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Variations in the *TNF-α* Gene (TNF-α -308G→A) Affect Attention and Action Selection Mechanisms in a Dissociated Fashion

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Beste C, Baune BT, Falkenstein M, Konrad C. Variations in the *TNF-α* gene (TNF-α -308G→A) affect attention and action selection mechanisms in a dissociated fashion. *J Neurophysiol* 104: 2523–2531, 2010. First published September 1, 2010; doi:10.1152/jn.00561.2010. There is growing interest to understand the molecular basis of complex cognitive processes. While neurotransmitter systems have frequently been examined, other, for example neuroimmunological factors have attracted much less interest. Recent evidence suggests that the A allele of the tumor necrosis factor alpha (TNF-α) 308G→A single nucleotide polymorphism (SNP; rs1800629) enhances cognitive functions. However, it is also known that TNF-α exerts divergent, region-specific effects on neuronal functioning. Thus the finding that the A allele is associated with enhanced cognitive performance may be due to regionally specific effects of TNF-α. In this study, associations between the *TNF-α* −308G→A single nucleotide polymorphism (rs1800629) and cognitive function in an event-related potential (ERP) study in healthy participants (n = 96) are investigated. We focus on subprocesses of stimulus-response compatibility that are known to be mediated by different brain systems. The results show a dissociative effect of the TNF- 308G→A SNP on ERPs reflecting attentional (N1) versus conflict and action selection processes [N2 and early-lateralized readiness potential (e-LRP)] between the AA/AG and the GG genotypes. Compared with the GG genotype group, attentional processes (N1) were enhanced in the combined AA/AG genotype group, while conflict processing functions (N2) and the selection of actions (LRP) were reduced. The results refine the picture of the effects of the TNF-α −308G→A SNP on cognitive functions and emphasize the known divergent effects of TNF-α on brain functions.

INTRODUCTION

There is growing interest in examining the relation between genetic factors and cognitive processes. The investigation of genes relevant to brain function may provide a better understanding of neurobiochemical and molecular mechanisms underlying cognitive processes. In this respect, mostly neurotransmitter systems have been examined (e.g., Goldberg and Weinberger 2004; Scerif and Karmiloff-Smith 2005). Until now only very few studies investigated other molecules, such as cytokines that also modulate cognitive functions (for review: McAfoose and Baune 2009). One interesting cytokine is the tumor necrosis factor alpha (TNF-α). With respect to cognitive processes, the role of TNF-α has mainly been examined in processes related to learning and memory. It has been shown that the absence of TNF-α leads to a decline in performance. Interestingly, the overexpression of TNF-α is related to decline of cognitive performance (for review: McAfoose and Baune 2009). These effects emerge mainly because TNF-α affects long-term potentiation processes.

With respect to TNF-α, the −308G→A single nucleotide polymorphism (SNP) (rs1800629) may be of functional importance because the −308A allele has been found to confer stronger transcriptional activity than the −308G allele, likely resulting in higher TNF-α levels (Hajeer and Hutchison 2001; Wilson et al. 1997). Examining associations of this SNP with cognitive performance, a recent study by Baune et al. (2008a) showed that the A allele speeds up reactions times (RTs) compared with G allele carriers, but one cannot be sure that these effects are driven by improved sensory processing. Numerous lines of evidence suggest that the effect of TNF-α in the brain is not uniform. Specifically, it is suggested that TNF-α exerts both neuroprotective and neurodegenerative effects (McAfoose and Baune 2009; Sriram and O'Callaghan 2007). While TNF-α likely induces dysfunctions in basal ganglia structures (McCoy and Tansey 2008; Sriram et al. 2006), it is hypothesized that TNF-α unfolds neuroprotective effects in occipital structures (Kaneko et al. 2008), likely because TNF-α modulates BDNF-TrkB signaling (Kaneko et al. 2008). Generally, the determining factors why TNF-α has these opposing effects in different brain structures are not fully understood. It is hypothesized that the extent of microglial activation in specific brain regions, the timing and threshold of TNF-α expression, and the conditions that stimulate regulation of TNF signaling eventually determine whether TNF-α plays a neurotoxic or neurotrophic role in the CNS (Sriram and O'Callaghan 2007). Other research suggests that different subtypes of the TNF-α receptors TNF-R1 and -R2 mediate apoptotic (Tartaglia et al. 1993) and neuroprotective effects, respectively (Bernardino et al. 2005), which also have an effect on cognitive functions (Baune et al. 2008b).

However, the finding that the A allele is associated with enhanced cognitive performance (Baune et al. 2008a) may be due to regionally specific effects of TNF-α. Cognitive processes mediated by other brain systems may in contrast not be enhanced in A allele carriers, especially if these brain systems are adversely affected by TNF-α.

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In this study, we examine the hypothesis of a region-specific or cognitive subprocess specific effect of TNF-alpha. This approach is suitable to investigate cognitive performance in a task requiring distinct subprocesses that are known to be mediated by different brain systems. In this regard, stimulus-response compatibility (SRC) effects may be useful (Lien and Proctor 2002) because subprocesses mediating SRC effects can be examined using well-defined event-related potentials (ERPs) that are known to be generated by different brain systems:

First, stimulus identification processes take place that can be measured by the N1 (Luck 1995). The N1 ERP-component is generated in the lateral extrastriate cortex with a contribution of dorsal occipito-parietal and ventral occipital temporal structures (Gomez Gonzalez et al. 1994; Herrmann and Knight 2001). In these early processing stages, attention has been assumed to act as gain control, modifying the magnitude of neural responses to incoming information (Hillyard et al. 1999; Mangun 1995; Wascher and Beste 2010).

Action selection processes, which become increasingly important once stimulus identification processes are finished, are most likely reflected by another ERP, the N2 ERP-component (e.g., Beste et al. 2008a, 2009a, 2010b; Gajewski et al. 2008; Van Veen and Carter 2002; Willemssen et al. 2009). Wild-Wall et al. (2008) showed that the N2 is usually small when stimulus-response mapping is easy, while it is enhanced when response selection is intensified due to a conflict occurring between intended and not intended responses (Folstein and Van Petten 2008; Van Veen and Carter 2002). The N2 is known to be generated by the anterior cingulate cortex (ACC), which is part of basal ganglia-prefrontal loops that are a critical neural substrate for action selection processes (Humphries et al. 2006). Corroborating this, the N2 has been shown to be modulated in basal ganglia diseases (Beste et al. 2008a; Willemssen et al. 2009). Besides the N2, the latency of parietal P3 (P3b) is affected by action selection processes with longer P3 latencies in complex as compared with easy tasks and in incompatible as compared with compatible S-R relations (e.g., Doucet and Stelmack 1999; Falkenstein et al. 1994; Leuthold and Sommer 1998).

For response initiation processes that become increasing important once an action has been selected, the lateralized readiness potential (LRP) can be used. The LRP was developed as an additional temporal marker of the beginning of hand-specific motor activation (Gratton et al. 1988). The LRP onset divides the RT interval into premotor and motor processing times (e.g., Osman and Moore 1993). Premotor and motor processing time can be separately determined at two sets of LRP waveforms. To examine the premotor processing time, the single-trial waveforms are best averaged time locked to the stimulus (Stahl et al. 2010). The stimulus-locked (s-LRP) waveforms in incongruent, conflicting trials in a Flanker task usually show an early activation of the incorrect response hand ("dip") (Beste et al. 2008a). This dip precedes the negative deflection that reflects the correct response activation may be strongly driven by stimulus processing (Falkenstein et al. 2006).

TNF-α exerts neuroprotective effects in occipital brain structures. Due to these neuroprotective effects, we hypothesize that attentional processes are improved in A allele carriers leading to an increased N1 because the N1 is likely generated in these brain areas (Gomez Gonzalez et al. 1994; Herrmann and Knight 2001). We further hypothesize that the dip of the e-LRP is also increased as a consequence of the stronger N1 (e.g., Falkenstein et al. 2006). On a behavioral level, we hypothesize that especially the e-LRP effect is reflected in increased compatibility effects in RTs in the A allele group, which is paralleled by a delay in the P3b. Furthermore we hypothesize that the LRP is delayed in its onset and peak latency because the basal ganglia-prefrontal loops are likely to be adversely affected by TNF-α (Sriram and O'Callaghan 2007). As to the N2, no clear hypothesis can be drawn: given that the N2 reflects response selection or conflict, the N2 may be elevated in A allele carriers due to the increased N1. In this case, the modulation of the N2 will be similarly influenced like the N1. Yet as the N2 is likely modulated by basal ganglia-prefrontal interactions that are adversely affected by TNF-α, the N2 and hence response selection or conflict monitoring might be attenuated in A allele carriers despite an increase of the N1. In this case, a higher saliency of conflicting flanker information would have no effect because processing in networks subsequent to stimulus processing is compromised by TNF-α. In this way, stimulus processing and action selection processes may dissociate from each other.

METHODS

Subjects

A sample of 96 genetically unrelated healthy participants of Caucasian descent was recruited by newspaper announcements. The mean age of the subjects was 23.7 $\pm$ 4.9 (SD) yr. The sample consisted of 39 males and 57 females. As the AA genotype had an expectedly low frequency (see following text), we combined the AA and AG genotype groups to one group. The frequency of sexes did not differ across the genotype groups (Mann-Whitney U test: $Z = -0.14$; $P > 0.4$). Hardy-Weinberg equilibrium was examined using the program Finetti provided as an on-line source (http://ihg.gsf.de/cgi-bin/hw/hwa1.pl; Wienker TF and Strom TM). The distribution of TNF-α -308G→A genotypes did not significantly differ from the expected numbers calculated on the basis of observed allele frequencies according to Hardy–Weinberg equilibrium (AA = 2, AG = 39, GG = 55; P = 0.143). All subjects enrolled into the study had no history of any neurological or psychiatric diseases. The genotype groups had a comparable socio-economic background and educational level. The study was approved by decision of the ethics committee of the University of Münster. All subjects gave written informed consent before any of the study procedures were commenced.

Genotyping

Genotyping of TNF-α -308G→A (rs1800629) located on chromosome 6p21.3 (position 31651010 5′ to the gene (possibly promoter/enhancer region) was carried out following published protocols applying the multiplex genotyping assay iPLEX for use with the MassARRAY platform (Oeth et al. 2007). The TNF-α -308G→A SNP (Hajeer and Hutchison 2001; Wilson et al. 1997) denotes a G(TNFα1)→A(TNFα2) single nucleotide exchange (Rainero et al. 2004). The -308A allele has been found to confer stronger transcriptional activity than the -308G allele. The genotyping completion rate was at 100%. Genotypes were determined by investigators blinded for the study.

Task

To measure the interrelation of attentional processing and response selection processes, we conducted a modified flanker task (Kopp et al.

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1996). In this task, vertically arranged visual stimuli were presented. The target-stimulus (arrowhead or circle) was presented in the center with the arrowhead pointing to the right or left. The central stimuli were flanked by two vertically adjacent arrowheads that pointed in the same (compatible) or opposite (incompatible) direction as the target. The flankers preceded the target by 100 ms (SOA 100 ms). The target was displayed for 300 ms. The response-stimulus interval was 1,600 ms. Flankers and target were switched off simultaneously. Four blocks of 105 stimuli each were presented in this task. Compatible (60%) and incompatible stimuli (20%) and nogo stimuli (circle; 20%) were presented randomly. The subjects had to react with the right or left thumb depending on the direction of the central arrowhead and to refrain from responding to circles.

Data processing and analysis

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 four electrodes above and below both eyes (IO1, IO2, FP1, FP2), and the horizontal EOG from two electrodes at the outer canthi of the eyes (LO1 and LO2). These electrodes were used for ocular artifact correction. The forehead was used as ground. The primary reference was Cz and re-referenced to linked mastoids for data analysis of the N1, N2, and P3. Electroencephalograpic and -oculographic (EEG and EOG) data were sampled with 500 Hz (Acquire, Neuroscan) and stored continuously on a PC hard disk together with stimulus and response markers. All impedances of electrodes were <8 kΩ. The data were analyzed off-line. The data were filtered using a band-pass filter from 0.5 to 16 Hz. Filtering using a low-pass filter of 16 Hz does not affect the measurement of the N1 and subsequent components. A recent study for example (Wascher et al. 2009) used an even stronger low-pass filter of 8 Hz. Even this stronger filter does not distort the amplitude of the N1 (see: Wascher et al. 2009 for a more detailed discussion of this topic). The high-pass filter was set to avoid that slow drifts contaminate the measurements of the ERPs.

EEG segments beginning 200 ms before and ending 1,000 ms after the stimulus were cut out (baseline: 200 ms till target stimulus presentation). These segments were checked off-line for artifacts (amplitude criterion ± 80 μV; low activity criterion 0.1 μV for 100 ms). The influence of remaining eye movements on electrocortical activity was corrected by the algorithm proposed by Gratton, Coles, and Donchin (1983). The ERP data were re-referenced to linked mastoids.

For the choice of electrodes, we used a scalp topography-driven strategy, i.e., these electrodes were included in data analysis that clearly revealed the respective ERP component. The N1 on the flanker was defined as the most negative peak within a time range of 0 (i.e., time point of target presentation) and 100 ms. The N1 was quantified at electrodes P3, P4, P7, and P8 and measured against the baseline. 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 to 260 ms after target onset until the N2 peak. The N2 was quantified at electrodes Fz and FCz as these electrodes revealed the maximum of the N2. The P3 was quantified at electrode Pz (measured against prestimulus baseline) and defined as the most positive peak within the time window of 300–600 ms. These electrodes were chosen because potentials are known to be strongest at these sites. Only trials with correct reactions were used for data analyses. All amplitude measures used the peak of the specific component (relative to baseline or, in case of the N2, relative to the preceding P2 peak). This peak measurement is favorable to the measurement of the mean amplitude in a predefined time interval because these mean amplitude measures contain more variance and are hence less reliable. The whole quantification procedure is comparable to Willemssen et al. (2009).

Additionally the s-LRP was evaluated. Here activity was measured using the electrodes C3 and C4 (Seiss and Praamstra 2004). Before LRP calculation, the current source density (CSD) of the signals was calculated (Nunez et al. 1997), replacing the potential at each electrode with the CSD, thus eliminating the reference potential (Yordanova et al. 2004). Because of the low signal-to-noise ratio of LRPs, a jackknifing procedure was applied before data quantification (Ulrich and Miller 2001). To obtain the jackknifed mean LRPs onset score or amplitude ji for each participant i ($i = 1 \dots n$), first, n grand-average waveforms are calculated across participants by successively omitting every participant once. For each of the n grand-average waveforms, the LRP onset or amplitude is measured. This results in n jackknifed LRP onset or amplitude scores ($j \dots jn$), with each ji being based on the data from all participants but i (see: Stahl and Gibbons 2004). Jackknifing reduces noise with the effect of a more reliable onset latency and peak amplitude measurement. The onset latency and amplitude of the e- and late LRP was defined as that point in time where the LRP reaches a value of 20% of its peak amplitude (e.g., Stahl and Gibbons 2004). The baseline of the stimulus-locked LRP was at -300 to -200 ms before stimulus onset. The procedure is comparable to Beste et al. (2008a, 2009b).

Statistical analysis

Behavioral parameters (RT, error rates) were analyzed in separate repeated measures ANOVAs with the within-subject factor "compatibility" (compatible vs. incompatible) and the between subject factor "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 genotype group. The P3 was subjected to a similar ANOVA model using the variable of the electrode Pz only. The N1 was analyzed in a repeated measures ANOVA using the within-subject factors electrode (P3, P7, P4, P8) and compatibility (compatible vs. incompatible) and the between subject factor genotype group. As jack-knifing leads to a massive reduction of variance in the LRP data, these F-values were adjusted using the method described by Ulrich and Miller (2001) and denoted as F_{corr} in the results section of the LRP data (Beste et al. 2009). 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 $z < 0.6$; $P > 0.3$; 1-tailed). For the descriptive statistics, the mean and standard error of the mean are given.

RESULTS

Behavioral data

RTs were longer for the incompatible [427 ± 7 (SD) ms] than for the compatible condition [399 ± 8 ms; $F(1,94) = 242.3$; $P < 0.001$; $\eta = 0.72$]). RTs were longer in the AA/AG (438 ± 10 ms) than for the GG genotype group (387 ± 9 ms) [$F(1,94) = 12.82$; $P = 0.001$]. Compatibility effects were differently large for the AA/AG and the GG genotype groups as indicated by the interaction compatibility × genotype [$F(1,94) = 52.60$; $P < 0.001$; $\eta = 0.35$; AA/AG genotype: compatible = 418 ± 10, incompatible = 459 ± 11; GG genotype: compatible = 380 ± 8, incompatible = 395 ± 9]. A univariate ANOVA across the difference in RTs between the incompatible and the compatible condition (compatibility effect) revealed that the compatibility effects were larger in the AA/AG (40 ± 3 ms) than in the GG genotype group [15 ± 3 ms; $F(1,94) = 49.68$; $P < 0.001$; $\eta = 0.34$].

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Error rates were higher in the incompatible (6.1 ± 0.2) compared with the compatible condition [2.5 ± 0.1; $F(1,94) = 265.42$; $P < 0.001$; $\eta = 0.73$] and also higher in the AA/AG genotype (4.9 ± 0.2) group compared with the GG genotype group [3.7 ± 0.2; $F(1,94) = 14.66$; $P < 0.001$; $\eta = 0.13$]. The interaction compatibility × genotype group was not significant [$F(1,94) = 0.26$; $P > 0.6$; $\eta = 0.003$].

Neurophysiological data

N1 EFFECTS. The scalp-topographical plots in Fig. 1 reveal a typical N1 topography. As depicted in the figure, activity related to the N1 covered electrodes P3, P4, P7, and P8. No other electrodes revealed activation related to the N1. Hence these four electrodes were included into data analysis.

The repeated measures ANOVA across the N1 amplitudes revealed that the N1 was strongest at electrode P7 (−4.68 ± 0.2) and lower at electrode P8 (−3.9 ± 0.1), differing from each other ($P = 0.01$). Amplitudes at electrodes P3 (3.19 ± 0.2) and P4 (3.16 ± 0.2) were lower than at P7 and P8 but did not differ from each other [$P > 0.9$; $F(3,282) = 114.32$; $P < 0.001$; $\eta = 0.54$]. This electrode effect was further modulated by the factor genotype group as indicated by the interaction electrode × genotype group [$F(3,282) = 13.86$; $P < 0.001$; $\eta = 0.13$]. Subsequent repeated measures ANOVAs for each genotype group separately revealed that this interaction was due to a larger electrode effect in the AA/AG genotype group [$F(3,120) = 65.84$; $P < 0.001$; $\eta = 0.62$] than in the GG genotype group [$F(3,162) = 39.74$; $P < 0.001$; $\eta = 0.42$]. In both groups, post hoc tests revealed the electrodes P3 and P4 did not differ from each other ($P > 0.8$). Also there was no difference between electrode P7 and P8 ($P > 0.9$). In each genotype group, the N1 was stronger at electrode P7/P8 (AA/AG: −6.1 ± 0.3; GG: −3.17 ± 0.2), compared with P3/P4 (AA/AG: −4.13 ± 0.4; GG: −2.21 ± 0.3). There was a main effect genotype group [$F(1,94) = 37.60$; $P < 0.001$; $\eta = 0.28$], showing that the N1 was larger in the AA/AG genotype group (−.11 ± 0.3) than in the GG genotype group (−2.71 ± 0.2). All other main or interaction effects were not significant (all F's <0.9; $P > 0.4$). There were no main or interaction effects for the latency of the N1 (all Fs < 0.9; $P > 0.4$). Also when using the mean amplitudes in the time interval (i.e., between 0 and 100 ms), the pattern of results remained the same.

N2 EFFECTS. The topographical maps in Fig. 2 reveal a typical N2 topography centered around electrodes Fz and FCz. Hence these electrodes were included into data analysis. The N2 at electrode Fz and FCz is shown in Fig. 2 for compatible and incompatible trials separated for each genotype group.

The N2 was larger at electrode Fz (−5.3 ± 0.2), compared with FCz [−3.3 ± 0.2; $F(1,94) = 96.88$; $P < 0.001$; $\eta = 0.51$] and was also larger for incompatible (−6.2 ± 0.3) than for compatible trials [−2.5 ± 0.2; $F(1,94) = 161.96$; $P < 0.001$; $\eta = 0.63$]. The latter effect was different for the two genotype groups as indicated by the interaction compatibility × genotype group [$F(1,94) = 19.85$; $P < 0.001$; $\eta = 0.17$]. Subsequent univariate ANOVAs using the genotype group as between-subject factor in separate analyses of the compatible and incompatible condition revealed that the groups differed in the N2 on incompatible [$F(1,94) = 61.53$; $P < 0.001$; $\eta = 0.39$] but not in compatible trials [$F(1,94) = 1.06$; $P = 0.306$; $\eta = 0.01$]. On incompatible trials, the AA/AG genotype group revealed a lower N2 (−6.7 ± 0.3) than the GG genotype group (−8.2 ± 0.2). The main effect genotype group was significant

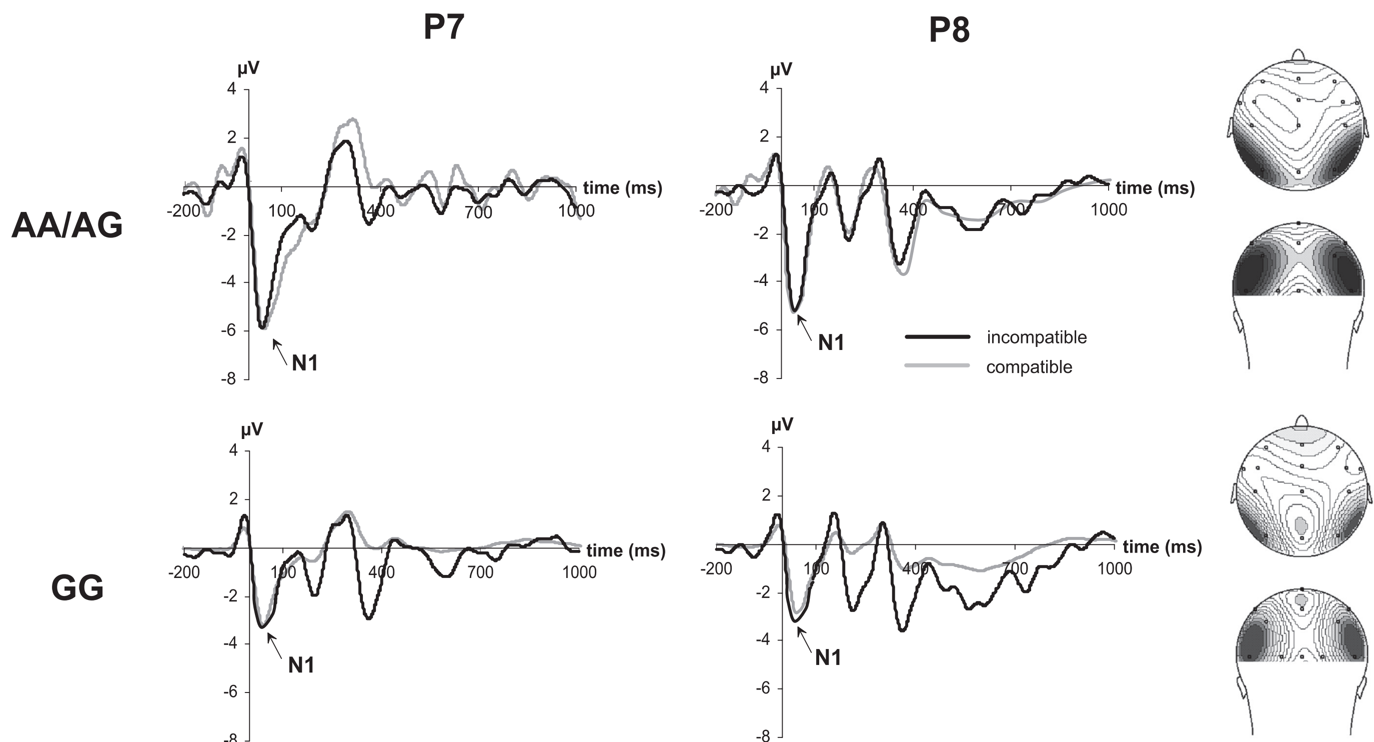

FIG. 1. The N1 on compatible (gray lines) and incompatible trials (black lines) is given. Time point 0 denotes the time point of stimulus delivery. *Left*: the N1 at electrode P7 is given; *right*: N1 at electrode P8. *Top*: the potentials of the AA/AG genotype group; *bottom*: the potentials for the GG genotype group. At the right side of the figure the scalp topographies of the N1 are given. Here, the scale denotes values between 0 (white) and negative values (grayscale).

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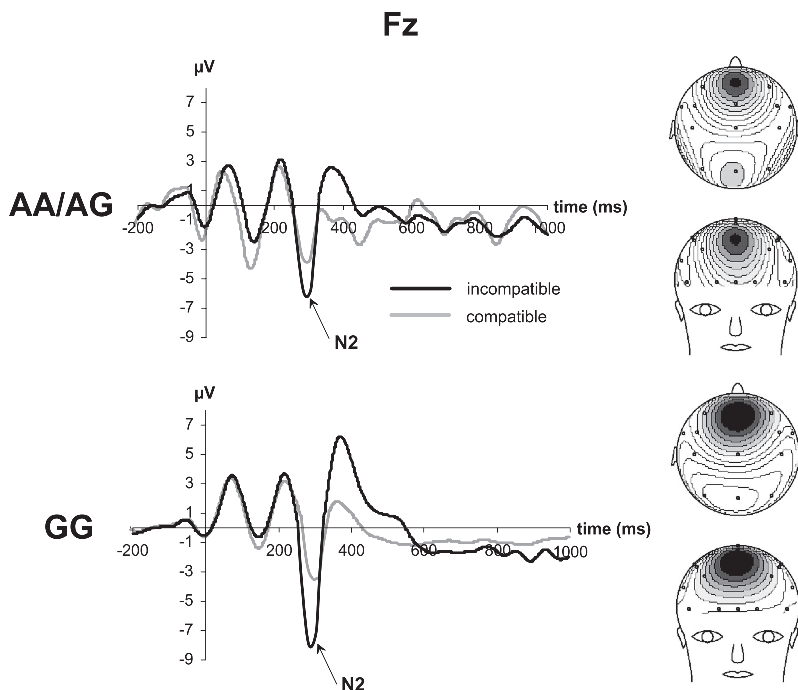

FIG. 2. The N2 on compatible (gray lines) and incompatible trials (black lines). Time point 0 denotes the time point of stimulus delivery. *Top*: the N2 for the AA/AG genotype group is given. *Bottom*: the N2 for the GG genotype group is given. *Right*: the scalp topography of the N2 on incompatible trials is given. Here the scale denotes values between 0 (white) and negative values (grayscale).

too [$F(1,94) = 18.34$; $P < 0.001$; $\eta = 0.21$], showing that the N2 was generally smaller in the AA/AG genotype group (-4.8 ± 0.2) than in the GG (-7.1 ± 0.2) group. All other main or interaction effects were not significant (all Fs < 1; $P > 0.2$). Similar, there was no effect in the latencies (all Fs < 0.6; $P > 0.5$). Also when using the mean amplitudes in the time interval (i.e., between 190 and 260 ms), the pattern of results remained the same.

LRP EFFECTS. LRP traces averaged to the stimulus onset are shown in Fig. 3 for the incompatible trials.

The amplitude of the "dip" was stronger in the AA/AG genotype group (4.97 ± 0.02) than in the GG genotype group [13 ± 0.03; $F_{corr}(1,94) = 5.29$; $P = 0.021$]. Onset and peak latencies of the dip did not differ between genotype groups (all $F\text{s}_{corr} < 0.5$; $P > 0.4$). The amplitude of the LRP did not differ between groups [$F_{corr}(1,94) = 0.29$; $P > 0.9$]. Opposed to the dip, onset and peak latency of the LRP differed between genotype groups. The onset of the LRP was earlier in the GG genotype group (281 ± 0.3) than in the AA/AG genotype group [340 ± 0.2; $F_{corr}(1,94) = 4.82$; $P = 0.031$]. Similar, also the peak latency was delayed in the AA/AG (370 ± 0.3) genotype group compared with the GG genotype group [322 ± 0.2; $F_{corr}(1,94) = 4.96$; $P = 0.028$].

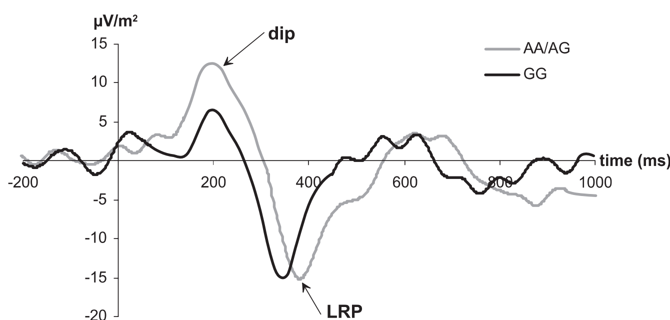

FIG. 3. The stimulus-locked lateralized readiness potential (LRP) on the incompatible trials is given. Time point 0 denotes the time point of stimulus delivery. Gray lines denote the potential for the AA/AG genotype group; black lines denote the potential for the GG genotype group.

P3 EFFECTS. The P3 on compatible and incompatible trials is shown in Fig. 4 for each genotype group separately.

For the amplitude, the ANOVA revealed there was a main effect condition, showing that potentials were lower in the incompatible (7.1 ± 0.3) than in the compatible condition [8.6 ± 0.3; $F(1,94) = 10.44$; $P < 0.001$; $\eta = 0.21$]. Neither the main effect genotype group nor the interaction compatibility × genotype was significant (all Fs < 0.56; $P > 0.5$). However, for the latencies, there was also a main effect compatibility [$F(1,94) = 54.34$; $P < 0.001$; $\eta = 0.39$]. The P3 peak latency was shorter in the compatible (350 ± 6) than in the incompatible condition (384 ± 4). This effect was further modulated by the factor group, as the interaction indicates [$F(1,94) = 17.71$; $P < 0.001$; $\eta = 0.16$]. While both genotype groups did not differ on compatible trials, the peak latency was longer in the AA/AG genotype group (413 ± 5) compared with the GG genotype group (370 ± 7) in the incompatible trials.

DISCUSSION

In the current study, we examined associations of the functional TNF-α −308G→A polymorphism (rs1800629) with SRC processes. The results show that the flanker N1 and the N2 reveal a dissociation between the AA/AG and the GG genotypes of the TNF-α −308G→A polymorphism. Compared with the GG genotype group, the N1 was enhanced in the AA/AG genotype group, while the N2 was reduced. Similar to the flanker-N1, the e-LRP amplitude was increased in the AA/AG genotype group compared with the GG genotype group, but no differences in the onset and peak latency of the e-LRP were evident. Yet for the subsequent occurring LRP, onset and peak latency were delayed in the AA/AG genotype group. Similar, also the P3b was delayed in the AA/AG genotype group. The behavioral data, showing increased com-

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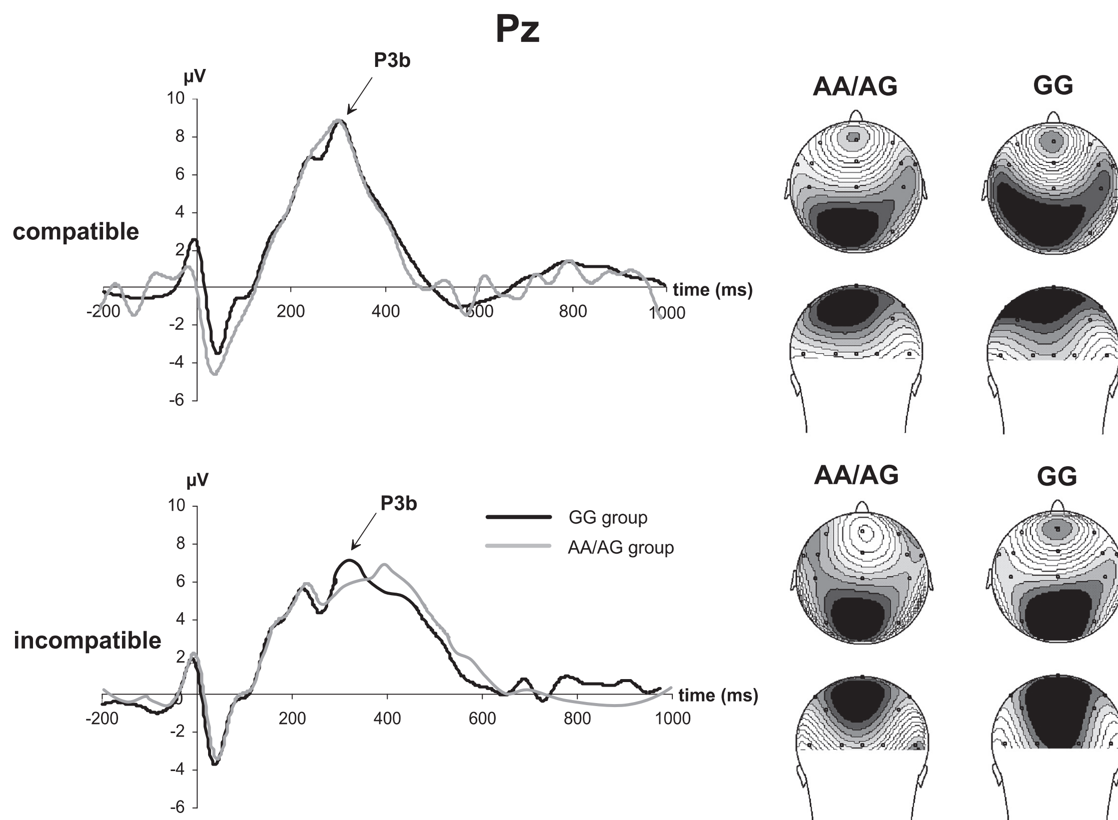

FIG. 4. The stimulus-locked P3 on compatible (*top*) and incompatible trials (*bottom*) at electrode Pz. Time point 0 denotes the time point of stimulus delivery. The scalp topography for the P3 on compatible and incompatible trials is given on the right. Gray lines denote the potential for the AA/AG genotype group; black lines denote the potential for the GG genotype group. Here the scale denotes values between 0 (white) and positive values (gray-scale).

patibility effects in the AA/AG genotype group, underline this pattern.

Attentional processing and action selection

The N1 is assumed to reflect attentional processes (e.g., Luck et al. 1995) and is hypothesized to be generated in lateral extrastriate cortex with a contribution of dorsal occipito-parietal and ventral occipital temporal structures (Gomez Gonzalez et al. 1994; Herrmann and Knight 2001). The increased flanker N1 in the AA/AG genotype group suggests that attentional processes are elevated in the AA/AG genotype group compared with the GG genotype group. As a possible explanation, this increase in attentional processing may be due to the suggested neuroprotective effects of TNF-α in occipital brain regions (Kaneko et al. 2008; Sriram and O'Callghan 2007). The increased attentional processing affects subsequent processing stages of action selection. Data from the e-LRP, reflecting automatic response activation (Falkenstein et al. 2006), show a stronger activation of the incorrect response hand on flanker presentation of the AA/AG genotype group. Hence the existence of an A allele seems to adversely affect action selection as driven by the flankers. This is likely a consequence of the increased saliency of distracting flanker information due to the enhanced processing as reflected in the N1. On a neuronal level, the process that may lead to the activation of the incorrect response hand (increased e-LRP) may be conceptualized as follows.

The basal ganglia are known to process sensation from various sensory modalities (Redgrave and Gurney 2006; Saft et al. 2008) and visual cortical areas are connected to the basal ganglia (Silkis 2006). Here visual input likely modulates basal ganglia networks that are important for action selection processes (e.g., Beste et al. 2010a,c; Redgrave and Gurney 2006). This may have important consequences for the process of action selection in the basal ganglia that operates as a "winner-take-all network" (WTA) (Beste et al. 2007, 2008b; Plenz 2003; Redgrave et al. 1999; Wild-Wall et al. 2008). Due to the WTA network, only the action is selected that produces strongest activation and is presented to the basal ganglia by cortical afferents. As a consequence of elevated attentional processing, flanker information in the basal ganglia may probably be stronger in the AA/AG genotype group compared with the GG genotype group and likely leads to a stronger activation of the incorrect response hand in the AA/AG genotype group. It is known that TNF alpha affects oligodendrocytes that in course can effect myelination (e.g., Huang et al. 2002), which could lead to differences in ERP latencies and response times. In fact, ERPs have been used as a measure to infer the status of myelination (Lippe et al. 2007; Picton and Taylor 2007).

However, the LRP onset and peak-latency reflecting processes of the selection of the desired response were also delayed in the AA/AG genotype group. This delay is in line with the average increase in RTs in the AA/AG genotype group and may also be explained within the WTA network of the basal ganglia:

Once activation in the basal ganglia has been established by cortical input, these initially established network states (i.e., due to flanker presentation) have to be suppressed to allow the

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establishment of a new focus that correlates with the novel cortical input (i.e., due to target presentation) (Plenz 2003). As outlined in the INTRODUCTION, the basal ganglia are especially likely to be adversely affected by TNF-α (McCoy and Tansey 2008; Sriram et al. 2006). Therefore the suppression of network states produced by the flankers may need more time in A allele carriers. Likely this may cause that the striatal focus on the target presentation is later established in the AA/AG genotype group compared with the GG genotype group, which may ultimately delay the LRP onset and peak latency as well as the RTs. However, it cannot be ruled out that the delay is simply because the activation of striatal networks on the flanker presentation was stronger in the AA/AG genotype group, and it may simply take longer to activate the desired response channel.

With respect to the N1 results, the current findings draw a picture in which some cognitive processes (i.e., attention) are enhanced, while processes related to the selection of the desired response become compromised by the same genotype (i.e., AA/AG genotype group). In this way, the results underline the complex and contradictory effects of TNF-α that may depend on various brain region specific factors (Sriram and O'Callaghan 2007) and suggest that the A allele of the TNF-α $-308G \rightarrow A$ polymorphism is not necessarily related to elevated cognitive performance. It is important to note that this highly depends on the way cognitive performance in examined.

Conflict processing

While it may be argued that the effects observed for the e-LRP (automatic activation of the incorrect response hand) may be predominantly driven by the increased N1 and hence do not reflect an immediate adverse modulation in the A allele group, the N2 results clearly suggest concomitant adverse effects on cognitive processes in the A allele group.

The N2 has been suggested to be related to action or response selection, which is intensified in case of response conflict (e.g., Folstein and Van Petten 2008; Gajewski et al. 2008; Van Veen and Carter 2002). Based on the increased flanker-N1 and e-LRP in the AA/AG genotype group, the distracting flanker information should lead to a higher degree of conflict and hence to an increased N2 in the AA/AG, compared with the GG genotype group. However, the opposite was the case because the N2 was decreased in amplitude in the AA/AG compared with the GG genotype group. The N2 was shown to be modulated by basal ganglia processes (Willemssen et al. 2009) and is known to be generated in the ACC (Botvinick et al. 2004), which is closely related to the basal ganglia (e.g., Chudasama and Robbins 2006). As the basal ganglia are assumed to consist of a WTA network, it is obvious to assume that the N2 is enhanced due to the stronger N1. However, as the basal ganglia are most likely to be adversely affected by TNF-α, the WTA network is compromised in its processing characteristics. This decrease in basal ganglia WTA network functioning, and hence basal ganglia-prefrontal circuits may affect the ACC, possibly leading to decreases in the N2. This would suggest that even though the conflict is evident it cannot be processed properly likely because basal ganglia-anterior cingulate interactions may be compromised. The N2 data may seem quite paradox with the data of the s-LRP. Assuming that the diminished incompatible N2 in the A allele group is a signal of a conflict processing deficit, why would the early LRP amplitude then be increased? Possibly the reason is due to different levels of conflict that are reflected by the s-LRP and N2: The s-LRP waveform is supposed to reflect premotor processes (e.g., Stahl et al. 2010). A previous study by our group revealed that the modulation of the N2 can be decoupled from the modulation of the s-LRP waveform and especially from the initial dip of the s-LRP (Beste et al. 2008a). In that study, the dip was not different between groups, but the N2 showed differences. We argued that the N2 does not reflect conflict on the motor or premotor level but, if at all, on a higher cognitive level only (Beste et al. 2008a). The current results support the suggestion that the N2 and the s-LRP may both reflect "conflict" but on different levels of processing; i.e., "cognitive" and "premotor" level.

However, the preceding interpretation of the N2 findings comprises an occipital-basal ganglia network to explain the results on a neuronal level, but there may also be an alternative interpretation of the results: Significant compatibility effects were found in both TNF allele groups: 40 ms in the AA/AG group and 15 ms in the GG group. This indicates that some level of conflict processing occurred in all participants but was greater in the former genotype group. The magnitude of this conflict processing effect might be mitigated by the cognitive processes associated with the increased N1. The N1 is not necessarily only an index of sensory gain control (Hillyard et al. 1999) but may also be interpreted as representative of stimulus discrimination (i.e., Hopf et al. 2002). Enhanced attentional processing as indexed by N1 could lead to better early differentiation of targets and flankers. This reduces subsequent conflict and leads to a reduction in N2. Yet the s-LRP data (the dip) shows that the enhanced flanker processing leads to an increased degree of conflict. Hence the alternative explanation of the findings does not seem justified.

Conclusions

In the current study, we analyzed subprocesses of SRC and their association with the TNF-α $-308G \rightarrow A$ SNP (rs1800629). Carriers of the A allele demonstrated elevated attentional processes as compared with G allele carriers. Subsequent processes of action selection and conflict processing were compromised in A allele carriers. This may be due to interrelated processes affecting mechanisms within the basal ganglia WTA network. The results underline the relevance of TNF-α as an important modulator of cognitive functions (McAfoose and Baune 2009). It is suggested that the dissociated effect of the A allele varies with respect to the examined cognitive function, putatively because specific brain areas are differentially affected by TNF-α.

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DISCLOSURES

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