongruent trials was not, it is highly unlikely that the N2 relies on the same neuronal sources and possibly also neuroanatomical structures in both conditions. That means that the idea that the N2 is simply enhanced and response selection is simply intensified on incongruent vs. congruent trials is not supported by the present data. Likewise, the idea that an additional process is superimposed on the ongoing response selection process in incompatible trials is not supported by the data. It rather appears that the normal response selection, as present during compatible trials, is replaced by a different process related to conflict processing or response control in incompatible trials. However, it cannot completely be ruled out that the N2 in congruent and incongruent trials is generated by a similar source, which is simply differentially modulated by the serotonin 1A receptor system, depending on task demand. In this case, effects of serotonin 1A receptors on this cognitive function are demand-specific rather than specific for a neural network.

Finally, the data shed some light on the relation between the frontal N2, when recorded in correct trials as reported in the present paper, and the fronto-central Ne/ERN, when recorded in error trials (Falkenstein et al. 1991; Gehring et al. 1993). In an influential paper, Yeung et al. (2004) argued that both Ne/ERN and N2 reflect the same process, namely conflict detection or processing. If so, the influence of genetic polymorphisms, such as 5HT1A C(-1019)G, should have the same impact on the N2 and the Ne/ERN. Recently, we have reported modulations of the Ne/ERN by the 5HT1A C(-1019)G variant when applying the same task in a partially overlapping sample (Beste et al., 2009). In that study (Beste et al., 2009) the Ne/ERN in the incongruent trials was clearly modulated by the 5-HT1A C(-1019)G variant, while the N2 in the current study was not. This and the observations of the present study argue against the notion that N2 and Ne/ERN reflect similar processes. Thus the study of modulatory effects of the 5-HT1A C(-1019)G polymorphism in an partially overlapping sample provides useful insights into the functional relevance of the N2 and Ne/ERN. Moreover, Burle et al. (2008) have conclusively shown that the Ne/ERN does not reflect conflict. Similar conclusion can be drawn from other studies (e.g. Carbonnell and Falkenstein, 2006). However, in both cases (N2 and Ne) there is at least a partial involvement of the anterior cingulate cortex (ACC), which may be the reason why the serotonin 1A system modulates both processes, even though N2 and Ne reflect different processes.

The 5-HT1A receptor polymorphism influences serotonergic neurotransmission (Albert and Lemonde, 2004). More precisely, the presence of a -1019 G allele is accompanied by a de-repression of 5-HT1A autoreceptor expression by disrupting an inhibitory transcription factor-binding site. This leads to a reduced serotonergic neurotransmission (Lemonde et al., 2003). The results suggest that increases in serotonergic neurotransmission lead to an equalization of neuronal processes underlying response selection in congruent and incongruent S–R relations that is driven by effects in congruent S–R relations. The outcome of this process is that response selection becomes equally demanding in congruent and incongruent conditions. As the modulation of response selection processes across the genotype groups was due to alterations in compatible S–R relations and the functional effect of the 5-HT1A C(-1019)G polymorphism is likely specific to serotonergic 1 A autoreceptors of serotonergic neurons in the DRN (Czesak et al., 2006; Parsey et al., 2006a; 2006b), the results suggest dissociable roles of the DRN serotonin 1 A system for response selection and control.

From the point of view of receptor distribution, evidence for a striatal presence of serotonin receptors is weaker than for a prefrontal distribution. While some authors have shown that the serotonin 1A system in the DRN influences serotonergic tone throughout the brain including the striatum (Kreiss and Lucki, 1994), other suggested that serotonin 1 A receptors are expressed in prefrontal cortical, but not in striatal structures (Pompeiano et al., 1992; for review: Alex and Pehek, 2008). From the functional point of view, as noted in the introduction, the striatum seems especially important for incompatible S–R relations compared to compatible trials (Willemssen et al., 2009; Gurney et al., 2004). Considering the unclear serotonergic receptor distribution in the striatum and the importance of this region for complex response selection processes, we suggest that serotonergic influences are less relevant for incompatible S–R relations. In contrast, considering previous evidence for an important role of the dopaminergic system in compatible reactions (Willemssen et al., 2009) in conjunction with evidence on the close interaction of dopaminergic and serotonin 1 A receptors (for review: De Almeida et al., 2008), it seems reasonable that especially compatible S–R relations are modulated by serotonergic neural transmission.

However, DRN projections strongly modulate limbic, paralimbic and prefrontal cortical regions and especially the ACC (Alex and Pehek, 2008; Frey et al., 2008; Varnäs et al., 2004). It may be especially this modulation of these prefrontal regions, known to generate the N2 (Folstein and Van Petten 2008), which may underlie genotype-dependent modulations in compatible response selection processes. The 5-HT1A receptor polymorphism has been shown to influence serotonergic neurotransmission (Albert and Lemonde, 2004), and 5-HT1A receptor agonists increase dopamine-release in the prefrontal cortex (PFC) (for review: De Almeida et al., 2008).

It is well known that the relation between dopamine level and cognitive performance follows an inverted U-shape (e.g. Goldman-Rakic et al., 2000; Seamans and Yang, 2004). Stimulating the DA-system in conditions with decreased DA-levels enhances performance. However, further stimulation above the optimal level again leads to a decline in performance, comparable to the effects occurring in decreased dopamine functioning (e.g. Goldman-Rakic et al., 2000; Seamans & Yang, 2004). The presence of G alleles leads to an increase in 5-HT1A autoreceptor levels by de-repression of receptor expression. Given the regulatory feedback function of 5-HT1A autoreceptors an increased expression conferred by the G allele results in a decrease in serotonergic neural transmission (e.g. Lemonde et al., 2003).

Because of this decrease in serotonergic neural transmission, the stimulating effects on the dopaminergic system (Dias-Mataix et al., 2005; De Almeida et al., 2008) are decreased in the CG and GG genotype groups relative to the CC genotype group. In turn, the relatively stronger stimulation of the dopaminergic system in the CC genotype group may lead to the observed results, because dopaminergic neural transmission is shifted above its optimal level (Goldman-Rakic et al., 2000). However, the GG genotype does not lead to a decline in performance, even though it may shift dopaminergic neural transmission below its optimal level. The reason for this remains elusive, but it may be speculated that the CC and GG genotypes affect dopaminergic neural transmission to a different degree, which leads to the asymmetrical pattern of results.

The finding that behavioural and neurophysiological parameters of response selection and control are stepwise modulated as a function of the number of -1019 G alleles in compatible trials is also of high clinical importance. In mood and anxiety disorders, the -1019 G allele is considered to be a risk allele (for review: Albert and Lemonde, 2004; Domschke et al., 2006). Executive functions are also impaired in these disorders (Fossati et al., 2002; Holmes and Pizzagalli, 2008). There is evidence that response selection is disturbed in impulsive behaviour and in a number of psychiatric disorders (Munro et al., 2007; Potts et al., 2006; Sprink et al., 2008). Understanding neuropharmacological processes underlying response monitoring represents a first step towards a rationally based treatment. Yet, for this it is necessary to more fully understand possible interrelations of the dopaminergic and serotonergic system.

In summary, the results show that the functional 5-HT1A C(-1019)G polymorphism differentially modulates response selection and control, since this modulation is restricted to compatible trials only. The modulation is a function of the number of -1019 G alleles and affects behavioural and neurophysiological processes. As the functional effect of the 5-HT1A C(-1019)G polymorphism is likely specific to serotonergic 1 A autoreceptors of serotonergic neurons in the DRN (Czesak et al., 2006; Parsey et al., 2006) the results suggest dissociable roles of the DRN serotonin 1 A autoreceptors for response selection and control in compatible and incompatible S–R relations. The results also imply that response-selection and control functions are closely regulated on a neuronal level and even slight changes can cause strong effects. Future studies may incorporate even larger samples enabling the examination of gene–gene interactions. In this respect, especially GABAergic polymorphisms may be of interest, as they may be important for striatal processes underlying response selection. The examination of other potentially relevant neurotransmitter systems may be of importance for clinical studies. To further disentangle and validate possible neocortical/striatal dissociations in response selection processes, functional imaging (fMRI) may be conducted in parallel.

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