

Original Article

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# Examining the independent and joint effects of genomic and exposomic liabilities for schizophrenia across the psychosis spectrum

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**Abstract**

**Aims.** Psychosis spectrum disorder has a complex pathoetiology characterised by interacting environmental and genetic vulnerabilities. The present study aims to investigate the role of gene–environment interaction using aggregate scores of genetic (polygenic risk score for schizophrenia (PRS-SCZ)) and environment liability for schizophrenia (exposome score for schizophrenia (ES-SCZ)) across the psychosis continuum.

**Methods.** The sample consisted of 1699 patients, 1753 unaffected siblings, and 1542 healthy comparison participants. The Structured Interview for Schizotypy-Revised (SIS-R) was administered to analyse scores of total, positive, and negative schizotypy in siblings and healthy comparison participants. The PRS-SCZ was trained using the Psychiatric Genomics Consortium results and the ES-SCZ was calculated guided by the approach validated in a previous report in the current data set. Regression models were applied to test the independent and joint effects of PRS-SCZ and ES-SCZ (adjusted for age, sex, and ancestry using 10 principal components).

**Results.** Both genetic and environmental vulnerability were associated with case-control status. Furthermore, there was evidence for additive interaction between binary modes of PRS-SCZ and ES-SCZ (above 75% of the control distribution) increasing the odds for schizophrenia spectrum diagnosis (relative excess risk due to interaction = 6.79, [95% confidential interval (CI) 3.32, 10.26],  $p < 0.001$ ). Sensitivity analyses using continuous PRS-SCZ and ES-SCZ confirmed gene–environment interaction (relative excess risk due to interaction = 1.80 [95% CI 1.01, 3.32],  $p = 0.004$ ). In siblings and healthy comparison participants, PRS-SCZ and ES-SCZ were associated with all SIS-R dimensions and evidence was found for an interaction between PRS-SCZ and ES-SCZ on the total ( $B = 0.006$  [95% CI 0.003, 0.009],  $p < 0.001$ ), positive ( $B = 0.006$  [95% CI, 0.002, 0.009],  $p = 0.002$ ), and negative ( $B = 0.006$ , [95% CI 0.004, 0.009],  $p < 0.001$ ) schizotypy dimensions.

**Conclusions.** The interplay between exposome load and schizophrenia genetic liability contributing to psychosis across the spectrum of expression provide further empirical support to the notion of aetiological continuity underlying an extended psychosis phenotype.

## Introduction

The psychosis spectrum ranges from serious, enduring, and disabling illness to transient, sub-threshold psychotic experiences in non-clinical populations (Guloksuz and van Os 2018). It represents a wide range of symptoms including aberrant thinking and reasoning, perceptual abnormalities, cognitive disturbance, as well as motivational and social deficits. Consistent with the extended psychosis phenotype model, prevalence is estimated at 5–8% for psychotic experiences in the general population, 3% for clinical psychotic disorders, and 0.5% for arguably the most severe end of the spectrum meeting diagnostic criteria for schizophrenia (van Os *et al.*, 2009).

The aetiological and pathophysiological theories of psychosis spectrum have evolved to encompass genetic and environmental factors and their interaction (EUGEI investigators, 2014). The concordance rates between twin pairs suggest the presence of genetic factors with heritability estimates of up to 80% for schizophrenia and 73% for the wider phenotype (Hilker *et al.*, 2018). More recent molecular genetic studies have confirmed that schizophrenia spectrum disorder, as a common complex trait, has a polygenic architecture, which is mainly shaped by many common allele variants with small effect sizes that are normally distributed among the general population (Schizophrenia Working Group of the Psychiatric Genomics Consortium, 2014). With the advent of the genome-wide association study approach, the Psychiatric Genomics Consortium has identified 145 significant loci associated with schizophrenia (Pardinas *et al.*, 2018). It is now possible to calculate an individual score summarising the level of genetic risk for schizophrenia, known as polygenic risk score for schizophrenia (PRS-SCZ) (Pardinas *et al.*, 2018).

Similarly, several environmental exposures have been associated with a schizophrenia spectrum disorder, such as childhood adversities, cannabis use, urbanicity, migration, ethnic minorities, hearing impairment, and perinatal factors (Linszen *et al.*, 2016;

Radua *et al.*, 2018; Stilo and Murray, 2019). In accordance with the diathesis-stress model, there is evidence supporting gene–environment interaction in the aetiology of schizophrenia (Guloksuz *et al.*, 2019) and mood disorders (Geoffroy *et al.*, 2013; Colodro-Conde *et al.*, 2018; Arnau-Soler *et al.*, 2019a, 2019b). A recent case-control study found evidence for additive interactions between molecular genetic liability for schizophrenia (i.e. PRS-SCZ) and emotional abuse, emotional neglect, sexual abuse, bullying, and regular cannabis use, suggesting that a multitude of environmental factors and PRS-SCZ are independently and jointly associated with schizophrenia (Guloksuz *et al.*, 2019).

To better accommodate the multiplicity of exposures associated with schizophrenia (Guloksuz *et al.*, 2018), a cumulative environmental exposure score for schizophrenia – exposome score for schizophrenia (ES-SCZ) – was recently designed and validated through predictive modelling approaches in training and validation data sets of two independent cohorts that followed identical measurement methods for environmental exposures (Pries *et al.*, 2019). This summary measure is generated using weighted coefficients derived from a single model to take into account the interdependency of exposures. Therefore, ES-SCZ prevents overestimation of the weights per exposure that are likely to occur when correlations between exposures are ignored, e.g. weighted estimates of individual exposures from meta-analyses or simple summation of exposures. Recent studies indicate that the ES-SCZ is associated with psychosis risk states (Guloksuz *et al.*, 2020) as well as mental and physical health (Pries *et al.*, 2020b) in the general population.

By leveraging aggregate scores of genetic (PRS-SCZ) and environmental liability (ES-SCZ) in the current study and in accordance with a previous study (Guloksuz *et al.*, 2019), we aimed to test gene–environment interaction across the psychosis spectrum in a multinational multicentre sample of patients diagnosed with schizophrenia spectrum disorder, their siblings, and healthy comparison participants.

## Methods

### Study population

Data were derived from the Workpackage 6 (WP6) of the European Network of National Networks studying Gene–Environment Interactions in Schizophrenia (EUGEI) and the Genetic Risk and Outcome for Psychosis (GROUP) studies, collected using uniform assessment schedules between 2010 and 2015 in the Netherlands, Turkey, Spain, and Serbia (Korver *et al.*, 2012). Both projects were approved by the Medical Ethics Committees of all participating sites and conducted in accordance with the Declaration of Helsinki. All respondents provided written informed consent and, in the case of minors, such a consent was also obtained from parents or legal guardians. Patients were diagnosed with schizophrenia spectrum disorders according to the DSM-IV-TR (average duration of illness since the age of the first contact with mental health services = 9.9 years). Unrelated controls with no lifetime psychotic disorder were recruited from the same population as the cases. Exclusion criteria for all participants were a diagnosis of psychotic disorder due to another medical condition, a history of head injury with loss of consciousness, and an intelligence quotient <70.

EUGEI WP6 ('vulnerability and severity') was a cross-sectional study specifically conducted to investigate the role of gene–environment interaction of the vulnerability and severity of schizophrenia spectrum disorder and its intermediate phenotypes in a family-based setting.

GROUP is a naturalistic longitudinal cohort study that started in 2004 in the Netherlands and Dutch-speaking part of Belgium and collected data at baseline, 3 and 6 years follow-ups over an approximate 10-year period, with the aim of studying the interplay of genetic and environmental factors impacting vulnerability and resilience in psychotic disorders. Individuals in the sibling group who manifested lifetime psychotic disorder over the study period were reassigned to the patient group.

Further details of the GROUP and EUGEI projects are provided elsewhere (Korver *et al.*, 2012; EUGEI investigators 2014). The current analyses used a merged data set of GROUP baseline data and EUGEI WP6 cross-sectional data including 1699 patients, 1753 siblings, and 1542 unrelated healthy comparison participants who were of Caucasian white ethnic origin and had available genotype data.

### Outcomes

#### Diagnosis of schizophrenia spectrum disorder

Patients were diagnosed with schizophrenia spectrum disorders according to the DSM-IV-TR. The diagnosis was confirmed by the Operational Criteria Checklist for Psychotic and Affective Illness (McGuffin *et al.*, 1991) in EUGEI WP6, and by the Schedules for Clinical Assessment in Neuropsychiatry (Wing *et al.*, 1990) and the Comprehensive Assessment of Symptoms and History (Andreasen *et al.*, 1992) in GROUP.

#### Schizotypy trait

In both GROUP and EUGEI, the Structured Interview for Schizotypy-Revised (SIS-R) was administered to siblings and healthy comparison participants. The SIS-R is a semi-structured interview containing 20 schizotypal symptoms and 11 schizotypal signs rated on a four-point scale (Kendler *et al.*, 1989; Vollema and Ormel, 2000). Symptoms are defined as verbal responses to standardised questions concerning, for example, magical ideation,

illusions, and referential thinking. Signs refer to behaviours that are rated by the interviewer such as goal-directedness of thinking and flatness of affect. Questions and rating procedures are standardised. Guided by previous research, 31 item scores were reduced *a priori* to two-dimensional scores representing the means of seven positive schizotypy items (covering the areas of referential thinking, psychotic phenomena, derealisation, magical ideation, illusions, and suspiciousness) and eight negative/disorganised schizotypy items (covering the areas of social isolation, sensitivity, introversion, restricted affect, disturbances in associative and goal-directed thinking, poverty of speech, and eccentric behaviour) (van Os *et al.*, 2020).

### Genetic and environmental liability measures

#### Exposome score for schizophrenia

The exposome score in the current analyses was calculated based on our previously validated estimates (Pries *et al.*, 2019) for constructing cumulative environmental load in this data set. Using the log odds from our previous report, we generated the ES-SCZ by summing log-odds weighted environmental exposures (each exposure defined as absent = '0' and present = '1') including cannabis use, hearing impairment, winter-birth, and childhood adversity domains (emotional and physical neglect, emotional, physical and sexual abuse, and bullying). The definition of each exposure conformed to previous work in this data set.

Childhood adversities were assessed using the Childhood Trauma Questionnaire (CTQ) Short Form (Bernstein *et al.*, 2003). This consists of 28 items, rated on a five-point Likert scale, measuring five domains of maltreatment (emotional and physical neglect; emotional, physical, and sexual abuse). The psychometric characteristics of the translated versions (Spanish, Turkish, Dutch, and Serbian) of the CTQ have been comprehensively studied (Sar *et al.*, 2004; Thombs *et al.*, 2009; Hernandez *et al.*, 2013). To dichotomise each childhood adversity domain (0 = 'absent' and 1 = 'present'), consistent with previous work in the EUGEI (Guloksuz *et al.*, 2019), we used the following cut-off scores for each domain:  $\geq 9$  for emotional abuse;  $\geq 8$  for physical abuse;  $\geq 6$  for sexual abuse;  $\geq 10$  for emotional neglect; and  $\geq 8$  for physical neglect.

Cannabis use was assessed by a modified version of the Cannabis Experiences Questionnaire (Barkus *et al.*, 2006) in the EUGEI WP6 (0 = 'none'; 1 = 'only once or twice'; 2 = 'a few times a year'; 3 = 'a few times a month'; 4 = 'once or more a week'; 5 = 'everyday'), and by the L section of the Composite International Diagnostic Interview (Robins *et al.*, 1988) in the GROUP (0 = 'none'; 1 = 'less than weekly'; 2 = 'weekly'; 3 = 'daily'). Consistent with previous work (van Winkel *et al.*, 2011; Pries *et al.*, 2018; Guloksuz *et al.*, 2019; Radhakrishnan *et al.*, 2019), a binary regular cannabis use variable was constructed by using the cut-off value of one or more per week during the lifetime period of most frequent use.

In accordance with previous studies investigating the association between season of birth and schizophrenia in the Northern hemisphere sites (Davies *et al.*, 2003), the high-risk birth period was defined based on the winter solstice (December–March), and a binary winter-birth exposure was constructed. Hearing impairment was defined based on self-reported hearing impairment in the last 12 months (0 = 'absent' and 1 = 'present').

The history of bullying by peers (emotional, psychological or physical violence) before 17 years of age was assessed using the

short version of the Retrospective Bullying Questionnaire (Hunter *et al.*, 2004; Schäfer *et al.*, 2004) that measures the severity of the bullying experience: 0 = 'none'; 1 = 'some (no physical injuries)'; 2 = 'moderate (minor injuries or transient emotional reactions)'; 3 = 'marked (severe and frequent physical or psychological harm)'. Exposure to childhood bullying was dichotomised using  $\geq 1$  as the cut-off point (0 = 'absent' and  $\geq 1$  = 'present').

#### *Polygenic risk score for schizophrenia*

Samples of all individuals were genotyped at Cardiff University Institute of Psychological Medicine and Clinical Neurology, using a custom Illumina HumanCoreExome-24 BeadChip genotyping arrays containing probes for 5 70 038 genetic variants (Illumina, San Diego, CA). Genotype data were called using the GenomeStudio package and transferred into PLINK format for further analysis. Quality control was conducted in PLINK v1.07 (Purcell *et al.*, 2007) or with custom Perl scripts. Variants with a call rate  $< 98\%$  were excluded from the data set. Hardy-Weinberg equilibrium  $p$  value was calculated separately in Turkish, northern European, and southern European samples. Variants with Hardy-Weinberg equilibrium  $p$  value  $< 1 \times 10^{-6}$  in any of these three regions were excluded from the data set. After QC, 5 59 505 variants remained. Samples with a call rate  $< 98\%$  were excluded from the data set. A linkage disequilibrium pruned set of variants was calculated using the `- indep-pairwise` command in PLINK (maximum  $r^2 = 0.25$ , window size = 500 single nucleotide polymorphisms (SNPs), window step size = 50 SNPs) and used for further analyses. Homozygosity  $F$  values were calculated using the `- het` command in PLINK, and outlier samples ( $F < -0.11$  or  $F > 0.15$ ) were excluded. The genotypic sex of samples was calculated from X chromosome data using the `check-sex` command in PLINK, and samples with different genotypic sex to their database sex were excluded. Identity-by-descent values were calculated for the sample in PLINK. Samples with one or more siblings among the genotyped samples according to the database but no identified genotypic siblings (defined as  $\hat{p} > 0.35$  and  $< 0.65$ ) were excluded. After these were removed from consideration, samples with two or more siblings in the database that were not supported by the genotypic data were also excluded. After visually observing the clustering of errors by genotyping chips, we decided to exclude chips with a high proportion of errors. All samples on chips with five or more sample exclusions due to heterozygosity or call rate (out of 12 possible samples) were excluded. All samples on chips with four or more sample exclusions due to sex or relative checks were also excluded unless their identity was corroborated by concordance between database and genotype relatedness data with a sample on another chip. Principal components (PCs) were calculated in PLINK using linkage disequilibrium (LD) pruned variants after combining the data set with the Thousand Genomes reference data set. After quality control, genotypes were imputed on the Michigan Imputation Server using the Haplotype Reference Consortium reference panel (version 1.1) and the programmes Eagle for haplotype phasing and Minimac3 for imputation (Das *et al.*, 2016; Loh *et al.*, 2016). After imputation, variants with an imputation  $r^2 > 0.6$ , minor allele frequency (MAF)  $> 0.1\%$  and call rate  $> 99\%$  were retained (82 77 535 variants). Best-guess genotypes were generated from genotype probabilities using PLINK.

PRS-SCZ was constructed using summary statistics from the Psychiatric Genomics Consortium (PGC2) genome-wide association study in both samples (Schizophrenia Working Group of the Psychiatric Genomics Consortium, 2014). There was no

overlap between the PGC2 and the current data sets. Clumping was performed in imputed best-guess genotypes for each data set using PLINK (maximum  $r^2 = 0.2$ , window size = 500 kb, minimum MAF = 10%, minimum INFO score = 0.7), and variants within regions of long-range LD around the genome (including the major histocompatibility complex) excluded (Price *et al.*, 2008). PRS-SCZ were then constructed from best-guess genotypes using PLINK at 10 different  $p$ -value thresholds (PT = 1, 0.5, 0.3, 0.2, 0.1, 0.05, 0.01,  $1 \times 10^{-4}$ ,  $1 \times 10^{-6}$ ,  $5 \times 10^{-8}$ ). Consistent with previous research in the field (Allardyce *et al.*, 2018; Sorensen *et al.*, 2018) and previous work in this data set, we used PT = 0.05 for our primary analysis, as this threshold optimally captures liability to the disorder in the Psychiatric Genomics Consortium analysis (Schizophrenia Working Group of the Psychiatric Genomics Consortium, 2014).

#### *Statistical analysis*

Stata software version 15.0 was used for the analysis (StataCorp, 2017). Supplementary Table S1 reports missing data. The analyses were conducted on both multiple imputed data and raw data. Under the assumption of missing at random, the multiple imputation chained equation (Royston and White, 2011) was applied with 20 imputations restricted to in-range values (relative efficiency  $\geq 99\%$ ). ES-SCZ was calculated after imputing missing values of the environmental exposures (cannabis use, hearing impairment, winter-birth, and childhood adversity domains). All the analyses were run on multiple imputed data and pooled using Rubin's rules (Rubin, 2004). To test gene-environment interaction, additive models were chosen over multiplicative models prior to data collection (EUGEI consortium meeting, 14 December 2013), consistent with previous work (Guloksuz *et al.*, 2019), and given that they provide a superior representation of biological synergy (Rothman, 1976) and inform public health decisions within the sufficient cause framework (Rothman *et al.*, 1980; Kendler and Gardner, 2010). For all analyses, random intercept multilevel mixed regression models, taking into account the clustering of participants within countries, were applied. Models including PRS-SCZ were *a priori* adjusted for ancestry using 10 PCs and adjusted models included age and sex as covariates. The nominal significance threshold was set to  $p = 0.05$ .

For the case-control analyses, as utilised in previous studies (Guloksuz *et al.*, 2019; Guloksuz *et al.*, 2020), ES-SCZ and PRS-SCZ were dichotomised at the quartile cut-off points based on the control distribution within each country (to account for differences between countries that may arise due to ethnic and geographical variation). The highest quartile was considered the binary risk state for schizophrenia (hereafter PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub>). Multilevel logistic regression models were applied to test the independent and joint effects of PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub> (independent variables) on the diagnosis of schizophrenia (i.e. case-control status; dependent variable). Departure from additivity was tested using the relative excess risk due to interaction (RERI) (Knol and VanderWeele, 2012; VanderWeele and Knol, 2014). RERI greater than zero was defined as a positive deviation from additivity and considered significant when the 95% confidence interval (CI) did not contain zero. Conforming to early work in this sample, we applied the delta method to calculate the RERI using the odds ratios derived from the model (Guloksuz *et al.*, 2019). Furthermore, sensitivity analyses were conducted using the bootstrap percentile method to estimate additive interaction between continuous PRS-SCZ and ES-SCZ

**Table 1.** Sample characteristics

		Patients <i>n</i> = 1699	Siblings <i>n</i> = 1753	Controls <i>n</i> = 1542	Total <i>N</i> = 4994
Age	Mean (s.d.)	31.49 (8.95)	31.73 (9.62)	33.45 (10.62)	32.18 (9.76)
Sex	% female	30.02	53.17	50.58	44.49
SIS-R total	Mean (s.d.)	–	0.39 (0.33)	0.23 (0.24)	0.32 (0.30) <sup>a</sup>
SIS-R positive	Mean (s.d.)	–	0.41 (0.42)	0.24 (0.31)	0.33 (0.38) <sup>a</sup>
SIS-R negative	Mean (s.d.)	–	0.38 (0.34)	0.23 (0.24)	0.31 (0.31) <sup>a</sup>
PRS-SCZ <sub>75</sub>	% > 75 <sup>th</sup>	45.44	33.09	23.74	34.40
ES-SCZ <sub>75</sub>	% > 75 <sup>th</sup>	58.79	36.30	21.88	37.64

ES-SCZ<sub>75</sub>, exposome score for schizophrenia (75% cut-point); *n*, number of individuals; PRS-SCZ<sub>75</sub>, polygenic risk score for schizophrenia (75% cut-point); s.d., standard deviation; SIS-R, the structured interview for schizotypy – revised.

<sup>a</sup>Within siblings and control groups (3295 individuals).

**Table 2.** Interaction of PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub> on case-control status

	PRS-SCZ <sub>75</sub> = 0	PRS-SCZ <sub>75</sub> = 1	RERI (95% CI)
	Odds ratio (95% CI)	Odds ratio (95% CI)	
ES-SCZ <sub>75</sub> = 0	1.0	2.79 (2.24–3.47) <i>P</i> < 0.001	6.79 (3.32–10.26) <i>P</i> < 0.001
ES-SCZ <sub>75</sub> = 1	4.86 (3.92–6.02) <i>P</i> < 0.001	13.44 (10.21–17.69) <i>P</i> < 0.001	

CI, confidence interval; ES-SCZ<sub>75</sub>, exposome score for schizophrenia (75% cut-point); PRS-SCZ<sub>75</sub>, polygenic risk score for schizophrenia (75% cut-point); RERI, relative excess risk due to interaction.

Adjusted for sex, age, and ten PCs.

in unimputed data (*N* = 1000 bootstrap replications) (Richardson and Kaufman, 2009).

In unaffected siblings and healthy comparison participants, the effects of continuous measures of PRS-SCZ, ES-SCZ, and their interaction on continuous measures of schizotypy dimensions (total, positive, and negative) as dependent variables were tested with multilevel linear regression models, where the coefficient of the product term (PRS-SCZ×ES-SCZ) reflects the departure from additivity (Knol *et al.*, 2007).

Previous analyses did not indicate a gene–environment correlation between the individual environmental exposures and PRS-SCZ<sub>75</sub> in the control sample (Guloksuz *et al.*, 2019). Furthermore, for the current analyses, we tested gene–environment correlation between the continuous (ES-SCZ and PRS-SCZ) and dichotomised (ES-SCZ<sub>75</sub> and PRS-SCZ<sub>75</sub>) exposome and genetic risk scores applying multilevel linear and logistic regression, respectively. Nagelkerke's R<sup>2</sup> was calculated based on logistic regression with case-control status as the dependent variable.

## Results

Sample demographic data, SIS-R scores, PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub> distributions are reported in Table 1. Missing data are reported in the Supplementary material (Table S1).

PRS-SCZ explained 15% of the variance in case-control status (OR = 1.30 [95% CI 1.25, 1.34], *p* < 0.001) and 20% after adjusting for age, sex, and country (OR = 1.30 [95% CI 1.26, 1.35], *p* < 0.001). ES-SCZ explained 28% of the variance in case-control status (OR = 2.52 [95% CI 2.29, 2.78], *p* < 0.001) and 33% after adjusting for age, sex, and country (OR = 2.40 [95% CI 2.17, 2.66], *p* < 0.001).

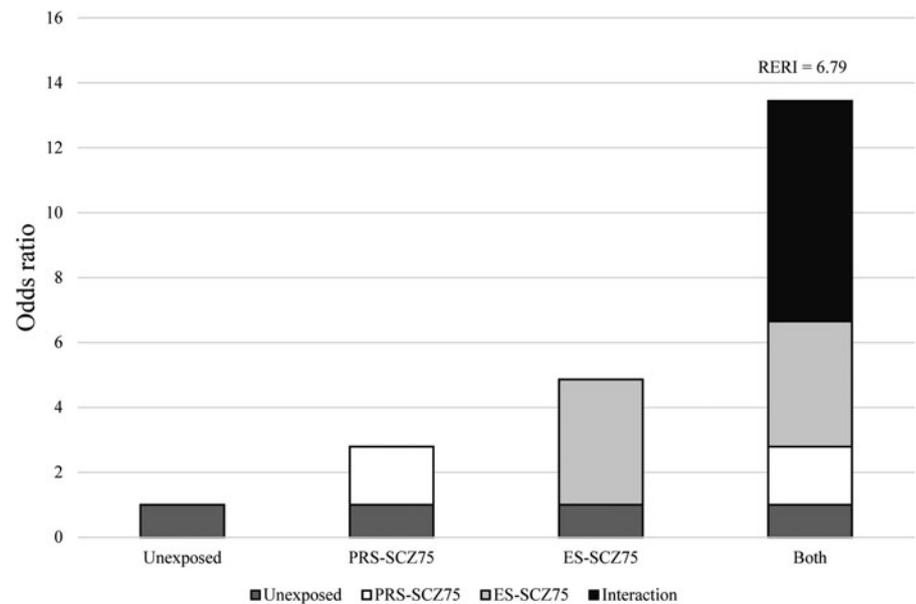
There was no evidence for gene–environment correlation, as PRS-SCZ<sub>75</sub> was not strongly or significantly associated with ES-SCZ<sub>75</sub> in the control group (OR = 1.08 [95% CI 0.78, 1.51], *p* = 0.635), neither after adjusting for age and sex (OR = 1.08 [95% CI 0.78, 1.51], *p* = 0.638) nor when using the continuous scores; PRS-SCZ and ES-SCZ (*B* = −0.008 [95% CI −0.028, 0.013], *p* = 0.478; adjusted *B* = −0.008 [95% CI −0.029, 0.012], *p* = 0.429).

### Main and joint effects of PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub> on case-control status

PRS-SCZ<sub>75</sub> was associated with case status (OR = 2.91 [95% CI 2.48, 3.40], *p* < 0.001; adjusted for age and sex: OR = 2.85 [95% CI 2.43, 3.35], *p* < 0.001); and ES-SCZ<sub>75</sub> was associated with case status (OR = 4.99 [95% CI 4.22, 5.90], *p* < 0.001, adjusted for age and sex: OR = 4.90 [95% CI 4.14, 5.81], *p* < 0.001). There was evidence for a positive additive interaction between PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub> (RERI = 7.29 [95% CI 3.73, 10.85], *p* < 0.001), also after adjusting for age and sex (Table 2 and Fig. 1). Sensitivity analyses using continuous PRS-SCZ and ES-SCZ confirmed G × E interaction (RERI = 1.77 [95% CI 1.00, 3.24], *p* = 0.003; adjusted RERI = 1.80 [95% CI 1.01, 3.32], *p* = 0.004). Results from the analyses using unimputed data corroborated these results and are reported in the Supplementary material (Table S2 and Figure S3).

### Main and joint effects of continuous PRS-SCZ and ES-SCZ on SIS-R dimensions

PRS-SCZ was significantly associated with the SIS-R dimensions in the unaffected sibling/healthy comparison participants sample



**Fig. 1.** Additive effect of the polygenic risk score for schizophrenia, 75% cut-point (PRS-SCZ<sub>75</sub>), and the exposome score for schizophrenia, 75% cut-point (ES-SCZ<sub>75</sub>) on case-control status, adjusted for age, sex, and ten PCs; RERI: relative excess risk due to interaction.

**Table 3.** Main and joint effects of PRS-SCZ and ES-SCZ on SIS-R scores

Psychopathology measures	Main effect PRS-SCZ <sup>a</sup>			Main effect ES-SCZ			Interaction <sup>a</sup>		
	<i>B</i>	95% CI	<i>p</i> -value	<i>B</i>	95% CI	<i>p</i> -value	<i>B</i>	95% CI	<i>p</i> -value
SIS-R total	0.011	0.007–0.015	<0.001	0.088	0.078–0.098	<0.001	0.006	0.003–0.009	<0.001
SIS-R positive	0.012	0.007–0.018	<0.001	0.103	0.091–0.116	<0.001	0.006	0.002–0.009	0.002
SIS-R negative	0.010	0.005–0.014	<0.001	0.074	0.064–0.085	<0.001	0.006	0.004–0.009	<0.001

*B*, regression coefficient from the multilevel model; CI, confidence interval; ES-SCZ, exposome score for schizophrenia; PRS-SCZ, polygenic risk score for schizophrenia; SIS-R, the structured interview for schizotypy – revised.

All analyses were adjusted for age and sex.

<sup>a</sup>Additionally adjusted for ten PCs.

(total:  $B = 0.011$  [95% CI 0.006, 0.015],  $p < 0.001$ ; positive:  $B = 0.012$  [95% CI 0.007, 0.018],  $p < 0.001$ ; negative:  $B = 0.010$  [95% CI 0.005, 0.014],  $p < 0.001$ ) also after adjusting for age and sex (Table 3). ES-SCZ was also significantly associated with the SIS-R dimensions (total:  $B = 0.088$  [95% CI 0.078, 0.098],  $p < 0.001$ ; positive:  $B = 0.103$  [95% CI 0.090, 0.116],  $p < 0.001$ ; negative:  $B = 0.074$  [95% CI 0.064, 0.085],  $p < 0.001$ ), also after adjusting for age and sex (Table 3). There was evidence for a significant interaction between ES-SCZ and PRS-SCZ on the SIS-R dimensions (total:  $B = 0.006$  [95% CI 0.003, 0.009],  $p < 0.001$ ; positive:  $B = 0.005$  [95% CI 0.002, 0.009],  $p = 0.002$ ; and negative:  $B = 0.006$  [95% CI 0.003, 0.009],  $p < 0.001$ ), also after adjusting for age and sex (Table 3). Results from the analyses in unimputed data confirmed the results in imputed data and are reported in the Supplementary material (Table S4).

## Discussion

To the best of our knowledge, this is the first study testing the role of gene–environment interaction using aggregate scores of environmental and genetic liability across the spectrum of psychosis expression. In the case-control design, we found evidence for additive interaction between PRS-SCZ and ES-SCZ increasing the odds for schizophrenia. Similarly, evidence emerged for interaction between PRS-SCZ and ES-SCZ on schizotypal traits when

investigating G×E interaction in the group of unaffected siblings and healthy comparison participants.

By using aggregate scores for genetic and environmental liability for schizophrenia, we provided further support for the role of gene–environment interaction in schizophrenia spectrum disorder (Bernardo *et al.*, 2017; Guloksuz *et al.*, 2019) and replicated recent findings of suggestive, but not nominally statistically significant, additive interaction between PRS-SCZ and environmental risk score for schizophrenia in a first episode psychosis cohort (Mas *et al.*, 2020). When PRS-SCZ<sub>75</sub> and ES-SCZ<sub>75</sub> were analysed as binary modes of risk factors, the relative excess risk due to the interaction was 6.79 and the corresponding 95% CI was above 2, suggesting a ‘mechanistic’ interaction, which means that the risk of developing schizophrenia for some individuals exists only when both genetic and environmental risks are present together but not when either genetic or environmental risk is present alone. The results further suggest that the PRS-SCZ and ES-SCZ explain 15 and 28% of the variance in case-control samples, respectively.

In a previous study, we demonstrated that the extent of sub-threshold phenotypic expression of schizophrenia polygenic risk is contingent on having a sibling with a psychotic disorder, suggesting a gene–environment interaction underlying schizotypy expression (van Os *et al.*, 2020). In the light of this new evidence, we tested for the first time the putative role of gene–environment

interaction in schizotypy traits. In line with our previous inference, we have now demonstrated that molecular genetic liability for schizophrenia moderates the effect of environmental liability for schizophrenia on phenotypic expression of overall, positive, and negative schizotypy traits in unaffected participants. Although much research has investigated the role of familial sensitivity to individual environmental exposures (e.g. cannabis use and childhood adversities) underlying subclinical psychosis expression (Modinos *et al.*, 2013; EUGEI investigators, 2014), only a few studies have utilised PRS to investigate the role of G×E in intermediate psychotic phenotypes (Ronald and Pain, 2018). A recent study from the 1966 Northern Finland Birth Cohort showed that high birth weight, a risk factor for familial schizophrenia in this cohort, increased the association between PRS-SCZ and social anhedonia, suggesting a gene–environment interaction (Liuhanen *et al.*, 2018). Similarly, a study conducted in a general population twin cohort demonstrated that while PRS-SCZ was not independently associated with affective dysregulation and psychosis proneness, PRS-SCZ increased sensitivity to the effect of childhood adversities on affective dysregulation and psychosis proneness (Pries *et al.*, 2020a). Although not a direct test of gene–environment interaction, a study of healthy young males assessed during their compulsory military service showed that there was a negative association between PRS-SCZ and positive schizotypy at military induction (stressful condition) but not at follow-up, providing further support for the key role of environment in the phenotypic expression of schizotypy traits (Hatzimanolis *et al.*, 2018). Taken together, although warranting further replication in independent cohorts, these findings imply that the phenotypic expression of schizotypal traits involves underlying genomic liability for schizophrenia that operates, at least in part, through sensitising individuals to the exposome.

The major strengths related to the study population were threefold: sufficient sample size to detect gene–environment interactions, access to comprehensive genotype, phenotype, and exposure data collected through validated interviews conducted by trained psychiatrists, psychologists or research assistants, and the geographical and cultural diversity of the sample that may increase the variation of environmental exposures and thereby provide increased power and replicability to detect interaction effects across populations (Ritz *et al.*, 2017).

The ES-SCZ was constructed using predictive modelling that mutually adjusted for the interdependency of exposures to prevent overestimation of the weights per exposure. ES-SCZ was fully compatible with this study population and clearly outperformed other aggregate scores that were based on meta-analytical estimates or simple summation of exposures as shown previously. Notwithstanding, ES-SCZ was limited by the degree to which exposures were available in the data set, and therefore did not include other exposures that might be of importance, such as obstetric and pregnancy complications (Garcia-Rizo and Bitanirwe, 2020). Furthermore, childhood adversities and cannabis use were retrospectively assessed and may be affected by recall bias (Baldwin *et al.*, 2019). Although population stratification was controlled using PCs and no gene–environment correlation was detected, unmeasured environmental confounding might still be present. The cross-sectional design did not allow for investigating the dynamic nature of gene–environment interaction over time.

In conclusion, we have shown that the interplay between exposure load and genetic liability for schizophrenia contributes to the phenotypic expression of psychosis across the extended

phenotype. Our findings provide further empirical support to the notion of aetiological continuity of psychosis spectrum and pave the way for future longitudinal studies of schizotypy traits in the general population. As some individuals may only develop schizophrenia spectrum disorder if both the genetic and environmental vulnerabilities are present, the current findings highlight the importance of the combined effect of genomic and exposomic liability for clinical practice. Furthermore, the results suggest that health care strategies may benefit from focusing on modifiable environmental factors.

**Supplementary material.** The supplementary material for this article can be found at <https://doi.org/10.1017/S2045796020000943>.

**Data.** The data that support the findings of this study are available from the corresponding author upon reasonable request under the condition of the approval of the EUGEI and GROUP steering committees.

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**Author contributions.** SG and L-KP conceived the idea of this study and developed the plan for analysis. SG, L-KP, and GAdF performed the statistical analysis and wrote the first draft. SG, JvO, and BPF provided supervision and expert knowledge on gene–environment interaction. ALR and MoD provided expert knowledge on psychiatric genetics and processed genotyping data. SG and L-KP contributed to data cleaning and database management for initial use and later reuse. All authors contributed to the collection of data and interpretation of the results, revised the manuscript and approved the final version.

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**Ethical standards.** The projects were approved by the Medical Ethics Committees of all participating sites and conducted in accordance with the Declaration of Helsinki. All respondents provided written informed consent and, in the case of minors, such consent was also obtained from parents or legal guardians.

## References

- Allardyce J, Leonenko G, Hamshere M, Pardinas AF, Forty L, Knott S, Gordon-Smith K, Porteous DJ, Haywood C, Di Florio A, Jones L, McIntosh AM, Owen MJ, Holmans P, Walters JTR, Craddock N, Jones I, O'Donovan MC and Escott-Price V (2018) Association between schizophrenia-related polygenic liability and the occurrence and level of mood-incongruent psychotic symptoms in bipolar disorder. *JAMA Psychiatry* 75, 28–35.

- Andreasen NC, Flaum M and Arndt S (1992) The Comprehensive assessment of symptoms and history (CASH). An instrument for assessing diagnosis and psychopathology. *Archives of General Psychiatry* **49**, 615–623.
- Arnau-Soler A, Adams MJ, Clarke TK, MacIntyre DJ, Milburn K, Navrady L, Generation Scotland Major Depressive Disorder Working Group of the Psychiatric Genomics Consortium, Hayward C, McIntosh A and Thomson PA (2019a). A validation of the diathesis-stress model for depression in Generation Scotland. *Translational Psychiatry* **9**, 25.
- Arnau-Soler A, Macdonald-Dunlop E, Adams MJ, Clarke TK, MacIntyre DJ, Milburn K, Navrady L, Generation Scotland Major Depressive Disorder Working Group of the Psychiatric Genomics Consortium, Hayward C, McIntosh AM and Thomson PA (2019b). Genome-wide by environment interaction studies of depressive symptoms and psychosocial stress in UK Biobank and Generation Scotland. *Translational Psychiatry* **9**, 14.
- Baldwin JR, Reuben A, Newbury JB and Danese A (2019) Agreement between prospective and retrospective measures of childhood maltreatment: a systematic review and meta-analysis. *JAMA Psychiatry* **76**, 584–593.
- Barkus EJ, Stirling J, Hopkins RS and Lewis S (2006) Cannabis-induced psychosis-like experiences are associated with high schizotypy. *Psychopathology* **39**, 175–178.
- Bernardo M, Bioque M, Cabrera B, Lobo A, Gonzalez-Pinto A, Pina L, Corripio I, Sanjuan J, Mane A, Castro-Fornieles J, Vieta E, Arango C, Mezquida G, Gasso P, Parellada M, Saiz-Ruiz J, Cuesta MJ, Mas S and PEPs GROUP (2017) Modelling gene-environment interaction in first episodes of psychosis. *Schizophrenia Bulletin* **189**, 181–189.
- Bernstein DP, Stein JA, Newcomb MD, Walker E, Pogge D, Ahluvalia T, Stokes J, Handelsman L, Medrano M, Desmond D and Zule W (2003) Development and validation of a brief screening version of the childhood trauma questionnaire. *Child Abuse & Neglect* **27**, 169–190.
- Colodro-Conde L, Couvy-Duchesne B, Zhu G, Coventry WL, Byrne EM, Gordon S, Wright MJ, Montgomery GW, Madden PA and Ripke S (2018) A direct test of the diathesis-stress model for depression. *Molecular Psychiatry* **23**, 1590–1596.
- Das S, Forer L, Schonherr S, Sidore C, Locke AE, Kwong A, Vrieze SI, Chew EY, Levy S, McGue M, Schlessinger D, Stambolian D, Loh PR, Iacono WG, Swaroop A, Scott LJ, Cucca F, Kronenberg F, Boehnke M, Abecasis GR and Fuchsberger C (2016) Next-generation genotype imputation service and methods. *Nature Genetics* **48**, 1284–1287.
- Davies G, Welham J, Chant D, Torrey EF and McGrath J (2003) A systematic review and meta-analysis of northern hemisphere season of birth studies in schizophrenia. *Schizophrenia Bulletin* **29**, 587–593.
- EUGEI investigators (2014) Identifying gene-environment interactions in schizophrenia: contemporary challenges for integrated, large-scale investigations. *Schizophrenia Bulletin* **40**, 729–736.
- Garcia-Rizo C and Bitanhihrwe BKY (2020) Implications of early life stress on fetal metabolic programming of schizophrenia: a focus on epiphenomena underlying morbidity and early mortality. *Progress in Neuro-Psychopharmacology & Biological Psychiatry* **101**, 109910.
- Geoffroy PA, Etain B and Houenou J (2013) Gene-environment interactions in schizophrenia and bipolar disorder: evidence from neuroimaging. *Frontiers in Psychiatry* **4**, 136.
- Guloksuz S and van Os J (2018) The slow death of the concept of schizophrenia and the painful birth of the psychosis spectrum. *Psychological Medicine* **48**, 229–244.
- Guloksuz S, Rutten BPF, Pries LK, Ten Have M, de Graaf R, van Dorselaer S, Klingenberg B, van Os J, Ioannidis JPA and European Network of National Schizophrenia Networks Studying Gene-Environment Interactions Work Package 6 (EU-GEI WP6) Group (2018) The complexities of evaluating the exposome in psychiatry: a data-driven illustration of challenges and some propositions for amendments. *Schizophrenia Bulletin* **44**, 1175–1179.
- Guloksuz S, Pries LK, Delespaul P, Kenis G, Luyckx JJ, Lin BD, Richards AL, Akdede B, Binbay T, Altinyazar V, Yalincetin B, Gumus-Akay G, Cihan B, Soygur H, Ulas H, Cankurtaran E, Kaymak SU, Mihaljevic MM, Petrovic SA, Mirjanic T, Bernardon E, Cabrera B, Bobes J, Saiz PA, Garcia-Portilla MP, Sanjuan J, Aguilar EJ, Santos JL, Jimenez-Lopez E, Arrojo M, Carracedo A, Lopez G, Gonzalez-Penas J, Parellada M, Maric NP, Atbasog Lu C, Ucock A, Alptekin K, Saka MC, Genetic Risk Outcome of Psychosis investigators, Arango C, O'Donovan M, Rutten BPF and van Os J (2019) Examining the independent and joint effects of molecular genetic liability and environmental exposures in schizophrenia: results from the EUGEI study. *World Psychiatry* **18**, 173–182.
- Guloksuz S, Pries LK, Ten Have M, de Graaf R, van Dorselaer S, Klingenberg B, Bak M, Lin BD, van Eijk KR, Delespaul P, van Amelsvoort T, Luyckx JJ, Rutten BPF and van Os J (2020) Association of preceding psychosis risk states and non-psychotic mental disorders with incidence of clinical psychosis in the general population: a prospective study in the NEMESIS-2 cohort. *World Psychiatry* **19**, 199–205.
- Hatzimanolis A, Avramopoulos D, Arking DE, Moes A, Bhatnagar P, Lencz T, Malhotra AK, Giakoumaki SG, Roussos P, Smyrnis N, Bitsios P and Stefanis NC (2018) Stress-dependent association between polygenic risk for schizophrenia and schizotypal traits in young army recruits. *Schizophrenia Bulletin* **44**, 338–347.
- Hernandez A, Gallardo-Pujol D, Pereda N, Arntz A, Bernstein DP, Gaviria AM, Labad A, Valero J and Gutierrez-Zotes JA (2013) Initial validation of the Spanish childhood trauma questionnaire-short form: factor structure, reliability and association with parenting. *Journal of Interpersonal Violence* **28**, 1498–1518.
- Hilker R, Helenius D, Fagerlund B, Skytthe A, Christensen K, Werge TM, Nordentoft M and Glenthøj B (2018) Heritability of schizophrenia and schizophrenia spectrum based on the nationwide Danish twin register. *Biological Psychiatry* **83**, 492–498.
- Hunter SC, Mora-Merchan J and Ortega R (2004) The long-term effects of coping strategy use in victims of bullying. *The Spanish Journal of Psychology* **7**, 3–12.
- Kendler KS and Gardner CO (2010) Interpretation of interactions: guide for the perplexed. *British Journal of Psychiatry* **197**, 170–171.
- Kendler KS, Lieberman JA and Walsh D (1989) The Structured Interview for Schizotypy (SIS): a preliminary report. *Schizophrenia Bulletin* **15**, 559–571.
- Knol MJ and VanderWeele TJ (2012) Recommendations for presenting analyses of effect modification and interaction. *International Journal of Epidemiology* **41**, 514–520.
- Knol MJ, van der Tweel I, Grobbee DE, Numans ME and Geerlings MI (2007) Estimating interaction on an additive scale between continuous determinants in a logistic regression model. *International Journal of Epidemiology* **36**, 1111–1118.
- Korver N, Quee PJ, Boos HB, Simons CJ, de Haan L and GROUP investigators (2012) Genetic risk and outcome of psychosis (GROUP), a multi-site longitudinal cohort study focused on gene-environment interaction: objectives, sample characteristics, recruitment and assessment methods. *International Journal of Methods in Psychiatric Research* **21**, 205–221.
- Linszen MM, Brouwer RM, Heringa SM and Sommer IE (2016) Increased risk of psychosis in patients with hearing impairment: review and meta-analyses. *Neuroscience & Biobehavioral Reviews* **62**, 1–20.
- Liuhanan J, Suvisaari J, Kajantie E, Miettunen J, Sarin AP, Jarvelin MR, Lonnqvist J, Veijola J and Paunio T (2018) Interaction between compound genetic risk for schizophrenia and high birth weight contributes to social anhedonia and schizophrenia in women. *Psychiatry Research* **259**, 148–153.
- Loh P-R, Danecek P, Palamara PF, Fuchsberger C, Reshef YA, Finucane HK, Schoenherr S, Forer L, McCarthy S, Abecasis GR, Durbin R and Price LA (2016) Reference-based phasing using the haplotype reference consortium panel. *Nature Genetics* **48**, 1443–1448.
- Mas S, Boloc D, Rodriguez N, Mezquida G, Amoretti S, Cuesta MJ, Gonzalez-Penas J, Garcia-Alcon A, Lobo A, Gonzalez-Pinto A, Corripio I, Vieta E, Castro-Fornieles J, Mane A, Saiz-Ruiz J, Gasso P, Bioque M, Bernardo M and PEPs Group (2020) Examining gene-environment interactions using aggregate scores in a first-episode psychosis cohort. *Schizophrenia Bulletin* **46**, 1019–1025.
- McGuffin P, Farmer A and Harvey I (1991) A polydiagnostic application of operational criteria in studies of psychotic illness. Development and reliability of the OPCRIT system. *Archives of General Psychiatry* **48**, 764–770.
- Modinos G, Iyegbe C, Prata D, Rivera M, Kempton MJ, Valmaggia LR, Sham PC, van Os J and McGuire P (2013) Molecular genetic gene-environment studies using candidate genes in schizophrenia: a systematic review. *Schizophrenia Bulletin* **150**, 356–365.

- Pardinas AF, Holmans P, Pocklington AJ, Escott-Price V, Ripke S, Carrera N, Legge SE, Bishop S, Cameron D, Hamshere ML, Han J, Hubbard L, Lynham A, Mantripragada K, Rees E, MacCabe JH, McCarroll SA, Baune BT, Breen G, Byrne EM, Dannlowski U, Eley TC, Hayward C, Martin NG, McIntosh AM, Plomin R, Porteous DJ, Wray NR, Caballero A, Geschwind DH, Huckins LM, Ruderfer DM, Santiago E, Sklar P, Stahl EA, Won H, Agerbo E, Als TD, Andreassen OA, Baekvad-Hansen M, Mortensen PB, Pedersen CB, Borglum AD, Bybjerg-Grauholm J, Djurovic S, Durmishi N, Pedersen MG, Golimbet V, Grove J, Hougaard DM, Mattheisen M, Molden E, Mors O, Nordentoft M, Pejovic-Milovancevic M, Sigurdsson E, Silagadze T, Hansen CS, Stefansson K, Stefansson H, Steinberg S, Tosato S, Werge T, Gerad Consortium, Rester Consortium, Collier DA, Rujescu D, Kirov G, Owen MJ, O'Donovan MC and Walters JTR (2018) Common schizophrenia alleles are enriched in mutation-intolerant genes and in regions under strong background selection. *Nature Genetics* 50, 381–389.
- Price AL, Weale ME, Patterson N, Myers SR, Need AC, Shianna KV, Ge D, Rotter JJ, Torres E, Taylor KD, Goldstein DB and Reich D (2008) Long-range LD can confound genome scans in admixed populations. *American Journal of Human Genetics* 83, 132–135; author reply 135–139.
- Pries LK, Guloksuz S, Ten Have M, de Graaf R, van Dorsselaer S, Gunther N, Rauschenberg C, Reininghaus U, Radhakrishnan R, Bak M, Rutten BPF and van Os J (2018) Evidence that environmental and familial risks for psychosis additively impact a multidimensional subthreshold psychosis syndrome. *Schizophrenia Bulletin* 44, 710–719.
- Pries LK, Lage-Castellanos A, Delepaul P, Kenis G, Luyck JJ, Lin BD, Richards AL, Akdede B, Binbay T, Altinyazar V, Yalincetin B, Gumus-Akay G, Cihan B, Soygur H, Ulas H, Cankurtaran ES, Kaymak SU, Mihaljevic MM, Petrovic SA, Mirjanic T, Bernardo M, Cabrera B, Bobes J, Saiz PA, Garcia-Portilla MP, Sanjuan J, Aguilar EJ, Santos JL, Jimenez-Lopez E, Arrojo M, Carracedo A, Lopez G, Gonzalez-Penas J, Parellada M, Maric NP, Atbasoglu C, Ucok A, Alptekin K, Saka MC, Genetic Risk Outcome of Psychosis investigators, Arango C, O'Donovan M, Rutten BPF, van Os J and Guloksuz S (2019) Estimating exposome score for schizophrenia using predictive modeling approach in two independent samples: the results from the EUGEI study. *Schizophrenia Bulletin* 45, 960–965.
- Pries LK, Klingenberg B, Menne-Lothmann C, Decoster J, van Winkel R, Collip D, Delepaul P, De Hert M, Derom C, Thiery E, Jacobs N, Wichers M, Cinar O, Lin BD, Luyck JJ, Rutten BPF, van Os J and Guloksuz S (2020a). Polygenic liability for schizophrenia and childhood adversity influences daily-life emotion dysregulation and psychosis proneness. *Acta Psychiatrica Scandinavica* 141, 465–475.
- Pries LK, van Os J, Ten Have M, de Graaf R, van Dorsselaer S, Bak M, Lin BD, van Eijk KR, Kenis G, Richards A, O'Donovan MC, Luyck JJ, Rutten BPF and Guloksuz S (2020b). Association of recent stressful life events with mental and physical health in the context of genomic and exposomic liability for schizophrenia. *JAMA Psychiatry*. doi: 10.1001/jamapsychiatry.2020.2304.
- Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MA, Bender D, Maller J, Sklar P, de Bakker PI, Daly MJ and Sham PC (2007) PLINK: a tool set for whole-genome association and population-based linkage analyses. *American Journal of Human Genetics* 81, 559–575.
- Radhakrishnan R, Guloksuz S, Ten Have M, de Graaf R, van Dorsselaer S, Gunther N, Rauschenberg C, Reininghaus U, Pries LK, Bak M and van Os J (2019) Interaction between environmental and familial affective risk impacts psychosis admixture in states of affective dysregulation. *Psychological Medicine* 49, 1879–1889.
- Radua J, Ramella-Cravaro V, Ioannidis JPA, Reichenberg A, Phiphophatsanee N, Amir T, Yenn Thoo H, Oliver D, Davies C, Morgan C, McGuire P, Murray RM and Fusar-Poli P (2018) What causes psychosis? An umbrella review of risk and protective factors. *World Psychiatry* 17, 49–66.
- Richardson DB and Kaufman JS (2009) Estimation of the relative excess risk due to interaction and associated confidence bounds. *American Journal of Epidemiology* 169, 756–760.
- Ritz BR, Chatterjee N, Garcia-Closas M, Gauderman WJ, Pierce BL, Kraft P, Tanner CM, Mechanic LE and McAllister K (2017) Lessons learned from past gene-environment interaction successes. *American Journal of Epidemiology* 186, 778–786.
- Robins LN, Wing J, Wittchen HU, Helzer JE, Babor TF, Burke J, Farmer A, Jablenski A, Pickens R, Regier DA, Sartorius N and Towle LH (1988) The composite international diagnostic interview: an epidemiologic instrument suitable for use in conjunction with different diagnostic systems and in different cultures. *Archives of General Psychiatry* 45, 1069–1077.
- Ronald A and Pain O (2018) A systematic review of genome-wide research on psychotic experiences and negative symptom traits: new revelations and implications for psychiatry. *Human Molecular Genetics* 27, R136–R152.
- Rothman KJ (1976) The estimation of synergy or antagonism. *American Journal of Epidemiology* 103, 506–511.
- Rothman KJ, Greenland S and Walker AM (1980) Concepts of interaction. *American Journal of Epidemiology* 112, 467–470.
- Royston P and White IR (2011) Multiple imputation by chained equations (MICE): implementation in Stata. *Journal of Statistical Software* 45, 1–20.
- Rubin DB (2004) *Multiple Imputation for Nonresponse in Surveys*. New York: John Wiley & Sons.
- Sar V, Akyuz G, Kundakci T, Kiziltan E and Dogan O (2004) Childhood trauma, dissociation, and psychiatric comorbidity in patients with conversion disorder. *The American Journal of Psychiatry* 161, 2271–2276.
- Schäfer M, Korn S, Smith PK, Hunter SC, Mora-Merchán JA, Singer MM and Meulen K (2004) Lonely in the crowd: recollections of bullying. *British Journal of Developmental Psychology* 22, 379–394.
- Schizophrenia Working Group of the Psychiatric Genomics Consortium (2014) Biological insights from 108 schizophrenia-associated genetic loci. *Nature* 511, 421–427.
- Sorensen HJ, Debost JC, Agerbo E, Benros ME, McGrath JJ, Mortensen PB, Ranning A, Hjorthoj C, Mors O, Nordentoft M and Petersen L (2018) Polygenic risk scores, school achievement, and risk for schizophrenia: a Danish population-based study. *Biological Psychiatry* 84, 684–691.
- StataCorp (2017) *Stata Statistical Software: Release 15*. College Station, TX: StataCorp LLC.
- Stilo SA and Murray RM (2019) Non-genetic factors in schizophrenia. *Current Psychiatry Reports* 21, 100.
- Thombs BD, Bernstein DP, Lobbestael J and Arntz A (2009) A validation study of the dutch childhood trauma questionnaire-short form: factor structure, reliability, and known-groups validity. *Child Abuse & Neglect* 33, 518–523.
- VanderWeele TJ and Knol MJ (2014) A tutorial on interaction. *Epidemiologic Methods* 3, 33–72.
- van Os J, Linscott RJ, Myin-Germeys I, Delepaul P and Krabbendam L (2009) A systematic review and meta-analysis of the psychosis continuum: evidence for a psychosis proneness-persistence-impairment model of psychotic disorder. *Psychological Medicine* 39, 179–195.
- van Os J, Pries LK, Delepaul P, Kenis G, Luyck JJ, Lin BD, Richards AL, Akdede B, Binbay T, Altinyazar V, Yalincetin B, Gumus-Akay G, Cihan B, Soygur H, Ulas H, Cankurtaran ES, Kaymak SU, Mihaljevic MM, Petrovic SA, Mirjanic T, Bernardo M, Cabrera B, Bobes J, Saiz PA, Garcia-Portilla MP, Sanjuan J, Aguilar EJ, Santos JL, Jimenez-Lopez E, Arrojo M, Carracedo A, Lopez G, Gonzalez-Penas J, Parellada M, Maric NP, Atbasoglu C, Ucok A, Alptekin K, Saka MC, Genetic Risk Outcome Investigators, Arango C, O'Donovan M, Rutten BPF and Guloksuz S (2020) Replicated evidence that endophenotypic expression of schizophrenia polygenic risk is greater in healthy siblings of patients compared to controls, suggesting gene-environment interaction. The EUGEI study. *Psychological Medicine* 50, 1884–1897.
- van Winkel R, van Beveren NJ and Simons C, Genetic Risk Outcome of Psychosis Investigators (2011) AKT1 Moderation of cannabis-induced cognitive alterations in psychotic disorder. *Neuropsychopharmacology* 36, 2529–2537.
- Vollema MG and Ormel J (2000) The reliability of the structured interview for schizotypy-revised. *Schizophrenia Bulletin* 26, 619–629.
- Wing JK, Babor T, Brugha T, Burke J, Cooper JE, Giel R, Jablenski A, Regier D and Sartorius N (1990) SCAN: schedules for clinical assessment in neuropsychiatry. *Archives of General Psychiatry* 47, 589–593.

## Appendix

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