Data-driven subphenotypic dissection of the clinical heterogeneity of schizophrenia spectrum disorders

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CHAPTER 3

Heterogeneity of clinical trajectories, disease liability and underlying factors in schizophrenia spectrum disorders: data-driven subphenotypic analyses

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ABSTRACT

Background: Clinical heterogeneity has been a longstanding issue in mental disorders, with its course and underlying factors yet to be elucidated given that the evidence from cross-sectional studies and longitudinal studies is inconclusive and insufficient.

Aim: As the first study to date, we combined sub-phenotyping, polygenic risk scoring and data-driven approaches to investigate the long-term clinical course of positive and negative symptoms and identify underlying genetic, metabolic, sociodemographic and clinical factors in patients with schizophrenia spectrum disorders, unaffected siblings and healthy controls.

Methods: We included 1,119 patients with a schizophrenia spectrum disorder, 1,059 unaffected siblings and 586 controls. Positive and negative symptoms and sociodemographic and clinical factors were assessed at baseline, and after three and six years. Genotype data and metabolic data were collected at baseline and the third year, respectively. Genetic predisposition to schizophrenia was measured by calculating polygenic risk scores (PRS_{SCZ}) using a standard approach. Censored normal group-based trajectory modeling was applied to identify symptom trajectories. A multinomial random-effects logistic regression model was used to investigate the association between symptom trajectories and underlying factors.

Results: Decreasing, increasing and relapsing trajectories of positive and negative symptoms were identified in patients. In unaffected siblings and healthy controls, four and three trajectories were found that could be characterized as stability, decreasing or increasing symptoms scores. Variability of trajectories differed substantially across samples, while similar levels of variability were observed across symptom domains. Poor premorbid adjustment, low quality of life and severe positive and negative symptoms at baseline significantly predicted the six-year trajectories of positive and negative symptoms in patients and siblings. A small yet significant correlation and longitudinal association between PRS_{SCZ} and trajectories of positive and negative symptoms in patients, siblings and controls were found, but this finding did not pertain after adjustment for confounders and multiple comparisons.

Conclusions: Three to four clinically and statistically meaningful trajectories of positive and negative symptoms were identified. Low premorbid functioning, poor

quality of life and severity of symptoms at baseline may better predict poor clinical outcomes of schizophrenia spectrum disorders than PRS_{SCZ}. Identified trajectories may contribute to research on personalized psychiatric care.

Keywords: Schizophrenia, psychosis, positive and negative symptoms, trajectory analysis, group-based trajectory modeling, polygenic risk score

INTRODUCTION

Schizophrenia spectrum disorders are a highly heterogeneous clinical syndrome manifested by positive (i.e. delusions, hallucinations, and disorganized thinking, speech and motor behavior) and negative (i.e. diminished emotional expression, avolition, alogia, anhedonia and asociality) symptoms.(1) Schizophrenia is a severe, chronic prototype of schizophrenia spectrum disorders. Positive and negative symptoms are often followed by poor clinical and functional outcomes.(2) Positive symptoms often respond to antipsychotic treatment, whereas negative symptoms are the most disabling, yet harder to treat even by behavioral interventions.(2,3) The pathophysiologic mechanism that underpins negative symptoms remain poorly understood compared to positive symptoms.(4)

Positive and negative symptoms may share part of their etiopathogenic mechanisms, they may invoke each other, or symptom-specific mechanisms may contribute to their presentations.(3,5,6) As a result, patient symptoms may follow a different clinical course from the time of onset. While positive symptoms generally follow a pattern of reduction and stabilization over time, negative symptoms present a persistent course over time.(7,8) On the other hand, another study showed that these symptoms follow parallel trajectories over time and are positively associated with each other. (3) Hence, different symptom domains may depend on each other through a unified underlying pathological disease process.

Clinically, schizophrenia is heterogeneous with wide variations in symptom presentation. Its pathogenic mechanisms remain largely unknown despite advances in technology that facilitate biological inquiry to disentangle the molecular complexity.(9) In an attempt to address the wide difference in disease presentation and course, the fifth edition of Diagnostic and Statistical Manual of Mental Disorders (DSM-5) endorses a dimensional approach to identify intermediate categories

based on individual experience on the severity of symptoms.(1) Following DSM-5 recommendations, some studies have focused on sub-phenotyping to examine heterogeneity and molecular mechanisms of symptoms.(10-12) Additionally, cross-sectional and longitudinal data-driven studies have explored heterogeneity within symptoms in a specific study population (e.g., patients, siblings, controls) and identified several sociodemographic, and clinical factors that distinguish subtypes of symptoms.(13) Additionally, there has been evidence that cardiometabolic profile in patients with schizophrenia is linked with more severe positive and negative symptoms and poorer functioning.(13-15) Findings from both sub-phenotyping and data-driven studies collectively highlight the difference in symptomatology patterns and similarity across positive and negative symptoms.

The phenotypic heterogeneity and molecular complexity can be attributed to genetic and environmental factors, such as patient characteristics, illness phase, or illness severity. We have previously found that symptom subtypes identified in cross-sectional and longitudinal studies were consistently linked with age, gender, ethnicity, age of illness onset, diagnosis, duration of untreated psychosis, duration of illness, premorbid adjustment, global functioning and quality of life, and cognitive performance.(13) Genetic etiology of symptoms in schizophrenia is supported by candidate gene and polygenic risk score (PRS) association studies.(10-12,16,17) Findings from the Psychiatric Genomics Consortium (PGC) genome-wide association study (GWAS) provide strong evidence for an association of genetic risk with positive and negative symptoms. Several genes like RFX8 on 2q11.2, WDR72 / UNC13C on 15q21.3, *NKAIN2* and *NRG1* are associated with positive and negative symptoms. (18,19) Moreover, the polygenic risk score for schizophrenia (PRS_{SCZ}), which is a measure of cumulative genetic risk, has been associated with positive and negative symptoms in patients with schizophrenia and the healthy population, though large inconsistencies have been observed.(10-12,20)

Despite the advantages of longitudinal studies and trajectory analyses, only a few studies with a relatively small sample size have examined the trajectories of positive and negative symptoms in patients and siblings while comparing them to those of healthy people.(13) Additionally, identified predictors were based only on pair-wise comparisons and univariable regression in some studies. So that, adjustment to confounders was mostly neglected and the reported effect estimates may have been confounded and biased. Besides, evidence on sub-threshold positive and

negative symptoms from an unaffected group of individuals (i.e., siblings and healthy controls or general population) is remaining to be exploited. Lastly, results on the longitudinal pattern of positive and negative symptoms, and the extent of symptom fluctuations within the identified trajectories remains inconclusive.(3) Of interest, the role of genetic factors to predict the long-term trajectories of positive and negative symptoms has not been investigated using data-driven approaches.(10,21) Mostly, small-sized cross-sectional studies examined the association between PRS_{SCZ} and positive and negative symptoms by using analyses that adjusted to no or limited confounding factors.(10-12,20,21)

In the present study, we combined sub-phenotyping, polygenic risk scoring and data-driven approaches to strengthen previous efforts for tackling clinical heterogeneity of schizophrenia spectrum disorders. Therefore, we aimed to investigate the heterogeneity and clinical outcomes of schizophrenia spectrum disorders over time and the role of genetic, sociodemographic and clinical risk factors. To evaluate the effect of disease liability in relation to clinical trajectories, we investigated the heterogeneity of trajectories of positive and negative symptoms across patients with a schizophrenia spectrum disorder representing full liability for the disorder, their non-affected siblings who share a larger proportion of disease liability, and healthy controls representing the least disease liability.

METHODS AND MATERIALS

Study population

Data of a six-year longitudinal multi-center national study was analyzed with 1,119 patients, 1,059 unaffected siblings and 586 healthy controls who were eligible at baseline, using the 7th official release of the Genetic Risk and OUtcome of Psychosis (GROUP) cohort data. Patients were included if diagnosed with a schizophrenia spectrum disorder, age range of 16 to 50 years, good command of the Dutch language, and willing and capable of giving written informed consent. Siblings and controls were included if they had no known lifetime psychotic disorder. The Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSMIV) criteria were used to diagnose schizophrenia spectrum disorder. Sociodemographic and clinical data were collected at baseline, and after three years and six years using standardized tools. Details regarding sample characteristics, recruitment and assessment procedures published elsewhere.(22)

Measurement of variables

Positive and negative symptoms

The Positive and Negative Syndrome Scale (PANSS) for schizophrenia was administered to measure the severity of a variety of positive and negative symptoms in patients.(23) The PANSS is a 30-item, clinician-rated interview in which severity of symptoms is rated in an incremental seven-point Likert severity scale (from 1 = none, 2 = minimal, to up to 7 = extreme). The positive symptom subscale score was calculated as the sum of positive symptom items, and the negative subscale score was the sum of negative symptoms. The PANSS was developed in the 1980s as a well-operationalized instrument that provides a balanced representation of negative and positive symptoms, and has been proven a reliable and valid tool and is the most widely used scale.(23) Positive and negative schizotypy in siblings and healthy controls were assessed with the Structured Interview for Schizotypyrevised (SISR).(24-26) The SISR is reliable to assess schizotypal symptoms and signs.(24) The SISR items are rated on an incremental four-point Likert severity scale (from 0 = absent, 1 = mild, 2 = moderate, and 3 = severe). The positive dimension score was calculated as the mean of positive schizotypy (referential thinking, delusional mood, magical ideation, illusions and suspiciousness), and the negative subscale score was calculated as the mean of negative schizotypy (social isolation, social anxiety, introversion, and restricted affect).

Sociodemographic and clinical factors

The sociodemographic variables were age, gender, marital status, ethnicity and educational status (as measured by years of education). Clinical variables were age of onset of illness, duration of illness, number of psychotic episodes, premorbid adjustment, estimated IQ, general cognition, psychotic symptoms or psychotic-like experiences, depression, social and global functioning, and quality of life. We also collected physical health data, such as body mass index, waist circumference, glycated hemoglobin, triglycerides, low-density lipoprotein, high-density lipoprotein, systolic and diastolic blood pressure and pulse rate through physical and laboratory examination. Cognitive function was assessed using a comprehensive neuropsychological test battery(27-29) that included the Word Learning Task (i.e. immediate recall and delayed recall), Continuous Performance TestHQ (CPTHQ) (CPT performance index and CPT variability), WAISIII Digit Symbol Substitution Test, WAIS

III Information, WAISIII Calculation, and WAISIII Block Design test. A composite score as a measure of general cognition was generated out of these different cognitive tests. Details on the assessment of these tasks, scoring system and calculation of composite score published elsewhere.(22,30) The Community Assessment of Psychic Experiences (CAPE) was used to assess psychotic experiences (CAPE-42; www.cape42.homestead.com). The CAPE is a 42-item reliable and valid self-report questionnaire to assess psychotic-like experiences in the community and in individuals at an increased risk for developing psychosis.(31,32) The Calgary Depression Scale for Schizophrenia (CDSS) was used to assess depression.(33) Premorbid adjustment, which is a measure of the degree of achievement of developmental goals at each of several periods of a subject's life before the onset of schizophrenia, was assessed using the premorbid adjustment scale.(34) The World Health Organization Quality of LifeBREF (WHOQOLBREF) questionnaire, which has high construct validity and reliability, was used to assess the quality of life(35); the Social Functioning Scale (SFS) and Global Assessment of Functioning (GAF) – disability were used to measure social (total score of SFS), occupational (sub-score of SFS) and global functioning.(36,37)

Genotyping, quality control (QC) and polygenic risk score (PRS)

Genotype data for 2,812 individuals were generated on a customized Illumina, IPMCN array with 570,038 single nucleotide polymorphisms (SNPs). This chip contains ~250k common SNPs, 250K Exome chip variants (rare, exomic, nonsynonymous, MAF < 1%), and ~50K psychiatric-related variants. Quality control (QC) procedures were performed using PLINK v1.9 (Supplementary methods).(38) In total, 2,505 individuals and 275,021 SNPs passed QC steps. SNPs were imputed on the Michigan server(39) using the HRC r1.1 2016 reference panel with European samples after phasing with Eagle v2.3. Finally, PRSs for 2,505 samples were calculated using schizophrenia-associated alleles and effect sizes reported in the GWAS summary statistics from the Psychiatric genetics consortium (PGC) 2014(40) by excluding Dutch subjects (Supplementary methods). PRSs were calculated using PLINK's score function for four schizophrenia GWAS p-value thresholds of 5×10^{-8}, 0.05, 0.1 and 0.5. Each PRS separately modeled to compare results and identify the most predictive and discriminant PRS for observed trajectories.

Statistical analysis

Symptom trajectory modeling

A trajectory is defined as a group of individuals who have a symptom profile that is homogenous within the group but significantly heterogeneous between groups.(41) Group-based trajectory modeling (GBTM) using PROC TRAJ (in SAS version 9 for Windows) was applied to identify distinct subgroups that have a unique symptom trajectory.(42) The censored normal drop-out model was used assuming upper and lower censoring of symptom scores and presence of attrition during follow-up.(42) The drop-out model, which includes a logistic model of drop-out probability per follow-up period, was used to examine the effect of attrition rates on group membership probabilities assuming outcome observations during follow-up intervals depend on preceding responses.(43) Moreover, missing data were handled by using a full information maximum likelihood model that provides parameter estimates by maximizing the likelihood function of the incomplete data as it produces unbiased results compared with other methods.(44) Initially, the optimum number of trajectory groups was identified using a quadratic polynomial order. The most parsimonious number of trajectory subgroups was determined using Bayesian information criterion (BIC), logged Bayes factor (i.e. 2*ΔBIC) and our clinical judgment. A 10-point difference was taken to favor the lower BIC.(45) Then, the polynomial order was adjusted until it became significant for all subgroups at the significance level of 0.05. To measure the model accuracy to distinguish individuals, we calculated the average group posterior probability (AvePP). A model with an AvePP greater than 0.7 for all identified groups is believed to be accurate.(46) A complete explanation of the theoretical concept, function, and application of GBTM is published elsewhere.(47-49) To achieve our objectives, trajectory analyses were performed per group (i.e., patient, sibling, healthy control) and symptom domain (i.e., positive and negative).

Statistical modeling

Differences in patient, sibling and control characteristics at baseline were explored using a linear mixed-effects model for continuous variables and Pearson's Chi-square tests for categorical variables. Since individuals are clustered in families, observations from the same family are likely correlated. Therefore, we declared family as a random effect to set up a common correlation among all observations from the same family. Family as a random effect was ignored if the G-matrix was not positive definite. The Maximum Likelihood (ML) method was used to estimate

the model parameters and fixed-effects (i.e., type III overall group comparison tests) model results were interpreted. Then, the bivariate association between PRS_{SCZ} and baseline positive and negative symptoms was examined using the Spearman's correlations test. Finally, univariable and multivariable multinomial random-effects logistic regression models(50-53) using PROC NLMIXED (in SAS version 9.4 for Windows) were fitted using symptomatic trajectories as an outcome variable, and PRS_{SCZ} and environmental factors as predictors. Initial parameters () used in the random-effects logistic regression model and explained variance (*Nagelkerke* R^2) were estimated using PROC LOGISTIC while ignoring the family clustering effect. PROC NLMIXED maximizes the likelihood function of the multinomial random-effects model by the Adaptive Gaussian quadrature method and Dual Quasi-Newton optimization technique, and therefore, provides stable parameter estimates. The multinomial random-effects logistic regression model was used because symptomatic trajectories had more than two nominal categories and study participants were clustered within a family. In the multivariate model, variables not available at baseline assessment (i.e., depressive symptoms, social and occupational functioning, and physical health parameters) were not included to avoid data separation points and to increase model convergence. All tests were adjusted for multiple comparisons; therefore, the significant threshold was set to be 0.025 (i.e., 0.05 divided by two comparisons made across all samples). Given the essential effect of genetic susceptibility for schizophrenia, the most predictive PRS_{SCZ} with high effect size (OR) was included in the multivariable model, even when its effect was not significant. All analyses used full information ML estimation, which uses all data, including partial cases, to arrive at unbiased parameter estimates.

RESULTS

Baseline characteristics of participants

Participants significantly differed in PRS_{SCZ} and environmental baseline characteristics at baseline (Table 1). The mean PRS_{SCZ} at P_T of 0.05, 0.1 and 0.5 of patients was significantly higher than that of siblings and controls ($p < 0.001$) and the mean PRS_{SCZ} of siblings significantly higher than that of controls ($p < 0.001$). Participants also differed significantly in their baseline sociodemographic and clinical characteristics, in which three-fourths of patients were male ($p < 0.001$) and patients were younger than controls ($p < 0.001$). In general, characteristics of unaffected siblings laid between patients and controls.

Trajectories of positive and negative symptoms

As Figure 1 shows, multiple clinically and statistically meaningful trajectories of positive and negative symptoms were identified across the study populations. In 1,136 eligible patients, we identified three trajectories of positive symptoms that were labeled as low (67.8%, quadratic shape), moderate (23.3%, flat shape) and high (8.9%, quadratic shape) and three trajectories of negative symptoms, labeled as low (69.5%, linear shape), high-decreased (16.4%, quadratic shape) and high-increased (14.1%, linear shape). In 1,045 eligible siblings, we found four trajectories of positive symptoms (i.e., low (21.7%, flat shape), moderate (55.4%, linear shape), high (17.8%, flat shape) and high-decreased (5.1%, quadratic shape)) and four trajectories of negative symptoms (i.e., low (35.2%, linear shape), moderate (51.7%, flat shape), high-decreased (5.1%, quadratic shape) and high-increased (8.0%, quadratic shape)). In 583 controls, we found three trajectories of positive symptoms (i.e., low (39.7%, flat shape), moderate (55.0%, flat shape) and high-decreased (5.3%, linear shape)) and three flat/stable trajectories of negative symptoms (i.e., low (21.8%), moderate (65.0%) and high (13.2%)). The model accuracy in all group modeling ranged from 70 to 91%. In general, larger variation in trajectories were observed in patients with stability and persistence of symptoms in about two-thirds of patients, reduction of symptoms in more than one-fifth of patients, and worsening symptoms in about one-tenth of patients. Trajectory model fit indices and parameter estimates are presented in Supplementary Table S1 – S8.

Trajectory groups were named by consensus considering the group mean score and the nature of change over time. In patients, the 'Low' group represents individuals with positive and negative symptoms sum score of < 15 on the PANSS, 'Moderate' group represents individuals with a sum score of approximately 15 to 20 and 'High' group represents individuals with a sum score of > 20. Based on the change throughout follow-up, we also labeled trajectories as 'High-Decreased' – start with high symptoms and ameliorate over time or 'High-Increased' – start with high symptoms and worsen over time. In siblings and controls, we followed a similar approach: 'Low' with mean score < 0.25 on the SIS-R, 'Moderate' with mean score approximately 0.25 to 0.5, and 'High' with a mean score of > 0.5. We also named trajectories in siblings and controls as 'High-Decreased' or 'High-Increased'.

Table 1: Participants background characteristics

Variable	Participants			Overall group difference	Pair-wise comparisons
	Controls (C)	Siblings (S)	Patients (P)		
Background characteristics					
Age, mean(SE)	30.60(0.39)	27.70(0.28)	27.40(0.27)	F=24.0, p<0.001	P<C; S<C
Gender, male n(%)	269(46.1)	477(45.6)	857(75.4)	X^2=240.97, p<0.001	
Ethnicity, Caucasian n(%)	523(92.1)	871(83.6)	871(79.1)	X^2=45.84, p<0.001	
Years of education, mean(SE)	15.6(0.16)	13.5(0.12)	12.4(0.12)	F= 64.5, p<0.001	P<S & C; S<C
Marital status n(%)					
Not married	319(57.6)	589(57.5)	960(87.8)	X^2=303.17, p<0.001	
Married/Living together	219(39.5)	412(40.2)	104(9.5)		
Other (divorced and widowed)	16(2.9)	24(2.3)	30(2.7)		
Estimated current IQ, mean(SE)	109.61(0.69)	102.58(0.52)	94.81(0.49)	F=190.93, p<0.001	P<S & C; S<C
Premorbid adjustment, mean(SE)	1.13(0.03)	1.11(0.02)	1.98(0.02)	F=448.7, p<0.001	P>S; P>C
Age onset illness, mean(SE)	-	-	23.1(0.23)	-	-
Duration of illness, mean(SE)	-	-	4.98(4.46)	-	-
Number of psychotic episodes, mean(SE)	-	-	1.72(1.17)	-	-
Use of antipsychotics[†] n(%)	-	-			
Not currently using	-	-	38(5.22)	-	-
Currently using	-	-	574(78.85)	-	-
Unknown if currently using	-	-	116(15.93)	-	-
Polygenic risk score of SCZ, mean(SE)					
PRS_{SCZ} (P_T=5x10^{-8})	-0.10(0.03)	-0.02(0.02)	0.02(0.02)	F= 7.01, p=0.001	P>C
PRS_{SCZ} (P_T=0.05)	-7.38(0.35)	-4.92(0.24)	-3.02(0.24)	F= 67.22, p<0.001	P>S & C; S>C
PRS_{SCZ} (P_T=0.1)	-8.29(0.43)	-5.59(0.30)	-3.29(0.29)	F= 64.36,p<0.001	P>S & C; S>C
PRS_{SCZ} (P_T=0.5)	-5.08(0.59)	-1.48(0.40)	1.24(0.39)	F= 56.50, p<0.001	P>S & C; S>C

Table 1: Continued.

Variable	Participants			Overall group difference	Pair-wise comparisons
	Controls (C)	Siblings (S)	Patients (P)		
Schizotypy, mean(SE)					
Positive	0.30(0.02)	0.38(0.01)	-	F= 59.45, p<0.001	S>C
Negative	0.23(0.01)	0.27(0.0.01)	-	F= 49.25, p<0.001	S>C
Psychotic symptoms, mean(SE)					
Positive	-	-	13.90(0.20)	-	-
Negative	-	-	14.99(0.21)	-	-
Disorganization	-	-	16.77(0.20)	-	-
Emotional distress	-	-	15.82(0.18)	-	-
Excitement	-	-	12.05(0.13)	-	-
Psychotic experiences, mean(SE)					
Positive symptoms frequency	0.19(0.01)	0.20(0.01)	0.67(0.01)	F= 571.97, p<0.001	P>S & C
Positive symptoms distress	0.43(0.03)	0.46(0.02)	1.25(0.02)	F=511.36, p<0.001	P>S & C
Negative symptoms frequency	0.49 (0.02)	0.54(0.01)	1.02(0.01)	F= 401.58, p<0.001	P>S & C
Negative symptoms distress	0.67(0.02)	0.68(0.02)	1.25(0.02)	F= 305.75, p<0.001	P>S & C
Depressive symptoms frequency	0.58(0.02)	0.62(0.02)	1.00(0.02)	F= 217.76, p <0.001	P>S & C
Depressive symptoms distress	0.88(0.03)	0.92(0.02)	1.44(0.02)	F= 212.19, p<0.001	P>S & C
General cognition	0.62(0.07)	0.13(0.05)	-1.18(0.05)	F= 312.77, p<0.001	P<S & C; S<C
Functioning and quality of life, mean(SE)					
Occupational functioning[†]	8.93(0.13)	8.92(0.10)	5.91(0.10)	F=292.98, p<0.0001	P<S & C
Social functioning[†]	124.04(0.36)	122.32(0.28)	112.51(0.27)	F=474.62, p<0.0001	P<S & C; S<C
Global functioning	-	-	54.50(0.52)	-	-
Quality of life	4.07(0.02)	3.97(0.02)	3.40(0.02)	F=492.16, p<0.0001	P<S & C; S<C

[†] = *Used from the second wave at three-year follow-up; SE=Standard Error*

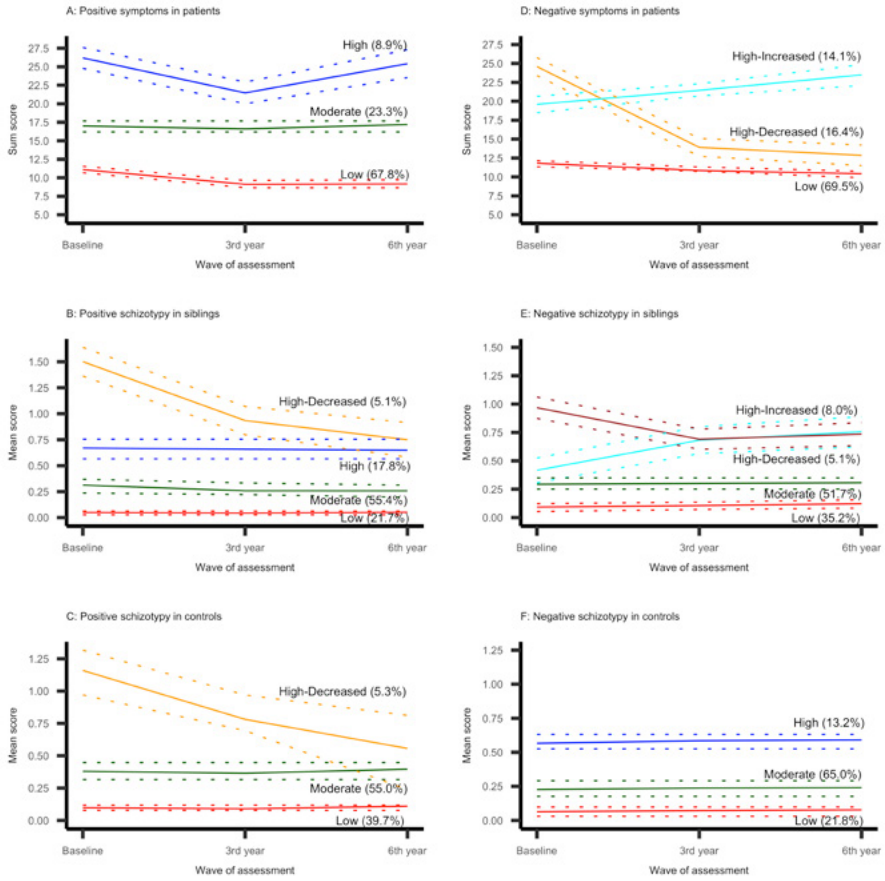

Figure 1: Trajectories of positive and negative symptoms in patients, siblings and controls.

Correlation between PRS_{SCZ}, positive and negative symptoms

Baseline negative symptoms in patients were significantly correlated with PRS_{SCZ} at P_T of 0.05 (Spearman's correlation (r_s) = 0.11, p = 0.007), 0.1 (r_s = 0.10, p = 0.01) and 0.5 (r_s = 0.10, p = 0.01). In controls, PRS_{SCZ} only at P_T of 0.5 (r_s = 0.10, p = 0.048) was significantly correlated with baseline negative symptoms. Among siblings, baseline positive symptoms were significantly correlated with PRS_{SCZ} at P_T of 0.05 (r_s = 0.10, p = 0.006), 0.1 (r_s = 0.11, p = 0.004) and 0.5 (r_s = 0.10, p = 0.008). Additionally, positive symptoms were significantly correlated with negative symptoms in patients (r_s = 0.34, p < 0.001), siblings (r_s = 0.47, p < 0.001) and healthy controls (r_s = 0.54, p < 0.001).

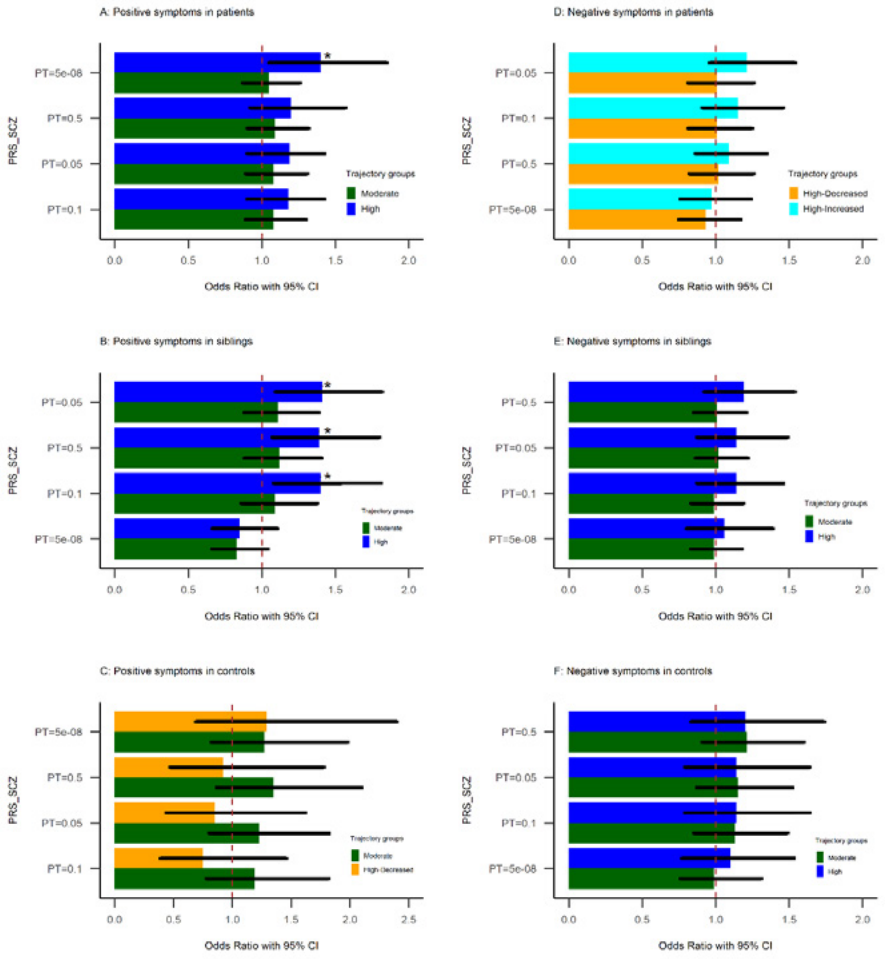

Figure 2: Predictive effect of PRS_{SCZ} on positive and negative symptoms trajectories in patients, siblings and controls (*p < 0.05).

Predictors of positive and negative symptoms trajectories

The results from the univariable regression analysis showed a significant association between PRS_{SCZ} at P_T threshold 5e-08 and positive symptoms trajectory in patients (High vs Low: OR = 1.40; 95% CI = 1.05 – 1.85; p = 0.02) (Figure 2). Environmental factors associated with positive symptom trajectories in patients were female gender, being married, ethnic minority, long duration of illness, frequent psychotic episodes, low estimated current IQ, poor premorbid adjustment, depressive symptoms, severe

baseline psychotic symptoms, low general cognition, poor quality of life, poor social, occupational and global functioning, high glycated hemoglobin level, high pulse rate, disturbed lipid profile and having metabolic syndrome (Table 2). The multivariable analyses showed that the explained variance of trajectories attributed to these factors, after excluding depressive symptoms, social functioning, occupational functioning and physical health parameters, was approximately 62.2%. Only a small fraction of the variance (0.04%) was explained by PRS_{SCZ}. Following adjustment for covariables and multiple comparisons, only baseline positive symptoms (High vs Low: OR = 2.91; 95% CI = 1.75 - 4.84; p <0.0001) were significantly associated with symptom trajectories (Supplementary Table S9). There was also evidence of an association between PRS_{SCZ} at P_T threshold 0.05 and negative symptom trajectories in patients though it was not statistically significant (High-Increased vs Low: OR = 1.21; 95% CI = 0.96 – 1.54; p = 0.11) (Figure 2). Female gender, more years of education, poor premorbid adjustment, low estimated current IQ, depressive symptoms, low general cognition, severe baseline psychotic symptoms, poor quality of life, poor social and occupational functioning, and high-density lipoprotein were environmental predictors of negative symptom trajectories (Table 2). After excluding depressive symptoms, high-density lipoprotein, and social and occupational functioning, the multivariable model showed that these factors explained approximately 61% of the variance of trajectories. Baseline negative symptoms (High-Decreased vs Low: OR = 2.22; 95% CI = 1.82 - 2.70; p < 0.0001) significantly predicted long-term trajectories of negative symptoms after adjustment for covariables and multiple comparisons (Supplementary Table S9).

Positive symptom trajectories (High vs Low) in siblings were significantly predicted by PRS_{SCZ} at P_T 0.05 (OR = 1.41; 95% CI = 1.09 – 1.82; p = 0.01), PRS_{SCZ} at 0.1 (OR = 1.40; 95% CI = 1.08 – 1.81; p = 0.01) and PRS_{SCZ} at 0.5 (OR = 1.39; 95% CI = 1.07 – 1.80; p = 0.01) in the univariable regression model (Figure 2). Older age, female gender, ethnic minority, more years of education, poor premorbid adjustment, low estimated current IQ, severe psychotic-like experiences, poor quality of life, and poor social and occupational functioning were also significantly associated with positive symptom trajectories (Table 3). These factors, after excluding social and occupational functioning, explained approximately 25% of the variance of trajectories. In the multivariable model, only quality of life (High vs Low: OR = 0.27; 95% CI = 0.11 - 0.70; p = 0.007) was found to be a strong predictor of positive symptoms trajectory

(Supplementary Table S10). There was evidence of an association between PRS_{SCZ} at P_T threshold 0.5 and negative symptom trajectories in siblings though it was not statistically significant (High vs Low: OR = 1.14; 95% CI = 0.87 – 1.49; p = 0.33) (Figure 2). Negative symptom trajectories were also associated with being married, poor premorbid adjustment, severe psychotic-like experiences, poor quality of life, and poor social and occupational functioning (Table 3). These factors (i.e., after excluding social and occupational functioning) explained 33.2% of the variance of trajectories and premorbid adjustment (High vs Low: OR = 4.19; 95% CI = 1.98 - 8.84; p = 0.0002) and quality of life (High vs Low: OR = 0.05; 95% CI = 0.01 - 0.20; p < 0.0001) were found to be a strong predictor of negative symptoms trajectory after adjustment for covariables and multiple comparisons correction in the multivariable model (Supplementary Table S10).

In controls, there was no evidence of significant association of PRS_{SCZ} at P_T threshold 5e-08 with positive symptom trajectories (High-Decreased vs Low: OR = 1.29; 95% CI = 0.69 – 2.40; p = 0.42) (Figure 2). Positive symptom trajectories were associated with older age, ethnic minority, poor premorbid adjustment, severe psychotic-like experiences, poor quality of life, and poor social and occupational functioning (Table 4). After excluding social and occupational functioning, these factors explained 26.8% of the variance of trajectories and none of these factors were significantly associated with positive symptoms trajectories after adjustment for covariables and multiple comparisons correction (Supplementary Table S11). There was no evidence of association between PRS_{SCZ} at P_T threshold 0.5 and negative symptom trajectories (High vs Low: OR = 1.20; 95% CI = 0.83 – 1.74; p = 0.33) (Figure 2). In addition, poor premorbid adjustment, severe psychotic-like experience, poor quality of life, and poor social and occupational functioning significantly associated with a severe negative symptoms trajectory, and they are attributed to approximately 32% of explained variance after excluding social and occupational functioning (Table 4). None of these factors were significantly associated with negative symptoms trajectories after adjustment for covariables and multiple comparisons correction in the multivariable model (Supplementary Table S11).

Table 2: Univariable multinomial random-effects logistic regression model on the environmental predictors of positive and negative symptoms trajectories in patients.

Predictors	Positive symptom trajectories (Ref=Low)				Negative symptom trajectories (Ref=Low)			
	Moderate		High		High-Decreased		High-Increased	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Demographic and clinical characteristics								
Age	0.98(0.97,1.01)	0.16	1.01(0.99,1.04)	0.28	0.97(0.95,1.00)	0.05	1.02(0.99,1.04)	0.22
Gender, female	0.64(0.45,0.91)	0.01	0.24(0.12,0.51)	0.0002	0.54(0.32,0.92)	0.023	0.60(0.34,1.05)	0.07
Marital status, married	0.29(0.15,0.59)	0.0006	0.16(0.04,0.68)	0.013	0.61(0.29,1.29)	0.19	0.60(0.27,1.36)	0.22
Ethnicity, non-Caucasian	1.13(0.78,1.63)	0.53	2.41(1.52,3.84)	0.0002	1.15(0.70,1.87)	0.58	1.54(0.94,2.53)	0.09
Years of education (full-time)	0.97(0.93,1.01)	0.13	0.97(0.92,1.03)	0.36	0.93(0.89,0.98)	0.006	0.96(0.91,1.02)	0.15
Age onset of first psychosis	0.98(0.96,0.99)	0.03	0.97(0.94,1.01)	0.06	0.97(0.95,0.99)	0.03	1.00(0.98,1.03)	0.75
Duration of illness	1.02(0.98,1.07)	0.30	1.09(1.03,1.15)	0.001	0.99(0.95,1.03)	0.64	1.02(0.98,1.06)	0.29
Number of psychotic episodes	1.06(0.93,1.20)	0.39	1.22(1.04,1.43)	0.012	0.93(0.78,1.10)	0.38	0.97(0.82,1.15)	0.72
Antipsychotics, first generation*	1.78(1.04,3.05)	0.035	1.76(0.84,3.71)	0.14	1.62(0.81,3.21)	0.17	1.60(0.77,3.32)	0.21
Premorbid adjustment	1.63(1.36,1.96)	<0.0001	1.68(1.28,2.19)	0.0001	1.86(1.22,2.82)	0.004	2.22(1.44,3.42)	0.0003
Estimated current IQ	0.99(0.97,0.99)	0.003	0.98(0.97,0.99)	0.016	0.98(0.97,0.99)	0.002	0.97(0.96,0.99)	0.0004
Depression‡	1.20(1.13,1.27)	<0.0001	1.21(1.11,1.31)	<0.0001	1.06(0.95,1.19)	0.30	1.22(1.09,1.36)	0.0004
General cognition	0.87(0.78,0.96)	0.009	0.80(0.69,0.92)	0.002	0.79(0.69,0.90)	<0.0001	0.78(0.68,0.90)	<0.0001
Psychotic symptoms								
Positive symptoms	1.39(1.31,1.42)	<0.0001	2.09(1.88,2.33)	<0.0001	1.10(1.05,1.15)	<0.0001	1.08(1.04,1.14)	<0.0001
Negative symptoms	1.06(1.04,1.09)	<0.0001	1.12(1.08,1.15)	<0.0001	2.19(1.97,2.45)	<0.0001	1.54(1.43,1.65)	<0.0001
Disorganization	1.15(1.08,1.22)	<0.0001	1.28(1.19,1.38)	<0.0001	1.26(1.11,1.42)	<0.0001	1.22(1.08,1.38)	<0.0001
Emotional distress	1.23(1.09,1.38)	<0.0001	1.43(1.26,1.64)	<0.0001	1.15(1.09,1.22)	<0.0001	1.11(1.04,1.18)	<0.0001

Table 2: Continued.

Predictors	Positive symptom trajectories (Ref=Low)				Negative symptom trajectories (Ref=Low)			
	Moderate		High		High-Decreased		High-Increased	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Excitement	1.24(1.13,1.37)	<0.0001	1.46(1.31,1.63)	<0.0001	1.35(1.22,1.48)	<0.0001	1.24(1.13,1.36)	<0.0001
Quality of life and functioning								
Quality of life	0.34(0.22,0.55)	<0.0001	0.22(0.12,0.40)	<0.0001	0.26(0.12,0.55)	<0.0001	0.24(0.11,0.51)	<0.0001
Social functioning[†]	0.93(0.92,0.95)	<0.0001	0.92(0.89,0.95)	<0.0001	0.90(0.84,0.97)	0.007	0.86(0.79,0.93)	0.0002
Global functioning	0.97(0.95,0.99)	0.01	0.94(0.92,0.97)	0.0001	0.94(0.91,0.97)	<0.0001	0.96(0.93,0.98)	<0.0001
Occupational functioning[†]	0.86(0.82,0.91)	<0.0001	0.87(0.80,0.95)	<0.0001	0.92(0.85,1.00)	0.06	0.84(0.77,0.92)	0.0001
Physical health[†]								
Body mass index (kg/m^2)	0.99(0.96,1.04)	0.93	1.01(0.95,1.06)	0.83	1.02(0.97,1.07)	0.39	1.01(0.96,1.06)	0.73
Waist circumference (cm)	1.00(0.99,1.02)	0.52	1.01(0.99,1.03)	0.44	1.01(0.99,1.03)	0.21	1.01(0.99,1.03)	0.22
Glycated haemoglobin (mmol/mol)	1.04(1.01,1.08)	0.022	1.07(1.03,1.12)	0.001	1.03(0.97,1.09)	0.36	1.05(0.99,1.11)	0.09
Triglycerides (mmol/l)	1.09(0.96,1.24)	0.17	1.05(0.86,1.28)	0.66	0.95(0.75,1.20)	0.65	0.92(0.73,1.17)	0.52
High density lipoprotein (mmol/l)	1.04(0.77,1.39)	0.80	0.31(0.11,0.83)	0.02	0.54(0.20,1.43)	0.21	0.26(0.09,0.76)	0.01
Low density lipoprotein (mmol/l)	1.06(0.85,1.32)	0.60	1.55(1.13,2.11)	0.006	1.01(0.71,1.44)	0.97	0.87(0.61,1.24)	0.45
Diastolic blood pressure (mmHg)	1.01(0.99,1.03)	0.21	1.01(0.99,1.04)	0.26	1.01(0.98,1.03)	0.54	1.01(0.99,1.03)	0.38
Systolic blood pressure (mmHg)	1.00(0.99,1.01)	0.85	1.01(0.99,1.03)	0.32	0.99(0.98,1.01)	0.89	0.99(0.97,1.01)	0.17
Pulse rate (beat/min)	1.01(1.01,1.03)	0.022	1.03(1.01,1.04)	0.004	1.02(0.99,1.03)	0.11	1.01(0.99,1.03)	0.16
Metabolic syndrome score	1.17(0.94,1.44)	0.15	1.46(1.09,1.95)	0.01	1.35(0.91,2.01)	0.14	1.45(0.98,2.14)	0.06

[†]= *Used from the second wave at a three-year follow-up.*
[*]= *Combination of medications and switching medication not considered, and the reference category is 'second' generation antipsychotics.*

DISCUSSION

We investigated the long-term trajectories of positive and negative symptoms among patients with schizophrenia-spectrum disorder, their unaffected siblings and healthy controls as well as the association of identified trajectories with genetic and non-genetic factors. We identified three trajectories of positive and negative symptoms in patients and controls, and four trajectories in siblings. Substantial differences in variability of trajectories were observed across samples, while a similar level of variability was observed across symptom domains. Poor premorbid adjustment, poor quality of life and severe positive and negative symptoms at baseline significantly predicted the six-year trajectories of positive and negative symptoms in patients and siblings. Findings suggested that the association between PRS_{SCZ} and both positive and negative symptoms was small but significant. Most findings became insignificant in the multivariate model corrected for multiple comparisons.

For positive symptoms, we identified three trajectory groups of patients and controls compared to four trajectory groups in siblings characterized by stability (more in siblings and controls), progressive decline (in siblings and controls) and relapsing of symptoms (only in patients). The majority (>65%) of patients belong to low trajectory groups and stays constant over time. Besides, the majority (>50%) of siblings and controls belong to trajectory groups with moderate severity of symptoms indicating their liability to develop psychosis over time. In our previous review (13), we have seen a similar number of trajectories or clusters that were identified by longitudinal and cross-sectional studies in patients, unaffected siblings and healthy controls. In this study, we also observed greater variation in trajectories of negative symptoms, i.e., the stability of symptoms observed in controls, a decline of symptoms observed in patients, and progressive worsening and persistence of symptoms observed in both patients and siblings. Results on both positive and negative symptoms are in line with our review.(13) We ensured the validity of identified trajectories in our current study by comparing results across the study population (e.g., patients vs controls) and findings from previous studies, as we summarized elsewhere.(13) In general, it has previously been observed that two to five trajectory groups were eminent across psychotic symptom dimensions and population groups (i.e., patients, siblings and controls), though the patterns were largely different.(13) Moreover, the clinical meaningfulness of trajectory groups was also carefully examined through directly involving clinicians during trajectory modeling. The observed variations in the num-

Table 3: Univariable multinomial random-effects logistic regression model on the environmental predictors of positive and negative symptoms trajectories in siblings.

Predictors	Positive symptom trajectories (Ref=Low)				Negative symptom trajectories (Ref=Low)			
	Moderate		High (high and high-decreased)		Moderate		High (Increased and Decreased)	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Demographic and clinical characteristics								
Age	0.98(0.96,1.01)	0.17	0.97(0.94,0.99)	0.016	1.00(0.98,1.02)	0.97	1.01(0.99,1.04)	0.29
Gender, female	1.62(1.12,2.32)	0.01	1.77(1.15,2.71)	0.01	1.27(0.94,1.73)	0.12	0.75(0.47,1.19)	0.22
Marital status, married	1.02(0.71,1.48)	0.91	0.63(0.40,0.98)	0.04	0.90(0.66,1.24)	0.53	0.45(0.27,0.76)	0.003
Ethnicity, non-Caucasian	2.46(1.37,4.42)	0.003	2.38(1.23,4.58)	0.01	1.08(0.70,1.67)	0.73	1.18(0.63,2.21)	0.61
Years of education (full-time)	0.94(0.89,0.99)	0.013	0.92(0.87,0.98)	0.008	0.99(0.95,1.03)	0.67	0.95(0.90,1.01)	0.10
Premorbid adjustment	1.70(1.22,2.36)	0.002	2.49(1.72,3.59)	<0.0001	2.78(1.98,3.92)	<0.0001	7.13(4.59,11.08)	<0.0001
Estimated current IQ	0.99(0.98,1.01)	0.19	0.98(0.97,0.99)	0.02	0.99(0.98,1.01)	0.46	0.99(0.98,1.01)	0.48
Depression[†]	1.01(0.52,1.97)	0.97	1.49(0.78,2.85)	0.23	0.99(0.68,1.45)	0.98	1.43(0.99,2.07)	0.06
General cognition	1.01(0.88,1.16)	0.87	0.95(0.82,1.10)	0.48	0.97(0.87,1.07)	0.55	0.93(0.80,1.07)	0.31
Psychotic-like experiences (distress)								
Positive symptoms	1.92(1.08,3.39)	0.026	6.58(3.56,12.15)	<0.0001	2.95(1.76,4.95)	<0.0001	9.98(5.32,18.70)	<0.0001
Negative symptoms	3.04(1.77,5.24)	<0.0001	14.04(7.65,25.77)	<0.0001	2.49(1.65,3.76)	<0.0001	8.48(4.98,14.46)	<0.0001
Depressive symptoms	1.96(1.29,2.99)	0.002	7.21(4.44,11.70)	<0.0001	2.23(1.54,3.23)	<0.0001	6.93(4.27,11.25)	<0.0001

Table 3: Continued.

Predictors	Positive symptom trajectories (Ref=Low)				Negative symptom trajectories (Ref=Low)			
	Moderate		High (high and high-decreased)		Moderate		High (Increased and Decreased)	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Quality of life and functioning								
Quality of life	0.28(0.17,0.47)	<0.0001	0.06(0.04,0.12)	<0.0001	0.15(0.09,0.26)	<0.0001	0.02(0.01,0.05)	<0.0001
Social functioning[†]	0.98(0.94,1.02)	0.30	0.93(0.89,0.97)	0.0004	0.91(0.87,0.95)	<0.0001	0.78(0.74,0.83)	<0.0001
Occupational functionin[†]	0.85(0.74,0.97)	0.019	0.76(0.66,0.88)	0.002	0.87(0.79,0.97)	0.009	0.76(0.68,0.85)	0.0001

[†]= *Used from the second wave at a three-year follow-up.*

ber of trajectories across studies may be due to several reasons. The use of different trajectory modeling techniques may lead to the identification of a different number of subgroups. For example, a study on the application of five statistical data-driven subtyping methods on longitudinal data showed that the number of trajectory groups derived from one method can be remarkably different from the other method using the same data structure.(54) Moreover, differences in patient characteristics, the use of different instruments to measure symptoms, different options for treatment in different countries and differences in frequency and duration of follow-up may affect the number of identified trajectories.(13)

PRS_{SCZ} significantly correlated with the severity of baseline negative symptoms in patients and controls while it correlated with positive symptoms in siblings. There was also some evidence for an association between PRS_{SCZ} and the six-year trajectory of positive symptoms in both patients and siblings, though all the associations disappeared after adjustment for environmental factors and multiple comparisons. These findings are not ignorable and suggest at least in part that genetic risk for schizophrenia might be expressed by negative symptoms at the onset of illness and might be expressed by positive symptoms later during illness progression. It is important to emphasize the dynamic changes of symptoms over time to obtain a more accurate estimate of the relationship between genetic factors and quantitative phenotypes. Previous studies on the association between psychosis phenotypes and PRS_{SCZ} have largely shown inconsistent results. A systematic review of PRS-based studies showed that PRS_{SCZ} was significantly associated with the severity of negative symptoms, but not with positive symptoms, in patients and healthy general population wherein most estimates were based on correlation tests.(20) A population-based birth cohort study also showed that PRS_{SCZ} was significantly associated with negative symptoms in adolescence in the general population, while others found no evidence for an association between PRS_{SCZ} and psychotic experiences/positive symptoms.(12,55) This is concordant with our findings from the bivariate correlation analyses. Other cross-sectional and longitudinal studies (10,21,56-58) found no association between PRS_{SCZ} and positive and/or negative symptoms in patients and healthy individuals. The inconsistencies between the genetic studies of quantitative phenotypes of schizophrenia could be due to the difference in the assessment instruments for positive and negative symptoms, sampling, sample size, genotyping and quality control methods used in GWAS, and inclusion of patients at vastly different stages of illness and with a diverse spectrum of symptoms. Additionally,

PRS is highly dependent on factors, such as the sample characteristics, sample size, stage and/or severity of the disease that leads to variation in findings across studies. (59)

This is the first study to investigate the effect of PRS_{SCZ} on the long-term trajectories of psychotic symptoms in patients, siblings and healthy controls. A genetic marker like PRS_{SCZ} is believed to be a specific and sensitive biomarker that shows the inherent heterogeneity in the course of symptoms. Thus, the results in our study may give some indication that both positive and negative symptoms are likely to be important markers of high genetic risk for schizophrenia both in patients and healthy populations though replication is still necessary to make a strong conclusion. Investigating the phenotypic manifestations of genetic susceptibility for schizophrenia (as measured by PRS) will contribute to the understanding of symptoms that have a stronger genetic liability and their underlying pathophysiological processes specific to a clinical subtype or symptom dimension.(55,60) This may eventually help to improve risk prediction, diag nosis and prognosis.(55,60) The evidence could also provide leads in the development of novel treatments since it is symptom dimensions, rather than the illness itself, that are ameliorated by medications.(19)

In agreement with a previous study(7), baseline positive and negative symptoms significantly predicted positive and negative symptoms trajectories in patients, respectively. However, there was no strong evidence that negative symptoms predicted later positive symptoms or vice versa. This finding suggests that patients who had severe symptoms at baseline showed persistence and stability of the initial level of symptoms.(19) On the other hand, an earlier study showed that the severity of positive symptoms initially decreased and became stable over time while negative symptoms showed persistence over time.(8) In siblings, poor premorbid adjustment and quality of life were found to be the strong predictors of positive and negative trajectories. In controls, none of the environmental factors survived adjustment for covariables and multiple comparisons. The lack of association following adjustment for covariables in our study is not surprising given that these factors were selected from previous group-based trajectory modeling studies that mostly performed univariable analyses or just compared trajectories using proportion or mean estimates.(13) In general, at least in the unadjusted model, multiple environmental factors were found to be strong predictors of positive and negative symptoms trajectories and this support the notion that positive and negative symptoms share a similar course and perhaps mechanisms.(3,5,6)

Table 4: Univariable multinomial random-effects logistic regression model on the environmental predictors of positive and negative symptoms trajectories in controls.

Predictors	Positive symptom trajectories (Ref=Low)				Negative symptom trajectories (Ref=Low)			
	Moderate		High-Decreased		Moderate		High	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Demographic and clinical characteristics								
Age	0.97(0.94,1.01)	0.09	0.94(0.89,0.99)	0.018	0.99(0.97,1.02)	0.85	1.01(0.98,1.04)	0.62
Gender, female	1.15(0.64,2.05)	0.64	2.63(0.95,7.32)	0.06	1.09(0.68,1.75)	0.73	1.24(0.65,2.35)	0.51
Marital status, married	0.81(0.44,1.51)	0.51	0.75(0.28,2.04)	0.58	1.32(0.79,2.20)	0.28	0.79(0.39,1.58)	0.50
Ethnicity, non-Caucasian	4.47(1.22,16.40)	0.024	3.33(0.49,22.52)	0.22	2.17(0.76,6.17)	0.14	1.32(0.32,5.41)	0.70
Years of education (full-time)	0.93(0.85,1.02)	0.11	0.93(0.80,1.08)	0.33	1.03(0.96,1.12)	0.39	1.03(0.93,1.13)	0.60
Premorbid adjustment	2.40(1.30,4.44)	0.005	3.83(1.67,8.80)	0.002	3.31(1.52,7.18)	0.003	10.08(4.28,23.72)	<0.0001
Estimated current IQ	0.98(0.96,0.99)	0.046	0.97(0.94,1.01)	0.05	1.01(0.99,1.02)	0.66	0.99(0.97,1.02)	0.68
Depression[†]	1.06(0.66,1.69)	0.82	1.13(0.62,2.06)	0.69	3.23(0.47,22.38)	0.23	3.90(0.56,27.39)	0.17
General cognition	0.90(0.73,1.09)	0.28	0.84(0.63,1.14)	0.26	1.09(0.95,1.26)	0.21	0.91(0.76,1.10)	0.34
Psychotic-like experiences (distress)								
Positive symptoms	4.02(1.53,10.54)	0.005	10.78(3.25,35.76)	0.0001	4.80(1.55,14.88)	0.007	18.11(5.27,62.23)	<0.0001
Negative symptoms	3.54(1.63,7.72)	0.002	13.46(4.49,40.35)	<0.0001	4.91(1.54,15.67)	0.007	13.78(3.96,48.00)	<0.0001
Depressive symptoms	2.98(1.53,5.80)	0.001	10.50(4.21,26.20)	<0.0001	3.57(1.74,7.32)	0.0005	8.80(3.85,20.10)	<0.0001

Table 4: Continued.

Predictors	Positive symptom trajectories (Ref=Low)				Negative symptom trajectories (Ref=Low)			
	Moderate		High-Decreased		Moderate		High	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Quality of life and functioning								
Quality of life	0.07(0.02,0.27)	0.0001	0.02(0.01,0.07)	<0.0001	0.11(0.03,0.40)	0.0008	0.02(0.01,0.06)	<0.0001
Social functioning[†]	0.66(0.51,0.85)	0.001	0.64(0.49,0.83)	0.002	0.89(0.83,0.96)	0.002	0.80(0.73,0.87)	<0.0001
Occupational functioning[†]	0.82(0.69,0.96)	0.016	0.73(0.59,0.90)	0.003	0.88(0.76,1.03)	0.11	0.85(0.71,1.01)	0.07

[†]= *Used from the second wave at a three-year follow-up.*

The identification of symptom trajectories and changes over time could have relevant implications for initiating preventive strategies and close monitoring. For example, people characterized by poor symptom trajectories could be the primary target to receive treatment(s), which could potentially improve the long term clinical and functional outcomes. Our study also provides evidence that environmental symptom level factors may be valuable and prognostic indicators of the trajectory of positive and negative symptoms compared to PRS_{SCZ}. To date, there was no study conducted to explore the long-term trajectories of positive and negative symptoms in siblings of probands. Thus, our findings in siblings could have potentially important implications for high-risk groups liable to develop psychosis, among whom current approaches for informing prediction of transition rely heavily on psychotic experiences.(12) It may also provide some clues to developing more effective treatments.(19) A better understanding of trajectories of symptoms and how genetic risk for schizophrenia manifests during illness could inform early recognition of disorders in those at greatest risk and potentially inform targeted interventions. Given that these symptoms contribute largely to morbidity and poor functioning in schizophrenia, the present findings hold promise to inform further research and potentially guide an objective approach to estimate disease liability, diagnosis of disease and development of precise therapeutic interventions.

In this study, we investigated the long-term trajectories of schizophrenia symptoms and their association with a broad range of genetic and environmental factors in patients and healthy individuals. This can help to easily compare the pattern of evidence. Interestingly, results were comparable across patients, unaffected siblings and healthy controls, and provided a similar trend of evidence, though the level of statistical significance was different. A longitudinal investigation of symptoms is a more optimal method for ascertaining the long-term variation in severity and trajectories of symptoms.(61) This study has also some limitations. PRS is based on common genetic variants and naturally does not capture copy number variants (CNV), or rare SNP contributions to variance. Besides, trajectory group identification and their association with PRS_{SCZ} may be different if it is done based on other symptom definitions, e.g. total PANSS score, a subdomain of positive (e.g., hallucination, delusion, disorganization) and negative (e.g., apathy, avolition, amotivation) symptoms, or PRS constructed based GWAS summary statistics of these quantitative phenotypes.

CONCLUSIONS

To our knowledge, this is the first study that examined the long-term trajectories of schizophrenia symptoms and the role of genetic susceptibility and multidimensional factors among patients, siblings and healthy controls. Three to four clinically and statistically meaningful trajectories were identified with large variations in patterns. Our study indicated that environmental factors contributed to the large variation in the severity of symptoms and were found to be a predictor of trajectories in schizophrenia spectrum disorders. Of interest is our finding of a small yet significant correlation and longitudinal association between PRS_{SCZ} and trajectories of positive and negative symptoms in patients and healthy individuals, though this did not survive the adjustment for confounders and multiple comparisons. This evidence, at least in part, provides additional support for genetic factors underlying the observed clinical heterogeneity in schizophrenia.(19) Taken together, we found some evidence that supports the notion that positive and negative symptoms share similar etiopathogenic mechanisms and they may be equally complex sub-phenotypes. Additionally, our results suggest that positive symptoms are as stable as negative symptoms.(19) Large-scale longitudinal studies with robust measures of quantitative phenotypes using a harmonized measuring instrument are required to determine how genetic risk for schizophrenia is expressed and whether this expression changes with time, to examine potential mediators and moderators of risk, and to determine the usefulness of PRS_{SCZ} for prediction of transition to psychosis.(12)

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