

## REGULAR RESEARCH ARTICLE

# Genetically Determined Levels of Serum Metabolites and Risk of Neuroticism: A Mendelian Randomization Study

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## Abstract

**Background:** Neuroticism is a strong predictor for a variety of social and behavioral outcomes, but the etiology is still unknown. Our study aims to provide a comprehensive investigation of causal effects of serum metabolome phenotypes on risk of neuroticism using Mendelian randomization (MR) approaches.

**Methods:** Genetic associations with 486 metabolic traits were utilized as exposures, and data from a large genome-wide association study of neuroticism were selected as outcome. For MR analysis, we used the standard inverse-variance weighted (IVW) method for primary MR analysis and 3 additional MR methods (MR-Egger, weighted median, and MR pleiotropy residual sum and outlier) for sensitivity analyses.

**Results:** Our study identified 31 metabolites that might have causal effects on neuroticism. Of the 31 metabolites, uric acid and paraxanthine showed robustly significant association with neuroticism in all MR methods. Using single nucleotide polymorphisms as instrumental variables, a 1-SD increase in uric acid was associated with approximately 30% lower risk of neuroticism (OR: 0.77; 95% CI: 0.62–0.95;  $P_{IVW} = 0.0145$ ), whereas a 1-SD increase in paraxanthine was associated with a 7% higher risk of neuroticism (OR: 1.07; 95% CI: 1.01–1.12;  $P_{IVW} = .0145$ ).

**Discussion:** Our study suggested an increased level of uric acid was associated with lower risk of neuroticism, whereas paraxanthine showed the contrary effect. Our study provided novel insight by combining metabolomics with genomics to help understand the pathogenesis of neuroticism.

**Key Words:** Neuroticism, genetically determined metabolite, Mendelian randomization, uric acid, paraxanthine

## Introduction

Neuroticism is a human personality trait characterized by a tendency to respond with negative emotions to threat, frustration, or loss (Lahey, 2009). Individuals with neuroticism often believe

that the world is dangerous and one's ability is not enough to cope with challenging events (Barlow et al., 2014). Neuroticism can also predict a variety of social and behavioral outcomes,

## Significance Statement

This is the first Mendelian randomization (MR) study, to our knowledge, to assess the causal relationship between serum metabolites and neuroticism. MR study could avoid confounding, reverse causation, and various biases compared with traditional observational study. Using genetic variants as instrumental variables, our results showed that urate had protective effects on neuroticism while paraxanthine had adverse effects on neuroticism. We also found some significant metabolic pathways that may be involved in neuroticism. By combining metabolomics with genomics, we provide new insight in the causal relationship between serum metabolites and neuroticism, which may contribute to the understanding of pathogenesis and potential therapeutic targets for neuroticism.

especially common psychiatric symptoms, including anxiety, depressive, social phobia, and substance use (Ormel et al., 2013b, 2013a). Genetic factors contribute substantially to the pathogenesis of neuroticism, accounting for 40% to 60% of the variance of this trait (Clark et al., 1994; Hettema et al., 2005; Fullerton, 2006). Recent genome-wide association studies (GWASs) have yielded substantial findings for improving our understanding of the etiological mechanism of neuroticism (Okbay et al., 2016; Luciano et al., 2018; Nagel et al., 2018). However, functional annotations towards biological processes are still lacking.

Substantial effort has been expended to understand the biological mechanisms through which GWAS-identified risk variants act on target diseases, including the introduction of expression quantitative trait loci, methylation quantitative trait loci, and protein quantitative trait loci (Westra et al., 2013; Consortium et al., 2017; McRae et al., 2018; Yao et al., 2018). Recent studies have proposed metabolic traits as functional intermediates to reveal the relevant biological processes of disease-related genetic variants (Gieger et al., 2008; Suhre et al., 2011). Metabolites are a group of small molecules widely distributed throughout the cells, tissues, and fluids of human bodies (Quinones and Kaddurah-Doouk, 2009). Alterations in the concentration of metabolites can serve as useful biomarkers and provide early evidence for the diagnosis and prognosis of pathologies (Assfalg et al., 2008; Holmes et al., 2008). Especially in recent years, the rapidly evolving field of metabolomics is providing a systematic readout of the metabolic state of an individual. GWASs with non-targeted metabolomics have been conducted and the concept of “genetically determined metabolotypes (GDMs)” proposed to further our understanding of the relationships and interactions between genetic variations and environmental triggers of disease. GDMs serve as useful intermediate phenotypes to explain the causal roles of genetic variants in human diseases.

Mendelian randomization (MR) is a novel technique that uses genetic variants as instrumental variables to assess causal inferences between risk factors and clinical outcomes of interest (Choi et al., 2019). The fundamental principle of the MR study design is that genetic variants, which indicate the biological effects or level of effects of a modifiable exposure, should be causally associated with exposure-related disease risk. This approach is largely independent of the biases inherent in observational studies, given that genetic variation is unlikely to be affected by environmental factors (Pierce et al., 2011). During the past years, the explosion in publicly available GWAS summary data has made the MR popular in inferring causality, especially the 2-sample MR design that utilizes just 1 pair of GWAS summary statistics (Zheng et al., 2017). Based on the 2-sample MR approach, we are able to link GDMs with neuroticism using GWAS estimates on both metabolic phenotypes and neuroticism. The aims of the present study were to: (1) assess the causal relationships between genetically determined levels of serum metabolites and risk of neuroticism, (2) investigate the

genetic variants that might have important roles in determining the variation of corresponding metabolites and risk of neuroticism, and (3) identify potential metabolic pathways that may be involved with the pathogenesis of neuroticism.

## Methods

### Genetically Determined Metabolites

Data for genetic associations with serum metabolites were downloaded from the Metabolomics GWAS server (<http://metabolomics.helmholtz-muenchen.de/gwas/>). Briefly, Shin et al. (Shin et al., 2014) conducted the most comprehensive genome-wide association estimates with high-throughput metabolic profiling as phenotypes and developed the atlas of genetic influences on human blood metabolites. The study sample was composed of 7824 adult individuals from 2 European population studies. All participants gave written informed consent, and the study was approved by local ethics committees. Metabolic profiling was performed based on non-targeted mass spectrometry analysis on human fasting serum. After merging the datasets of the 2 studies, a subset of 486 metabolites was available for GWAS analysis. Of the 486 total metabolites, 309 were classified as known, implying that their chemical identity had been determined according to Metabolon's spectra library (Metabolon, Inc., Morrisville, NC). The 309 known metabolites were further assigned to 8 biochemical classes (amino acids, peptides, lipids, cofactors and vitamins, carbohydrates, energy, nucleotides, and xenobiotics), as described in the Kyoto Encyclopedia of Genes and Genomes (KEGG) database. The genotyping and imputation steps for the 2 cohorts were described in detail in previous studies (Suhre et al., 2011; Shin et al., 2014). Finally, approximately 2.1 million single nucleotide polymorphisms (SNPs) present in both cohorts were tested in the primary association meta-analysis.

### Selection of Instrumental Variables

The principle of MR relies on the basic assumption of valid instrumental variables; this particularly requires the extracted SNPs to be (1) significantly related to exposure, (2) independent of any confounder, and (3) associated with the outcome directly through the exposure. To determine the instrumental variables for the 486 metabolites, we first extracted genetic variants with association thresholds at  $P < 1 \times 10^{-5}$  to ensure they conferred substantial variance for the corresponding metabolites. Second, we identified independent variants using a clumping procedure implemented in PLINK1.9 (<http://www.cog-genomics.org/plink2/>), setting a linkage-disequilibrium threshold of  $r^2 < 0.1$  with 500 kb in the European 1000 Genomes Project Phase 3 reference panel. We next assessed whether these instrumental variables were strong enough to predict

metabolite levels by 2 parameters: the explained variance (adjusted  $R^2$ ) and F statistic. The adjusted  $R^2$  was estimated using the “gtx” package in R software (<https://www.r-project.org/>), and the F statistic was calculated based on the formula  $F = R^2(n-1-k)/(1-R^2)k$ , where n is the sample size and k represents the number of genetic variants (Brion et al., 2013). A threshold of  $F > 10$  was typically recommended for MR analyses (Burgess et al., 2013).

### GWAS of Neuroticism

We obtained the GWAS summary statistics for neuroticism from the GWAS Catalog (<https://www.ebi.ac.uk/gwas/>). Briefly, Okbay et al. (Okbay et al., 2016) conducted the large GWAS meta-analysis on 170 910 individuals from 59 cohorts. Neuroticism was measured according to a 12-item version of the Eysenck Personality Inventory Neuroticism scale. Genotyping was performed separately for each cohort using different versions of Affymetrix arrays. Quality-control procedures were conducted using the EasyQC software following standard protocols (Winkler et al., 2014). Association analysis was carried out with a logistic regression model correcting for age, gender, and study-specific covariates. All participants provided written informed consent, and ethical approvals were obtained for each cohort. Detailed information for cohort descriptions, quality-control filters, and imputation procedures can be found in the previously published study. Finally, a total of 6 524 442 SNPs were included in the GWAS summary statistics.

### MR Analyses

We used the inverse-variance weighted (IVW) method as the primary MR analysis to determine the causal relationships between genetically determined levels of serum metabolites and neuroticism. Briefly, the IVW method provided a consistent estimate of the causal effect of the exposure when each variant satisfied the assumption of valid instrumental variable (Burgess et al., 2013). We then used additional methods to provide sensitivity analysis for the robustness of the MR estimates. MR-Egger was a useful sensitivity analysis that provided consistent estimates with invalid instruments (Bowden et al., 2015). The MR-Egger approach was able to detect certain violations of the standard instrumental variable assumptions and provide an effect estimate not subject to these violations. The weighted median method was another approach that provided consistent estimates even when up to 50% of the variants were deemed invalid instruments (Bowden et al., 2016). MR-PRESSO (MR pleiotropy residual sum and outlier) was another newly developed MR approach that could detect and correct for horizontal pleiotropic outliers to provide a corrected estimate (Verbanck et al., 2018). Furthermore, we conducted the MR-PRESSO Global test to evaluate whether horizontal pleiotropy existed. All analyses were performed using the MendelianRandomization and MR-PRESSO packages in R software version 3.4.4.  $P < .05$  was considered statistically significant.

### Metabolic Pathway Analysis

Metabolic pathway analysis was conducted using the web-based MetaboAnalyst 4.0 (<https://www.metaboanalyst.ca/>) (Chong et al., 2018). We analyzed the metabolite sets from 2 libraries in our study, including the Small Molecule Pathway Database and the KEGG Database (Frolkis et al., 2010; Kanehisa et al., 2012). All metabolites that passed the suggestive threshold of association

by IVW ( $P_{IVW} < .05$ ) were included in the metabolic pathway analysis.

## Results

### Strength of the Instrumental Variables

We performed a 2-sample MR analysis to assess the causal effects of human blood metabolites on neuroticism using a pair of GWAS summary statistics. The instrumental variables generated were composed of 3–675 SNPs and could explain 0.8%–83.5% of the variance of their corresponding metabolites. The minimum F statistic for indicating the strength of these instrumental variables was 20.33, meaning that all instrumental variables for the 486 metabolites were sufficiently effective ( $F$  statistic  $> 10$ ) for MR analysis.

### Effects of Genetically Determined Metabolites on Neuroticism

The IVW identified 17 known metabolites and 14 unknown metabolites that were significantly associated with neuroticism (Figure 1; supplementary Table 1). Of the 17 known metabolites, bilirubin (Z,Z) showed the most significant association with neuroticism ( $P_{IVW} = .0005$ ). Using 20 SNPs as proxy predictors, a 1-SD increase in the level of bilirubin (Z,Z) was associated with an approximately 10% lower risk of neuroticism (odds ratio [OR]: 0.93; 95% confidence interval [CI]: 0.89–0.97). We also identified 7 other metabolites that were associated with lower risk of neuroticism, including phenyllactate (OR: 0.87; 95% CI: 0.77–0.98;  $P_{IVW} = .0186$ ), glutamate (OR: 0.76; 95% CI: 0.62–0.93;  $P_{IVW} = .0094$ ), pyruvate (OR: 0.87; 95% CI: 0.78–0.98;  $P_{IVW} = .0256$ ), eicosapentaenoate (20:5n3) (OR: 0.88; 95% CI: 0.80–0.97;  $P_{IVW} = .0096$ ), uric acid (OR: 0.77; 95% CI: 0.62–0.95;  $P_{IVW} = .0145$ ), 4-ethylphenylsulfate (OR: 0.93; 95% CI: 0.87–0.99;  $P_{IVW} = .0480$ ), and 1-methylxanthine (OR: 0.90; 95% CI: 0.82–0.99;  $P_{IVW} = .0402$ ). The other 9 metabolites were associated with increased risk of neuroticism, including pantothenate (OR: 1.12; 95% CI: 1.01–1.23;  $P_{IVW} = .0251$ ), choline (OR: 1.29; 95% CI: 1.01–1.64;  $P_{IVW} = .0449$ ), oleoylcarnitine (OR: 1.17; 95% CI: 1.03–1.33;  $P_{IVW} = .0126$ ), 1-palmitoleoylglycerophosphocholine (OR: 1.15; 95% CI: 1.03–1.29;  $P_{IVW} = .0155$ ), hexanoylecarnitine (OR: 1.10; 95% CI: 1.01–1.21;  $P_{IVW} = .0391$ ), decanoylecarnitine (OR: 1.10; 95% CI: 1.01–1.20;  $P_{IVW} = .0478$ ), gamma-glutamylglutamate (OR: 1.08; 95% CI: 1.02–1.15;  $P_{IVW} = .0088$ ), paraxanthine (OR: 1.07; 95% CI: 1.01–1.12;  $P_{IVW} = .0117$ ), and metoprolol acid metabolite (OR: 1.01; 95% CI: 1.00–1.01;  $P_{IVW} = .0483$ ).

### Sensitivity Analysis

Although the IVW approach is highly effective for inferring causality of an exposure for a complex disease outcome, it is known to be vulnerable to weak instrument bias. We conducted sensitivity analyses to overcome these biases. Table 1 showed the sensitivity analysis results for the IVW identified metabolites. Two metabolites showed robust causal relationships with neuroticism when sensitivity analyses were applied, which were uric acid ( $P_{MR-Egger} = .0091$ ;  $P_{weighted-median} = .0006$  and  $P_{MR-PRESSO} = .0244$ ) and paraxanthine ( $P_{MR-Egger} = .0073$ ;  $P_{weighted-median} = .0004$  and  $P_{MR-PRESSO} = .0256$ ), respectively, and no evidence of horizontal pleiotropy was observed for either metabolite ( $P_{Global\ test} = .0573$  for uric acid and  $P_{Global\ test} = .1376$  for paraxanthine). Using 20 SNPs as predictors, a 1-SD increase in uric acid was associated with an approximately 30% lower risk of neuroticism (OR: 0.77; 95% CI: 0.62–0.95; Figure 2A), whereas a 1-SD increase in paraxanthine

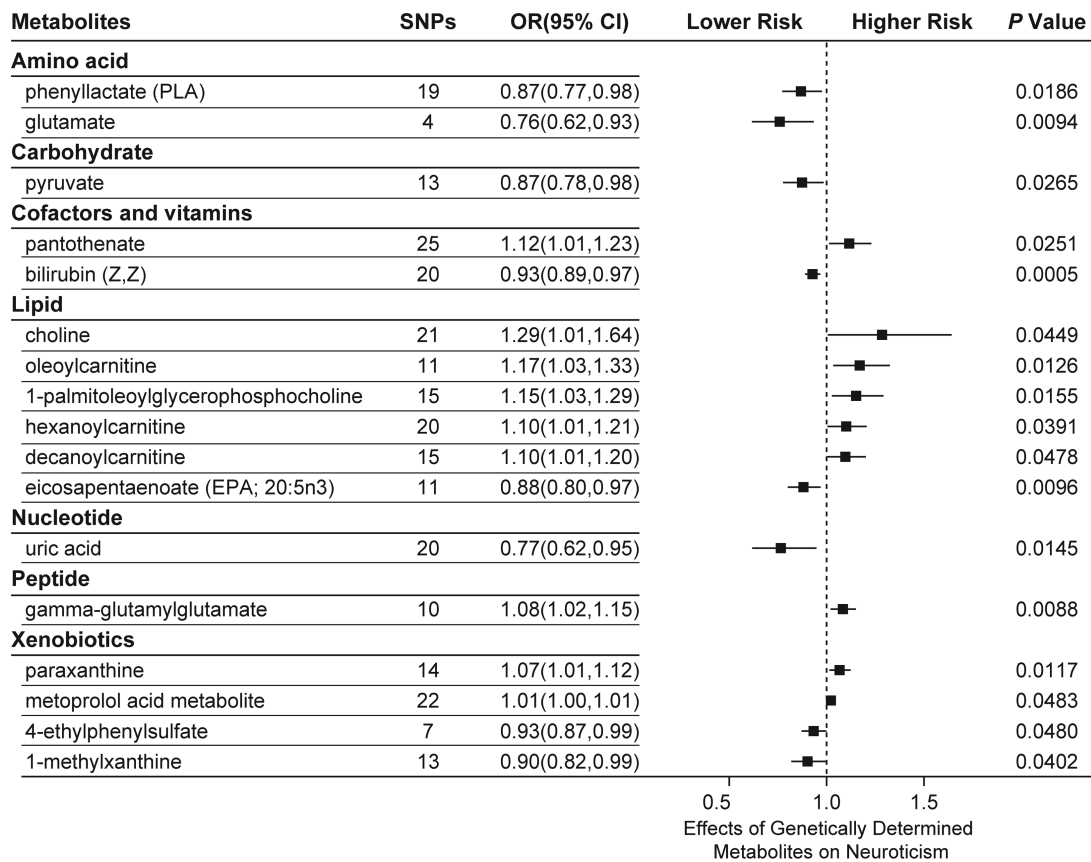


Figure 1. Mendelian randomization associations between serum metabolites and neuroticism based on inverse-variance weighted (IVW) method.

was associated with a 7% higher risk of neuroticism (OR: 1.07; 95% CI: 1.01–1.12; Figure 2B).

### Genetic Determinants

We also investigated the leading SNPs that played important roles in inferring the causal relationships between the identified metabolites and neuroticism. Among the 20 SNPs composing the instrumental variable of uric acid, rs17246501 showed the most significant association ( $P=2.35 \times 10^{-30}$ ) with uric acid (supplementary Table 2). Notably, the rs17246501 also showed a significant association signal ( $P=.0016$ ) with neuroticism. Of the 14 SNPs for paraxanthine, rs3768372 ( $P=.0103$ ), rs10791723 ( $P=.0120$ ), and rs4371296 ( $P=.0499$ ) were also significantly associated with neuroticism (supplementary Table 3). These SNPs might provide valuable information serving as diagnostic or therapeutic targets for neuroticism.

### Metabolic Pathway Analysis

Our study identified 7 significant metabolic pathways that were involved in the pathogenesis of neuroticism (Table 2). The most significant metabolic pathway was the alanine, aspartate, and glutamate metabolism ( $P=.0041$ ) from the KEGG database. Two neuroticism-related metabolites, pyruvate and glutamate, were involved in the metabolic pathway of alanine, aspartate, and glutamate metabolism. We also identified 5 other metabolic pathways from the KEGG database, including pantothenate and CoA biosynthesis ( $P=.0052$ ), butanoate metabolism ( $P=.0111$ ), glycine, serine and threonine metabolism ( $P=.0158$ ), arginine

and proline metabolism ( $P=.0385$ ), and D-glutamine and D-glutamate metabolism ( $P=.0449$ ). There was also a significant metabolic pathway identified from the Small Molecule Pathway Database, which was the caffeine metabolism ( $P=.0295$ ).

### Discussion

Our study performed a 2-sample MR analysis to provide an unbiased estimate of the causal relationships between genetically determined levels of serum metabolites and risk of neuroticism. To the best of our knowledge, this was the first MR study that linked metabolic phenotyping with genomics to assess causal relationships between human serum metabolites and neuroticism. Using SNPs as proxies, our study found that an increased level of uric acid was associated with lower risk of neuroticism, while an increased level of paraxanthine was associated with higher risk of neuroticism. Our study also reported several other metabolites that might contribute to the development of neuroticism.

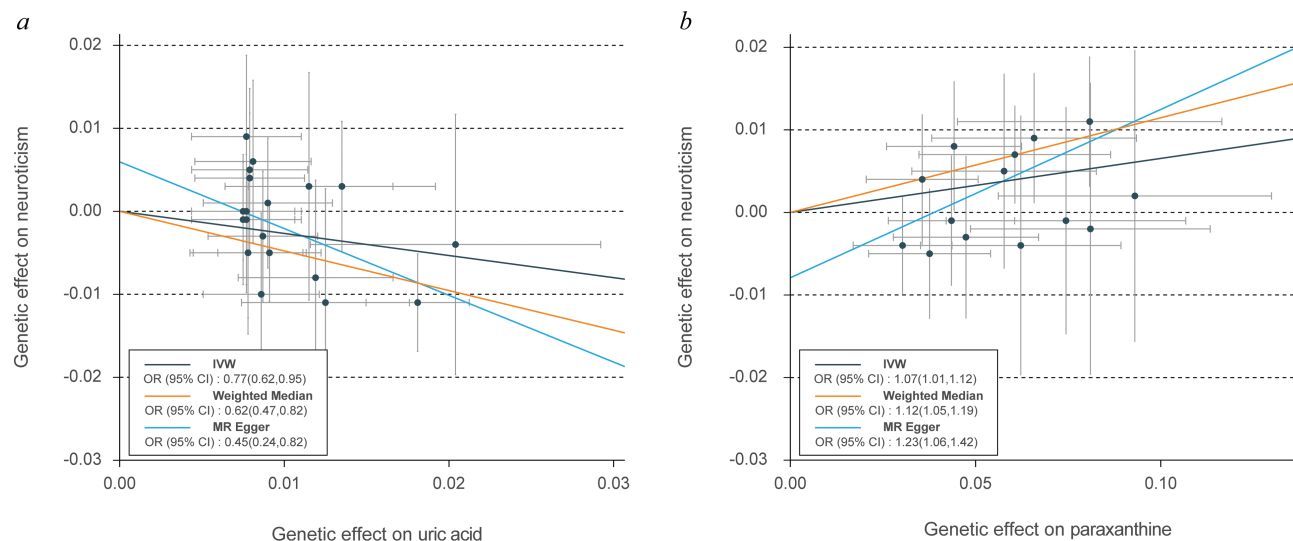
Uric acid is the end product of adenosine metabolism in humans. Lower uric acid has long been recognized as a risk factor for neurological and psychiatric disorders, such as Parkinson's disease, multiple sclerosis, and major depressive and anxiety disorders (Weisskopf et al., 2007; Moccia et al., 2015; Black et al., 2018). Ostojic et al. (Simeunovic Ostojic and Maas, 2018) suggested that uric acid could be a biomarker of mood dysfunction, personality traits, and behavioral patterns. Bartoli et al. (Bartoli et al., 2018b) reported that lower uric acid serum levels were associated with psychological distress and suicidal ideation severity. In addition, a recent meta-analysis further highlighted

Table 1. Sensitivity Analysis of Causal Associations Between IVW Identified Metabolites and Neuroticism

Metabolites	MR-Egger		Weighted median		MR-PRESSO		Global test	
	OR (95% CI)	P value	OR (95% CI)	P value	OR (95% CI)	P value	RSS	P value
Amino acid								
Phenylacetate	0.98 (0.70,1.37)	.9082	0.87 (0.74,1.01)	.0756	0.87 (0.77,0.98)	.0301	24.42	.2602
Glutamate	0.01 (0.00,540.44)	.3870	0.73 (0.55,0.96)	.0268	0.76 (0.64,0.90)	.0514	3.59	.5940
Carbohydrate								
Pyruvate	0.70 (0.48,1.04)	.0752	0.81 (0.69,0.94)	.0070	0.87 (0.78,0.98)	.0466	17.50	.2715
Cofactors and vitamins								
Pantothenate	1.20 (0.91,1.59)	.2002	1.13 (0.98,1.30)	.1057	1.12 (1.03,1.22)	.0185	19.94	.7968
Bilirubin (Z,Z)	0.93 (0.81,1.07)	.2975	0.94 (0.89,0.99)	.0450	0.93 (0.89,0.97)	.0025	26.18	.2416
Lipid								
Choline	0.88 (0.32,2.45)	.8120	1.34 (0.95,1.89)	.1010	1.29 (1.03,1.60)	.0344	17.55	.7241
Oleoyl carnitine	1.68 (0.71,3.94)	.2369	1.19 (1.01,1.41)	.0375	1.17 (1.06,1.30)	.0129	8.14	.7654
1-Palmitoleoylglycerophosphocholine	1.28 (0.88,1.85)	.1917	1.19 (1.02,1.40)	.0292	1.15 (1.04,1.28)	.0210	13.86	.6112
Hexanoyl carnitine	1.06 (0.79,1.42)	.7129	1.15 (1.02,1.30)	.0223	1.10 (1.01,1.21)	.0530	26.67	.2009
Decanoyl carnitine	1.32 (0.83,2.11)	.2433	1.18 (1.04,1.33)	.0121	1.10 (1.01,1.20)	.0678	18.48	.3151
Eicosapentaenoate (20:5n3)	0.80 (0.65,1.01)	.0518	0.89 (0.77,1.02)	.0842	0.88 (0.80,0.97)	.0244	12.79	.4204
Nucleotide								
Uric acid	0.45 (0.24,0.82)	.0091	0.62 (0.47,0.82)	.0006	0.77 (0.62,0.95)	.0244	33.25	.0573
Peptide								
Gamma-glutamylglutamate	1.07 (0.91,1.25)	.4155	1.09 (0.99,1.19)	.0685	1.08 (1.02,1.15)	.0278	11.23	.4629
Xenobiotics								
Paraxanthine	1.23 (1.06,1.42)	.0073	1.12 (1.05,1.19)	.0004	1.07 (1.01,1.12)	.0256	22.18	.1376
Metoprolol acid metabolite	1.01 (0.99,1.02)	.0718	1.01 (0.99,1.01)	.1977	1.01 (1.00,1.01)	.0516	20.83	.5946
4-Ethylphenylsulfate	1.02 (0.83,1.25)	.8765	0.91 (0.83,0.99)	.0386	0.93 (0.88,0.99)	.0677	6.49	.5966
1-Methylxanthine	1.08 (0.53,2.20)	.8410	0.91 (0.80,1.04)	.1697	0.90 (0.84,0.97)	.0187	8.04	.8662

Abbreviations: CI, confidence interval; IVW, inverse-variance weighted; MR-PRESSO, MR pleiotropy residual sum and outlier; OR, odds ratio; RSS, residual sum of squares.





**Figure 2.** Genetic associations of uric acid and paraxanthine with neuroticism. (A) Genetic effect of uric acid on neuroticism. (B) Genetic effect of paraxanthine on neuroticism. Each of the single nucleotide polymorphisms (SNPs) associated with metabolite are represented by a black dot with the error bar depicting the SE of its association with metabolite (horizontal) and neuroticism (vertical). The slopes of each line represent the causal association for each method.

**Table 2.** Significant Metabolic Pathways Involved in the Pathogenesis of Neuroticism

Metabolic pathway	Metabolites involved	P value	Database
Alanine, aspartate, and glutamate metabolism	Pyruvate; glutamate	.0041	KEGG
Pantothenate and CoA biosynthesis	Pyruvate; pantothenate	.0052	KEGG
Butanoate metabolism	Pyruvate; glutamate	.0111	KEGG
Glycine, serine, and threonine metabolism	Pyruvate; choline	.0158	KEGG
Caffeine metabolism	Paraxanthine; 1-methylxanthine	.0295	SMPDB
Arginine and proline metabolism	Pyruvate; glutamate	.0385	KEGG
D-Glutamine and D-glutamate metabolism	Glutamate	.0449	KEGG

Abbreviations: KEGG, Kyoto Encyclopedia of Genes and Genomes; SMPDB, Small Molecule Pathway Database.

an inverse association between serum uric acid levels and depression (Bartoli et al., 2018a). The neurotic personality usually incorporated the tendency to experience frequent, intense negative emotions and was strongly associated with psychiatric symptoms. Our study reported that lower serum uric acid levels were associated with higher risk of neuroticism, which supported the findings from previous studies. In addition, rs17246501 (corresponding to the SLC2A9 gene) was a genetic predictor for uric acid, which also showed significant association with neuroticism. Interestingly, a previous cross-sectional analysis reported that serum uric acid and SLC2A9 variant were strongly associated with depressive and anxiety disorders (Lyngdoh et al., 2013). Though further evidence is needed, the identified SNPs might serve as important targets for revealing the pathogenesis and therapies of neuroticism.

Paraxanthine is another metabolite that showed causal association with neuroticism. Notably, paraxanthine also shows close interconnection with adenosine. Indeed, paraxanthine is a non-selective antagonist of adenosine A1 and A2A receptors that play important roles in caffeine metabolism. A previous study has reported that heavy caffeine ingestion might cause or exacerbate anxiety and might be associated with depression and increased use of anti-anxiety drugs. Caffeine might also exacerbate anxiety and panic in panic disorder patients (Clementz and Dailey, 1988). The effect of caffeine on anxiety was also shown in several subsequent studies (Childs and de Wit, 2006; Nardi

et al., 2009; Trapp et al., 2014; Yudko and McNiece, 2014; Botton et al., 2017). Furthermore, coffee and caffeine consumption had also been reported to have antidepressant effects (Yudko and McNiece, 2014; Wang et al., 2016). As a whole, our study showed consistent findings with previous studies, supporting the possible role of the purinergic system in neuroticism-related disorders, such as anxiety and depression (Cheffer et al., 2018; Bartoli et al., 2020).

Our study also reported several metabolic pathways that might be involved in the pathogenesis of neuroticism, such as the alanine, aspartate, and glutamate metabolism pathway. Glutamate is a common excitatory and inhibitory neurotransmitter in the human brain and has been proved to play a major role in the pathogenesis of mental disorders (Hashimoto et al., 2013; Hasler et al., 2019). Our study also suggested that pantothenate and CoA biosynthesis was associated with risk of neuroticism. Pantothenate and CoA biosynthesis was also reported to be associated with risk of major depression and other affective disorders (Chu et al., 2009; Hammerschlag et al., 2017). Although additional evidence is needed, these metabolic pathways might provide valuable information to help understand the underlying biological mechanisms in the pathogenesis of neuroticism.

Our study has several limitations. First, the strength of the instrumental variables relied on the sample size of GWASs, so more data should be collected to improve the veracity of the generated GDMs. Second, though MR proved to be a powerful

approach to assess the causal associations between serum metabolites and neuroticism, the results require further verification based on experimental data. Third, genetic architectures played an essential role in determining the causal relationships between metabolites and neuroticism. However, further work should be done to determine the role of genetic variants in affecting variations of metabolites in neuroticism.

## Conclusions

In conclusion, our study suggested an increased level of uric acid was associated with lower risk of neuroticism, whereas paraxanthine showed the contrary effect. Our MR study also identified multiple metabolic pathways and genetic risk variants that might have causal effects on neuroticism. Our study provided novel insight by combining metabolomics with genomics to help understand the pathogenesis of neuroticism.

## Supplementary Materials

Supplementary data are available at *International Journal of Neuropsychopharmacology (IJNPPY)* online.

Supplementary Table 1. Unknown metabolites identified by the Mendelian randomization estimates.

Supplementary Table 2. Genetic predictors of uric acid and their association with neuroticism.

Supplementary Table 3. Genetic predictors of paraxanthine and their association with neuroticism.

## Acknowledgments

We thank the High-Performance Computing Cluster of the First Affiliated Hospital of Xi'an Jiaotong University for data processing.

This work was supported by the National Natural Science Foundation Award of China (grant no. 81771471).

## Statement of Interest

None.

## Data Availability Statement

Data for GWAS scans with human serum metabolites are freely accessible through the Metabolomics GWAS Server (<http://metabolomics.helmholtz-muenchen.de/gwas/>). GWAS summary statistics for neuroticism can be freely downloaded from the GWAS Catalog website (<https://www.ebi.ac.uk/gwas/>).

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