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Authors

Greenwood, Tiffany A
Swerdlow, Neal R
Sprock, Joyce
et al.

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Heritability of Acoustic Startle Magnitude and Latency from the Consortium on the Genetics of Schizophrenia

Tiffany A. Greenwood, PhD[1], **Neal R. Swerdlow, MD, PhD**[1], **Joyce Sprock, BA**[1], **Monica E. Calkins, PhD**[2], **Robert Freedman, MD**[3], **Michael F. Green, PhD**[4,5], **Raquel E. Gur, MD, PhD**[2], **Ruben C. Gur, PhD**[2], **Laura C. Lazzeroni, PhD**[6,7], **Gregory A. Light, PhD**[1,8], **Keith H. Nuechterlein, PhD**[5], **Allen D. Radant, MD**[9,10], **Jeremy M. Silverman, PhD**[11,12], **William S. Stone, PhD**[13,14], **Catherine A. Sugar, PhD**[5,15], **Debby W. Tsuang, MD, MSc**[9,10], **Ming T. Tsuang, MD, PhD**[1], **Bruce I. Turetsky, MD**[2], **David L. Braff, MD**[1], **Erica Duncan, MD**[16,17]

[1]Department of Psychiatry, University of California San Diego, La Jolla, CA

[2]Department of Psychiatry, University of Pennsylvania, Philadelphia, PA

[3]Department of Psychiatry, University of Colorado Health Sciences Center, Denver, CO

[4]VA Greater Los Angeles Healthcare System, Los Angeles, CA

[5]Department of Psychiatry and Biobehavioral Sciences, University of California Los Angeles, Los Angeles, CA

[6]Departments of Psychiatry and Behavioral Sciences and of Biomedical Data Science, Stanford University, Stanford, CA

[7]Department of Veterans Affairs Health Care System, Palo Alto, CA

[8]VISN-22 Mental Illness, Research, Education and Clinical Center (MIRECC), VA San Diego Healthcare System, San Diego, CA

[9]VA Puget Sound Health Care System, Seattle, WA

[10]Department of Psychiatry and Behavioral Sciences, University of Washington, Seattle, WA

[11]James J. Peters VA Medical Center, New York, NY

[12]Department of Psychiatry, Icahn School of Medicine at Mount Sinai, New York, NY

[13]Department of Psychiatry, Harvard Medical School, Boston, MA

Corresponding author: Erica Duncan, MD, Mental Health/116A, Atlanta Veterans Affairs Medical Center, 1670 Clairmont Road, Decatur, GA 30033, 404-321-6111, ext. 207532, erica.duncan@va.gov.

Contributors

All other authors except Dr. Duncan participated in obtaining funding for the study, subject recruitment, phenotyping, and validation of the clinical and endophenotype data. Dr. Swerdlow supervised 7-site quality assurance for startle measures. Joyce Sprock performed quality assurance for all data and coordinated database activities. Secondary analyses of startle data were designed by Drs. Greenwood and Duncan. The data were analyzed and interpreted and the manuscript was written by Drs. Greenwood and Duncan. All authors reviewed and approved the final manuscript.

[14]Massachusetts Mental Health Center Public Psychiatry Division of the Beth Israel Deaconess Medical Center, Boston, MA

[15]Department of Biostatistics, University of California Los Angeles School of Public Health, Los Angeles, CA

[16]Atlanta Veterans Affairs Healthcare System, Decatur, GA

[17]Department of Psychiatry and Behavioral Sciences, Emory University School of Medicine, Atlanta, GA

Abstract

Background—Latency of the acoustic startle reflex is the time from presentation of the startling stimulus until the response, and provides an index of neural processing speed. Schizophrenia subjects exhibit slowed latency compared to healthy controls. One prior publication reported significant heritability of latency. The current study was undertaken to replicate and extend this solitary finding in a larger cohort.

Methods—Schizophrenia probands, their relatives, and control subjects from the Consortium on the Genetics of Schizophrenia (COGS-1) were tested in a paradigm to ascertain magnitude, latency, and prepulse inhibition of startle. Trial types in the paradigm were: pulse-alone, and trials with 30, 60, or 120ms between the prepulse and pulse. Comparisons of subject groups were conducted with ANCOVAs to assess startle latency and magnitude. Heritability of startle magnitude and latency was analyzed with a variance component method implemented in SOLAR v.4.3.1.

Results—980 subjects had analyzable startle results: 199 schizophrenia probands, 456 of their relatives, and 325 controls. A mixed-design ANCOVA on startle latency in the four trial types was significant for subject group ($F(2,973)=4.45$, $p=0.012$) such that probands were slowest, relatives were intermediate and controls were fastest. Magnitude to pulse-alone trials differed significantly between groups by ANCOVA ($F(2,974)=3.92$, $p=0.020$) such that controls were lowest, probands highest, and relatives intermediate. Heritability was significant ($p<0.0001$), with heritability of 34–41% for latency and 45–59% for magnitude.

Conclusion—Both startle latency and magnitude are significantly heritable in the COGS-1 cohort. Startle latency is a strong candidate for being an endophenotype in schizophrenia.

Keywords

Schizophrenia; Latency; Heritability; Acoustic startle; Endophenotype

1.1 Introduction

Despite decades of work, the neurobiological and genetic underpinnings of schizophrenia (SCZ) remain incompletely understood. One approach to this problem is to search for biologically ascertained characteristics that distinguish subsets of individuals with SCZ. The hope is that these biological phenotypes, or endophenotypes, will identify subtypes of SCZ with more uniform genetics and neurobiology that can be found by the study of groups of subjects defined by the clinical diagnosis alone (Braff and Freedman, 2002; Gottesman and

Gould, 2003). This approach will hopefully lead to a better understanding of the genetic architecture of these endophenotypes and their corresponding neurobiology so that targeted treatments can be found based on an individual patient's specific neurobiology. According to Gottesman and Gould (2003), for a biological measure to be considered a valid endophenotype it must be found in subjects with the diagnosis at hand, be stable across clinical states, found within unaffected relatives of individuals with the diagnosis at a higher rate than in the general population, must segregate with the illness in affected families, and be significantly heritable (Gottesman and Gould, 2003).

The acoustic startle reflex (ASR) is a reflexive contraction of muscles in response to a sudden stimulus that is seen ubiquitously in mammals, including humans (Koch, 1999; Landis and Hunt, 1939). The startle reflex is thought to prepare the animal for a quick response when a potentially threatening stimulus is detected in the environment. The ASR and its modulations have been studied extensively in psychiatric disease and animal models. Prepulse inhibition of acoustic startle is the inhibition of the acoustic startle reflex when a preliminary small non-startling sound is presented shortly before the startle-eliciting stimulus. Prepulse inhibition is used extensively as an operational measure of sensorimotor gating (Graham, 1975; Hoffman and Searle, 1968; Koch, 1999). Prepulse inhibition is a very well researched and well accepted endophenotype of SCZ (Braff and Freedman, 2002; Braff et al., 2001; Braff et al., 1992; Swerdlow et al., 2008), the study of which has already yielded significant genetic findings in the Consortium on the Genetics of Schizophrenia (COGS) cohort (Greenwood et al., 2011; Greenwood et al., 2013).

Latency of acoustic startle is the time in milliseconds (ms) between presentation of the startling stimulus and the reflexive blink (in human subjects) or flinch (in rodents). It has received much less study than prepulse inhibition in published papers. The slowing of latency in SCZ compared to healthy controls is an index of slowed or inefficient information processing and has been seen in most papers that have reported on this measure (Braff et al., 1978; Braff et al., 1999; Geyer and Braff, 1982; Hasenkamp et al., 2010; Ludewig et al., 2002; Storozheva et al., 2016; Swerdlow et al., 2006; Weike et al., 2000), including a recent COGS publication (Swerdlow et al., 2018). However, a few papers did not detect this slowing of latency in SCZ (Braff et al., 1992; Mackeprang et al., 2002; Parwani et al., 2000; Kumari et al., 2005). For the most part these above-mentioned studies reported on peak latency, although in some, onset latency was analyzed (Braff et al., 1992; Braff et al., 1999; Hasenkamp et al., 2010; Mackenprang et al., 2002; Parwani et al., 2000; Weike et al., 2000).

There are several new findings that support the pursuit of further research on latency as a potential endophenotype for the study of SCZ. In a cohort of SCZ and control subjects and their first-degree relatives, startle latency measured up to 90% heritable (Hasenkamp et al., 2010). An important methodological issue in the study of endophenotypes is the stability across disease and treatment states. Importantly, slowed latency was not normalized in those SCZ subjects who were treated with antipsychotic medications (Fargotstein et al., 2018; Swerdlow et al., 2006). The North American Prodromal Longitudinal Study (NAPLS) followed young subjects at risk for developing psychosis and healthy controls for a period of two years. In this cohort a slowing of startle latency at baseline predicted which subjects went on to develop psychosis, thus indicating that this information processing measure was

abnormally slowed even before a full psychotic picture had emerged (Cadenhead et al., 2013). Importantly, a preliminary genetic association study of two separate cohorts of human subjects found an association of slowed (prolonged) latency with variants in the neuregulin gene (NRG1) (Smith et al., 2017) which has been identified as being associated with numerous other SCZ endophenotypes in the COGS-1 family study (Greenwood et al., 2007).

Latency provides an index of neural information processing speed (Hasenkamp et al., 2010) that could serve as a useful endophenotype in SCZ patients where it is slowed (Fargotstein et al., 2018). The COGS cohort was collected specifically to analyze a variety of SCZ cognitive and psychophysiological endophenotypes in order to discover their genetic underpinnings. The significant heritabilities of the endophenotypes assessed ranged from 24 to 55 percent, including 32 percent for prepulse inhibition (Greenwood et al., 2007). Data on latency of startle were collected and a prior publication indicated that latency was slowed in SCZ subjects in the COGS-2 case control cohort (Swerdlow et al., 2018), but the heritability of latency in the COGS-1 family study has not been published heretofore. As a preliminary step to examining the genetics of latency in the COGS cohort, and to endeavor to replicate the finding of highly significant heritability of this measure in a prior cohort (Hasenkamp et al., 2010), we conducted a heritability analysis of startle latency in the COGS cohort.

1.2 Methods

1.2.1 Cohort Description

Subjects from the COGS-1 family study cohort were recruited at seven sites: University of California San Diego, University of Colorado Health Sciences Center, University of California Los Angeles, Harvard University, Icahn School of Medicine at Mount Sinai, University of Pennsylvania, and University of Washington. (Calkins et al., 2007). Families were ascertained through probands who met DSM-IV-TR criteria for SCZ. Each family minimally consisted of a proband with SCZ, an unaffected sibling, and both parents. Unrelated healthy comparison subjects without personal or family history of psychosis were also recruited to serve as controls and matched as a group to the SCZ probands on age, gender, and ancestry. All subjects underwent a standardized clinical assessment using the Diagnostic Interview for Genetic Studies (DIGS) (Nurnberger et al., 1994). Subjects ranged from 18 to 65 years of age and received urine toxicology screens prior to assessment to exclude potential subjects with active drug use. Details of the ascertainment, diagnostic, and screening procedures are provided elsewhere (Calkins et al., 2007). Clinical symptoms were ascertained by means of the Brief Psychiatric Rating Scale—Expanded Version, Anxiety Scale (BPRS)(Ventura et al., 1993), the Scale for the Assessment of Positive Symptoms (SAPS)(Andreasen, 1984), the Scale for the Assessment of Negative Symptoms (SANS) (Andreasen, 1983), and the Global Assessment of Functioning Scale (GAF)(Hall and Parks, 1995). The 980 individuals from COGS-1 with analyzable startle data were used in these analyses, including 199 SCZ probands, 456 relatives, and 325 controls (Greenwood et al., 2013).

1.2.2 Acoustic Startle Acquisition—Startle testing was conducted according to methods outlined in prior publications (Calkins et al., 2007; Greenwood et al., 2007;

Swerdlow et al., 2007; Swerdlow et al., 2018). First, subjects were tested for hearing acuity by means of audiometer testing and excluded using a threshold of 45 dB at 1000 Hz. Startle testing was carried out with the SR-LAB system (San Diego Instruments, San Diego, CA). The eyeblink component of the acoustic startle reflex was captured by means of electromyographic (EMG) recording of the orbicularis muscle bilaterally with two electrodes placed over this muscle just under each eye. The startle stimuli were generated by the SR-LAB system and presented binaurally via headphones. A standard startle session designed to assess prepulse inhibition was used. The paradigm had 70 dB[A] broadband noise continued as background throughout the session. The startling stimuli were 115 dB[A] broadband noise bursts of 40 ms duration. Prepulse stimuli were 20 ms bursts of broadband noise of 86 dB[A] intensity presented 30, 60, or 120 ms prior to the startling stimuli. The session consisted of an initial block of five pulse-alone trials. This was followed by two blocks, each containing nine pulse-alone trials and nine of each of the prepulse+pulse trials (30, 60, or 120 ms between the prepulse and pulse stimuli). Trial types were presented in pseudorandom order separated by inter-trial intervals of 11–19 s. Additional “nostim” trials were included, consisting of EMG recording not paired with stimulus presentation in order to assure a stable baseline. The final block consisted of five pulse alone trials. The session lasted approximately 40 minutes. The SR-LAB system records EMG activity every ms for 250 ms following the startling pulses. The system has a data reduction program that evaluates each trial for error (baseline excessively high, no or inadequately low startle response) and calculates the maximum amplitude from a rolling average of the EMG response from 21 to 120 ms following the startling stimuli. The software also calculates the latency in ms between the startling stimuli and the maximum (peak) of the EMG response for each trial. Additional details of data reduction methods and procedures to minimize intersite variability are included in prior publications (Calkins et al., 2007; Swerdlow et al., 2007). Trials were included in the analyses if there were no errors for unstable baseline. Subjects were excluded if the mean startle magnitude in block 2 was less than 10 machine units, in accord with prior COGS publications (Swerdlow et al., 2018; Swerdlow et al., 2007).

1.2.3 Statistical Methods

Statistical differences in age, sex, and medication status between subject groups were determined by use of Chi square (for categorical variables) or analysis of variance (ANOVA, for continuous variables). BPRS, SAPS, and SANS ratings were compared by use of ANOVAs. Magnitude and latency variables are reported and analyzed as the means for the right and left eye. These variables were examined for normality: magnitude did not have a normal distribution so was log transformed prior to inclusion in statistical models. Startle magnitude to pulse-alone trials in Blocks 2–3 was compared across subject groups by means of an ANCOVA with age, sex, and site as covariates. In order to determine whether subjects’ hearing threshold contributed to startle magnitude, we compared hearing threshold between subject groups for a subset of subjects for whom this variable was available by means of an ANCOVA with age, sex, and site as covariates. We also ran correlations between hearing threshold and startle magnitude. For purposes of clarification we also examined the correlation between latency and magnitude for each of the trial types for each subject group. Mean latency across trials in Blocks 2 and 3 was assessed between the three subject groups

by means of mixed-design ANCOVA with trial type as a repeated measure factor, subject group as a between group factor, and age, sex, and site as covariates. The latency ANCOVA was also repeated with the addition of magnitude to pulse-alone trials in Blocks 2–3 as an additional covariate.

Heritability of startle magnitude and latency was analyzed in accord with methods published in our prior paper examining heritability of prepulse inhibition (Greenwood et al., 2007; Greenwood et al., 2013). Briefly, the variance component method implemented in SOLAR v.4.3.1 was used to obtain heritability (h^2) estimates by comparing a "full" model, which assumes that some fraction of the phenotypic variation is explained by genetic factors, to a "reduced" model, which assumes that no variation is explained by genes (Greenwood et al., 2007). Factors such as age, sex, and recruitment site were assessed for their significance as covariates, and those showing a significant ($p < 0.05$) association were retained in the heritability analysis. These analyses used normalized trait values and were restricted to 630 individuals from informative families. A correction was also made for ascertainment bias, since families were recruited through the identification of a proband with SCZ and were thus not representative of the general population (Almasy and Blangero, 1998; Beaty and Liang, 1987; Greenwood et al., 2007).

1.3 Results

1.3.1 Demographic Results

980 subjects with analyzable startle results were included in the analysis. Demographic characteristics of the sample are shown in Table 1. The subject groups were significantly different on age and sex distribution and smoking status.

1.3.2 Between Group Startle Results

Means and standard deviations of startle magnitude and latency variables are shown in Table 2. Age, sex, and site were included as covariates in the model for magnitude and the mixed-design ANCOVA for latency. Smoking status was not significant in ANCOVA models on startle variables so was not included in the final models.

Magnitude to pulse-alone trials differed significantly between groups by ANCOVA ($F(2,974)=3.92$, $p=0.020$, $\eta^2=0.008$) such that controls were lowest, probands highest, and relatives intermediate. In this model, age was highly significant ($F(2,974)=83.68$, $p<0.001$, $\eta^2=0.079$) but sex and site were not. Post hoc testing indicated that probands were higher than controls ($p=0.045$) as were relatives higher than controls ($p=0.008$). This model was conducted on subjects that included 14 relatives who had schizophrenia themselves. When the same model was run excluding these 14 subjects the results were unchanged. We compared hearing threshold between subject groups for a subset of subjects (n=556) for whom this variable was available by means of an ANCOVA with age, sex, and site as covariates. Hearing threshold was not significant ($F(2,550)=1.14$, $p=0.32$). A Pearson correlation between hearing threshold and startle magnitude to pulse-alone trials was likewise not significant ($r=-0.014$, $p=0.73$). Similarly, hearing threshold was not significantly correlated with magnitude to the other trial types.

We examined the correlation between latency and magnitude for each of the trial types for each subject group. As shown in Table 2, there were significant negative correlations in control subjects such that higher magnitude was associated with faster latency. Correlations were not significant for relatives or SCZ subjects.

A mixed-design ANCOVA on startle latency with the four trial types as a repeated measures factor (pulse-alone, 30 ms prepulse+pulse, 60ms prepulse+pulse, and 120 ms prepulse +pulse), subject group as a between group factor, and covariates of age, sex, and site was significant for subject group ($F(2,974)=4.46$, $p=0.012$, $\eta^2=0.009$). In this model age was highly significant ($F(2,974)=111.35$, $p<0.001$, $\eta^2=0.103$), as was sex ($F(2,974)=6.06$, $p=0.014$, $\eta^2=0.006$) but site was not significant. Post hoc testing indicated that on pulse-alone trials, probands were slower than relatives ($p=0.004$) and relatives were slower than controls ($p=0.040$). In the 30 ms prepulse+pulse trials, probands were slower than controls ($p=0.023$) and relatives ($p=0.002$). In the 120 ms prepulse+pulse trials, probands were slower than relatives ($p=0.021$). When log transformed magnitude to pulse-alone trials was added to the model the results were essentially virtually identical. These models were conducted on subjects that included 14 relatives who had schizophrenia themselves. When the same models were run, excluding these 14 subjects, the results were essentially unchanged.

1.3.3 Heritability Results

We conducted heritability analyses to explore how much of the variation in startle magnitude and latency can be attributed to inherited genetic factors. As shown in Table 3, all variables were found to be significantly heritable ($p<0.0001$), with heritability estimates in the moderate to substantial range (45–59% for magnitude; 34% for pulse-alone latency and up to 41% for prepulse-modified latency). Age was found to be a highly significant covariate for all startle magnitude and latency variables ($p<0.001$) with decreased magnitude and slowed (increased) latency. Sex was only associated with latency in pulse-alone trials ($p=0.001$) and for log transformed magnitude ($p=0.030$). Recruitment site was significantly associated with amplitude ($p<0.05$) but not latency. Log transformed magnitude was also significantly associated with latency in pulse-alone trials ($p=0.008$).

1.4 Discussion

The main objective of this analysis was to examine the heritability of latency and magnitude of acoustic startle in the COGS-1 family cohort. We used a standard COGS paradigm designed to evaluate prepulse inhibition of startle as well as latency and magnitude of startle (Calkins et al., 2007; Greenwood et al., 2007; Swerdlow et al., 2018). The main result herein was significant heritability of peak latency in all four trial types, with values ranging from 34 to 41%. As expected, we also found significant between group differences in latency such that probands with SCZ were slower information processors than controls and relatives, a finding consistent with the vast literature on information processing deficits in schizophrenia.

Heritability analyses of latency had not previously been conducted on COGS data. In the entire, extensive literature on startle in SCZ, only one prior publication reported on the

heritability of latency in a cohort of SCZ and control probands and their families, and found onset latency heritability ranging between 39% and 90% across trial types, and peak latency ranging from 29% to 68% (Hasenkamp et al., 2010), but the sample size was rather small and this finding was never replicated. The heritability reported in the current paper is for peak latency, and falls within the range reported in the Hasenkamp et al. study.

The potential importance of startle latency stems from several prior findings. Slower latency has previously been found in SCZ compared to controls in several prior reports dating back to the Callaway lab report of 1978 (Braff et al., 1978; Braff et al., 1999; Geyer and Braff, 1982; Hasenkamp et al., 2010; Ludewig et al., 2002; Swerdlow et al., 2006; Weike et al., 2000), including an analysis of a larger subject set from COGS (Swerdlow et al., 2018), although a few studies did not detect this difference (Braff et al., 1992; Mackeprang et al., 2002; Parwani et al., 2000). The North American Prodromal Longitudinal Study (NAPLS) is a multisite project that enrolled adolescent and young adult subjects at high risk for psychosis and healthy controls, testing them with several putative endophenotypes for schizophrenia and for clinical outcome in a 2-year follow up period. Startle latency at baseline was slower for those high risk subjects who converted to psychosis during the 2-year follow up than the healthy controls and in those who did not convert (Cadenhead et al., 2013). This finding suggests that slowed startle latency is an important early biomarker for SCZ risk and that antipsychotic medication effects do not account for slowing of latency since a majority of the high-risk subjects (approximately 80%) were not on antipsychotic medication at baseline (Seidman et al., 2016). Additional evidence to this point is that a recent analysis of latency in medicated and unmedicated subjects with schizophrenia found that latency in unmedicated subjects did not differ from those on atypical or on typical antipsychotics (Fargotstein et al., 2018), nor did medication status affect latency in a prior large cohort (Swerdlow et al., 2006). By contrast, in both these papers antipsychotic medication did affect prepulse inhibition (Fargotstein et al., 2018).

Slowing of startle latency is a putative index of generalized slowing of neural transmission and resulting slowed information processing (Hasenkamp et al., 2010). In that latency is significantly heritable and not normalized with medication, it could be useful as an endophenotype for schizophrenia in addition to the more widely researched prepulse inhibition of startle magnitude. An endophenotype (also called intermediate phenotype) is an abnormal measure of brain function that is seen in members of a diagnostic group and their affected and unaffected relatives, and is stable over time and across clinical states (Gottesman and Gould, 2003). The endophenotype approach to schizophrenia is widely endorsed as a potential for discovery of underlying genetics and individualized pathophysiology (Braff and Freedman, 2002; Greenwood et al., 2007; Tamminga et al., 2013) because the endophenotype may be closer to the underlying genetics and molecular abnormalities of subjects displaying that endophenotype than the larger set of patients with a complex disease such as schizophrenia. Furthermore, in keeping with its significant heritability, there is already some evidence of a genetic component to slowing of latency (Smith et al., 2017), although more work is needed in this area. An earlier study implicated a polymorphism of the dopamine D3 receptor gene affecting startle latency (Roussos et al., 2008). Supporting the potential importance of latency in the study of schizophrenia is the finding that slower latency is associated with greater cognitive impairment (Massa et al.,

2019), although an earlier paper with a smaller sample size failed to detect this association (Hasenkamp et al., 2011).

Startle magnitude was also found to be heritable in the current analyses, with values ranging from 45 – 59%. Heritability of startle magnitude to pulse-alone trials was 67% in the Hasenkamp et al. study (Hasenkamp et al., 2010). One other study in healthy humans has reported heritability of magnitude: it was 68–70% heritable in a cohort of non-psychiatric female twins (Anokhin et al., 2003). Magnitude differed between groups in the current analysis such that controls were lowest, relatives intermediate and probands had highest magnitudes, an orderly pattern of results. This is counter to the results reported in the larger COGS-2 case-control cohort wherein schizophrenia subjects had lower magnitude than healthy controls (Swerdlow et al., 2018). The reason for this discrepancy is unclear but likely related to differences in demographics and ascertainment methods between the COGS-2 case-control cohort and the COGS-1 family study reported in the current manuscript. In most prior studies reporting startle magnitude, schizophrenia subjects did not differ from controls (Braff et al., 1992; Braff et al., 2005; Cadenhead et al., 2000; Hasenkamp et al., 2010; Ludewig et al., 2002; Parwani et al., 2000; Perry et al., 2002; Storozheva et al., 2016; Swerdlow et al., 2006; Takahashi et al., 2008; Wynn et al., 2004) but in other studies, magnitude was lower in schizophrenia than control subjects (Kumari et al., 2005; Matsuo et al., 2016; Minassian et al., 2007; Quednow et al., 2006). Furthermore, psychosis-prone young adults had lower startle magnitude than controls (Simon and Giardina, 1992). It is theoretically possible that small differences in hearing threshold could have contributed to our finding. Subjects were excluded if their threshold was above 45 dB, so the thresholds of individual subjects did not have a high variance and would be very unlikely to contribute significantly to our findings. We were able to examine hearing threshold in a subset of our subjects for whom these data were available and found no difference between subject groups, nor was hearing threshold significantly correlated with startle magnitude. However, it must be mentioned as a caveat that we did not include auditory threshold as a covariate in our analysis of magnitude for our full set of 980 subjects. Because lower magnitude of startle has not often been reported in schizophrenia, this measure is not being investigated as an endophenotype for schizophrenia. However, there are both human and rodent data indicating that low startle magnitude may be a marker of vulnerability to cocaine use (Corcoran et al., 2011; Wheeler et al., 2017).

In summary, this analysis demonstrates a significant heritability of both startle latency and startle magnitude in the COGS-1 family study cohort. Startle latency is therefore a strong candidate for being a neurobiologically informative endophenotype in schizophrenia. Further work on this measure will examine the molecular underpinnings of this biomarker and of schizophrenia itself.

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Table 1:

Demographic and clinical information by diagnostic group

Variable	Controls	Relatives	Probands	Chi Sq/F-value	p-value
Demographics, n	325	456	199		
Sex, n(%)				64.59	<.0001
Male	135(41.5)	189(41.4)	146(73.4)		
Female	190(58.5)	267(58.6)	53(26.6)		
[a] Smoker, n(%)				113.96	<.0001
Yes	43(13.3)	66(14.70)	97(48.70)		
No	281(86.70)	383(85.30)	102(51.30)		
[b] Medication, n(%)				823.91	<.0001
Atypical Antipsychotic	0(0.0)	16(3.50)	170(86.30)		
Typical Antipsychotic	0(0.0)	3(0.70)	10(5.10)		
Both Typical and Atypical	0(0.0)	0(0.0)	10(5.10)		
No Antipsychotic Medication	320(100)	433(95.80)	7(3.60)		
Age, Years, Mean ± SD	34.78 ± 12.41	41.54 ± 13.43	33.23 ± 10.29	43.07	<.0001
Ratings, Mean ± SD					
[c] BPRS total	1.69 ± 0.92	2.12 ± 1.29	2.68 ± 1.52	37.05	<.0001
[d] GAF (past month)	84.40 ± 7.92	79.02 ± 12.64	45.35 ± 13.42	761.9	<.0001

[a] Missing in 8 subjects

[b] Missing in 11 subjects

[c] Missing in 64 subjects

[d] Missing in 56 subjects

Table 2.

Startle variables by diagnostic group

Variable	Controls	Relatives	Probands	[a] F-value	p-value
Startle testing, n	325	456	199		
Latency, ms, Mean ± SD				4.46	0.012
[b,c] Pulse Alone	64.94 ± 6.26	65.05 ± 5.68	65.69 ± 5.37		
[b,d] 30ms Prepulse+Pulse	56.84 ± 6.58	57.55 ± 6.14	58.14 ± 5.87		
60ms Prepulse+Pulse	56.05 ± 6.14	56.69 ± 6.09	56.30 ± 6.46		
[b] 120ms Prepulse+Pulse	59.08 ± 5.94	59.04 ± 6.11	60.01 ± 6.15		
Magnitude, μV, Mean ± SD					
[b,c,d] Pulse Alone	59.79 ± 41.25	63.1 ± 47.28	69.77 ± 52.57	3.56	0.029
[e] Correlation of latency and Magnitude (r, p-value)					
Pulse Alone	−0.128, 0.021	−0.049, 0.295	−0.012, 0.872		
30ms Prepulse+Pulse	−0.156, 0.005	−0.032, 0.492	−0.013, 0.858		
60ms Prepulse+Pulse	−0.157, 0.005	0.003, 0.949	0.002, 0.983		
120ms Prepulse+Pulse	−0.132, 0.017	0.012, 0.794	0.090, 0.208		

[a] F and p-value are for repeated measures ANOVA for 4 trial types

[b] In post hocs, Proband > Relative, $p < 0.05$

[c] In post hocs, Relative > Control, $p < 0.05$

[d] In post hocs, Proband > Control, $p < 0.05$

[e] Correlations of latency and magnitude for the same trial type

Table 3.

Results of heritability analyses (N=630)

			Covariates				
[a] Variable	[b] $h^2_r \pm SE$	p-value	Age	Sex	Site	Log Magnitude	[c] Prop Var
Magnitude							
Pulse-alone, Block 2	0.59 ± 0.09	<0.0001	<0.0001	0.03	0.012	-	0.1
Pulse-alone, Block 3	0.45 ± 0.09	<0.0001	<0.0001	0.429	0.029	-	0.09
Pulse-alone, Blocks 2–3	0.55 ± 0.10	<0.0001	<0.0001	0.084	0.017	-	0.1
Latency, Blocks 2–3							
Pulse-alone	0.34 ± 0.09	<0.0001	<0.0001	0.001	0.944	0.008	0.12
30ms	0.41 ± 0.09	<0.0001	<0.0001	0.095	0.804	0.052	0.11
60ms	0.34 ± 0.08	<0.0001	<0.0001	0.321	0.083	0.451	0.14
120ms	0.34 ± 0.10	<0.0001	0.001	0.688	0.893	0.06	0.01

[a] All variables are inverse normalized

[b] Heritability estimate after adjustment for significant covariates

[c] The proportion of trait variance explained by all significant covariates