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# Is the P3 amplitude reduction seen in externalizing psychopathology attributable to stimulus sequence effects?

**Casey S. Gilmore**[1,2,*], **Stephen M. Malone**[2], and **William G. Iacono**[2]

[1]Neurobehavioral Research, Inc

[2]Department of Psychology, University of Minnesota

## Abstract

P3 amplitude reduction (P3-AR) is associated with biological vulnerability to a spectrum of externalizing (EXT) disorders, such as conduct disorder, antisocial behavior, and substance use disorders. P3 amplitude, however, can be affected by the context within which it is measured, e.g. by the position of the target in the sequence of stimuli during an oddball task. We hypothesized that EXT-related P3-AR may be due to attention or working memory deficits in EXT that would weaken these stimulus sequence effects. Using a community-based sample of adolescent males, we examined the relationship between P3 and EXT as a function of the number of standards preceding the target. Higher EXT was associated with significantly smaller P3 amplitude, regardless of the number of standards preceding the target. These results suggest that P3-AR in EXT does not vary as a function of stimulus sequence, further supporting P3-AR as an endophenotype for EXT disorders.

### Keywords

Externalizing; P3; Sequence effects; Context; Endophenotype; Substance use

Reduced amplitude of the P3 event-related potential (ERP) has been associated with externalizing (EXT) psychopathology (Patrick et al., 2006). EXT represents a latent factor underlying the common comorbidity among a spectrum of disorders characterized by behavioral disinhibition, such as substance use disorders (SUDs), attention deficit/hyperactivity disorder (ADHD), conduct disorder, and antisocial behavior (Kendler, Prescott, Myers, & Neale, 2003; Krueger et al., 2002; Young, Stallings, Corley, Krauter, & Hewitt, 2000). Shared genetic effects account for the association between EXT and P3 amplitude reduction (P3-AR; Hicks et al., 2007), supporting the hypothesis that P3-AR is an endophenotype for general vulnerability to the spectrum of externalizing disorders. Little is known, however, about the mechanism underlying the P3-EXT effect. For example, does P3-AR stem from a core dysfunction in EXT, or might P3-AR depend in part on the context within which it is measured?

The sequence of standard stimuli preceding a target in the oddball task is a contextual variable that has been shown to affect the amplitude of P3. P3 amplitude tends to increase as the number of standards preceding a target increases (Gilmore, Clementz, & Buckley, 2005; Kilpelainen et al., 1999; Leuthold & Sommer, 1993; Polich & Bondurant, 1997; Squires,

*Corresponding author: cgilmore@nbresearch.com; Neurobehavioral Research, Inc., 1585 Kapiolani Blvd, Ste 1030, Honolulu, HI 96814, USA.
Address reprint requests to: Casey Gilmore, Neurobehavioral Research, Inc., 1585 Kapiolani Blvd, Ste 1030, Honolulu, HI 96814, USA; cgilmore@nbresearch.com

Wickens, Squires, & Donchin, 1976; Stadler, Klimesch, Pouthas, & Ragot, 2006). Variation in P3 amplitude reflects attentional allocation to stimulus processing and the updating of working memory for preceding stimulus events (Curry & Polich, 1992; Donchin, 1981; Polich, 1989; Putnam & Roth, 1990). Targets occurring after a long string of standards evoke a larger P3 because memory for the preceding stimuli has not been recently updated (Gonsalvez et al., 1999; Johnson & Donchin, 1982). Further, during the oddball task, subjective expectancies about the probability of a particular stimulus occurring are formed, which, in turn, modulates the attentional resources allocated to stimulus processing (Duncan-Johnson & Donchin, 1977; Duncan-Johnson, Roth, & Kopell, 1984; Gill & Polich, 2002; Matt, Leuthold, & Sommer, 1992). Thus, working memory updating processes and variable attention allocation based on subjective expectancies work in concert to differentially affect P3 amplitude as a function of stimulus sequence.

The ability to effectively utilize context during the oddball task underlies the P3 sequence effects found in normal participants. Contextual processing refers to actively holding information in mind in such a form that it can be used to mediate task appropriate behavior – a cognitive function that has been associated with attention and working memory processes (Cohen, Barch, Carter, & Servan-Schreiber, 1999; Cohen & Servan-Schreiber, 1992). Diminished attentional capacity and control and working memory capacity have been associated with externalizing-spectrum disorders (Barnett, Maruff, & Vance, 2009; Bogg & Finn, 2010; Finn & Hall, 2004; Harden & Pihl, 1995). The ability to effectively utilize context, then, may be compromised in EXT, which would manifest in atypical P3-related stimulus sequence effects (e.g. P3 in EXT may differ from controls for targets following a long string of standards, but not for targets following a short string of standards). The question is whether factors related to compromised contextual processing, such as inattention or decreased motivation and vigilance, might affect P3-AR in EXT.

The present report investigated how P3-AR in EXT varies with stimulus sequence pattern in the oddball task. The relationship between P3 and EXT was examined as a function of the number of standards preceding the target.

## Methods

### Participants

Participants were 499 male youths (Mean age = 17.5 years, SD = 0.4) from the older cohort of the Minnesota Twin Family Study (MTFS), a longitudinal and epidemiological study investigating the development of substance use disorders and related psychopathology. A comprehensive description of the MTFS is found in Iacono & McGue (2002). All participants and their parents gave written informed assent or consent as appropriate.

### Diagnostic Assessment

Lifetime presence of DSM-III-R (American Psychiatric Association, 1987) disorders, the diagnostic system in place at the time the present cohort of participants were assessed, was determined via in-person, structured interviews by trained clinical interviewers. Symptoms of nicotine dependence, alcohol dependence, and illicit drug dependence were assessed using an expanded version of the Substance Abuse Module of the Composite International Diagnostic Interview (Robins, Babor, & Cottler, 1987). An interview designed by MTFS staff (Holdcraft, Iacono, & McGue, 1998) was used to assess symptoms of conduct disorder and adult antisocial behavior (i.e., the adult criteria for Antisocial Personality Disorder). Mothers of the twins reported on the substance use and childhood antisocial behaviors of each twin through interviews using the parent version of the Diagnostic Interview for Children and Adolescents (Reich, 2000). Symptoms were assigned on the basis of a

consensus, “best-estimate” approach (Leckman, Scholomskas, Thompson, Belanger, & Weisman, 1982) combining mother and twin interview data.

Our measure of Externalizing consisted of the first principal component (which accounted for 59% of the variance) of the log-transformed symptom counts of conduct disorder, adult antisocial behavior, alcohol abuse/dependence, nicotine dependence, and illicit drug abuse/dependence (c.f. Hicks, et al., 2007; Patrick, et al., 2006).

## Psychophysiological Assessment

A rotated-heads visual oddball task (Begleiter et al., 1984) was used. Each of the 240 stimuli comprising the task was presented on a computer screen for 98 ms, with the inter-trial interval, during which subjects fixated on a dot in the center of the screen, varying randomly between 1 and 2 seconds. Two-thirds of the trials consisted of a plain oval, to which no response was required. On the remaining third of the trials, participants saw a superior view of a stylized head, in which a nose and one ear were depicted on the oval. Subjects were required to respond to these “target” trials by pressing a button on either the left or right armrest of their chair, corresponding to the side of the head on which the ear appeared. On half the target trials the nose pointed up (such that the left ear appeared on the left side of the screen; an easy discrimination), while on the other half of target trials the head was rotated 180° so that the nose pointed down (left ear appeared on the right side of the screen; a hard discrimination).

**Electroencephalographic (EEG) data acquisition—**A Grass model 12A Neurodata Acquisition System recorded EEG and electrooculographic (EOG) data at a sampling rate of 256 Hz and filtered from 0.01–30 Hz (6 dB/octave rolloff). EEG, referenced to linked earlobes, was recorded from three parietal electrodes: on the midline at Pz, and over left and right hemispheres at P3 and P4, respectively. EOG was recorded using a pair of biopotential electrodes, one electrode placed superior to the eye and the other at the outer canthus. Impedances were kept below 5 kΩ for EEG and below 10 kΩ for EOG.

**EEG data processing and reduction—**Blinks and other ocular artifacts were corrected using the method of Gratton, Coles, and Donchin (1983). Trials consisted of 2 s of data, including a 500 ms prestimulus baseline. Trials with activity >100 μV were excluded from further processing. Averaged ERPs to targets were constructed separately according to the number of standards preceding the target: 1, 2, 3, or 4. P3 amplitude was defined as the point between 280 and 600 ms at which amplitude of the average waveform was maximal. Current analyses were performed on ERPs only from the Pz electrode and averaged over easy/hard target conditions.

## Data Analysis

We fit a general linear model in which continuous scores on the EXT factor was the between-subjects variable, the number of standards preceding the target (1, 2, 3, or 4) the within-subjects variable, and P3 amplitude was the dependent variable. We also performed this analysis with P3 latency as the dependent variable. Because twins violate the assumption of independence of observations, we used Generalized Estimating Equations in SAS PROC GENMOD. These take into account the nested nature of the sample and produce appropriate standard errors. We conducted a Type 3 analysis, which is similar to the standard ANOVA Type III sums of squares used in PROC GLM, except that likelihood ratios are used instead of sums of squares. This procedure produces a chi-square statistic for each effect.

## Results

For P3 amplitude, there was a significant main effect of externalizing ($\chi^2_{(1;\ N = 275)} = 5.54$, $p = .019$), wherein higher EXT was associated with significantly smaller P3 amplitude. The main effect of number of standards was significant, $\chi^2_{(3;\ N = 275)} = 51.29$, $p < .0001$. The greater the number of standards preceding the target, the larger the resulting P3 amplitude, with P3 increasing 1.5 μV as the number of preceding standards varied from 1 to 4 (the respective P3 amplitudes were 16.2, 16.9, 17.4, and 17.7 μV). A linear contrast of means was highly significant ($p < .0001$), whereas a quadratic contrast was not ($p = .613$). There was no interaction between EXT and number of standards. Higher EXT was associated with smaller P3 amplitude, and this relationship remained regardless of the number of standards preceding the target. Figure 1 illustrates this effect by comparing average ERPs for participants in the upper and lower deciles of the distribution of EXT factor scores, collapsing over “early” occurring targets (those preceded by 1 or 2 standards) and “late” targets (those preceded by 3 or 4 standards).

For P3 latency, the main effect of the number of standards preceding the target was also significant ($\chi^2_{(3;\ N = 275)} = 12.71$, $p = .005$). In general, when more standards preceded the target, latency was shorter (mean latencies were 448.8, 449.5, 445.1, and 445.9 ms, respectively, for the different conditions). However, this was not moderated by EXT ($\chi^2_{(3;\ N = 275)} = 1.28$, $p = .734$) nor was there a main effect of EXT ($\chi^2_{(1;\ N = 275)} = 0.30$, $p = .584$), in contrast to the findings for amplitude.

In order to further address the possibility that the relationship between P3 amplitude and EXT may reflect the fact that the task was more difficult for those higher in EXT or that they were less engaged in it, we also examined reaction time (RT) and a measure of discrimination accuracy. We performed correlations 1) between EXT and mean RT and 2) between EXT and d-prime. There were small, non-significant positive correlations between EXT and RT for both easy ($r = .065$) and hard ($r = .058$) target conditions (both p values $\geq .150$). There was a small, non-significant negative correlation between EXT and d-prime ($r = -.012$, $p = .716$).

## Discussion

The present report demonstrated that P3-AR in EXT was not affected by stimulus sequence effects. As expected, P3 amplitude increased as the number of standards preceding the target increased. P3-AR, however, remained associated with EXT regardless of the number of standards that preceded the target.

The finding that P3-related sequence effects are normal in those with an EXT disorder suggests that such individuals are able to effectively utilize context during the oddball task to form subjective expectancies about the probability of a target occurring. This ability to utilize context occurs despite previous findings of diminished attention and working memory abilities in EXT spectrum disorders (e.g. Barnett, et al., 2009; Bogg & Finn, 2010; Finn, 2002). It may be the case, however, that the oddball task is relatively easy, such that those with an externalizing disorder can compensate for the relatively low working memory and attention requirements. Previous studies showing compromised working memory and attention abilities in EXT disorders have used comparably more difficult measures, e.g. the Digit Span, Operation-Word Span, Spatial Span, and Auditory Consonant Trigram tests (Barnett, et al., 2009; Bogg & Finn, 2010; Finn, 2002). Thus, while the underlying neurophysiology responsible for P3-AR in EXT may be compromised, the concomitant executive functioning abnormalities may not be manifested unless the related cognitive systems are sufficiently taxed. This interpretation is consistent with other studies showing

abnormal brain activity alongside normal task performance in EXT spectrum disorders (e.g. Caldwell et al., 2005; Tapert et al., 2004).

Our finding that the P3-AR effect was not secondary to a contextual processing deficit also rules out another possible explanation for why P3-AR is associated with EXT. We found that all participants, including those high in EXT, showed the expected association between the number of standards preceding a target and P3 amplitude. This finding, along with our results showing no significant relationship between EXT and either reaction time or task performance, indicates that all participants were effectively engaged in the task, paying attention and processing the stimuli. Hence, the P3-AR effect cannot easily be attributable to uncooperativeness, poor motivation, or superficial inattention. Thus, P3-AR appears to tap a core dysfunction, lending further support to its status as an endophenotype indexing genetic risk for EXT.

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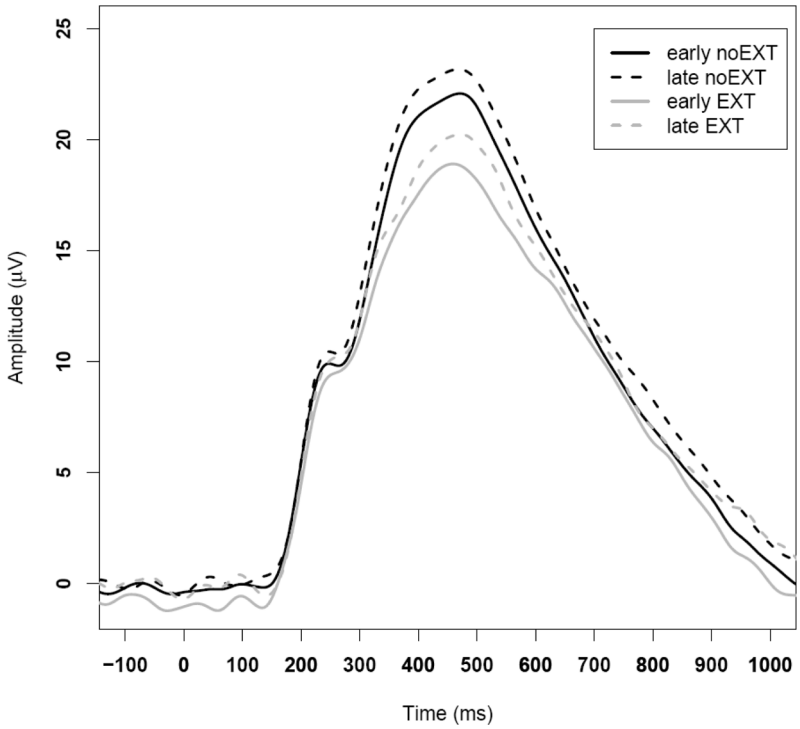


**Figure 1.**
Averaged target ERPs for participants in the upper (EXT) and lower (noEXT) deciles of the distribution of EXT factor scores, collapsing over “early” occurring targets (those preceded by 1 or 2 standards) and “late” targets (those preceded by 3 or 4 standards).