Original article

The lipidome in major depressive disorder: Shared genetic influence for ether-phosphatidylcholines, a plasma-based phenotype related to inflammation, and disease risk

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ABSTRACT

Background: The lipidome is rapidly garnering interest in the field of psychiatry. Recent studies have implicated lipidomic changes across numerous psychiatric disorders. In particular, there is growing evidence that the concentrations of several classes of lipids are altered in those diagnosed with MDD. However, for lipidomic abnormalities to be considered potential treatment targets for MDD (rather than secondary manifestations of the disease), a shared etiology between lipid concentrations and MDD should be demonstrated.

Methods: In a sample of 567 individuals from 37 extended pedigrees (average size 13.57 people, range = 3–80), we used mass spectrometry lipidomic measures to evaluate the genetic overlap between twenty-three biologically distinct lipid classes and a dimensional scale of MDD.

Results: We found that the lipid class with the largest endophenotype ranking value (ERV, a standardized parametric measure of pleiotropy) were ether-phosphodatidylcholines (alkylphosphatidylcholine, PC(O) and alkenylphosphatidylcholine, PC(P) subclasses). Furthermore, we examined the cluster structure of the twenty-five species within the top-ranked lipid class, and the relationship of those clusters with MDD. This analysis revealed that species containing arachidonic acid generally exhibited the greatest degree of genetic overlap with MDD.

Conclusions: This study is the first to demonstrate a shared genetic etiology between MDD and ether-phosphatidylcholine species containing arachidonic acid, an omega-6 fatty acid that is a precursor to inflammatory mediators, such as prostaglandins. The study highlights the potential utility of the well-characterized linoleic/arachidonic acid inflammation pathway as a diagnostic marker and/or treatment target for MDD.

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1. Introduction

Major Depressive Disorder (MDD) is a common and potentially life-threatening disorder of mood [1]. It affects 16.2% of individuals in the US during their lifetime [2] and as such it incurs great economic cost ($83.1 billion per annum in the US) [3]. This is not to mention the personal cost where the impact of MDD on well being and functioning is in line with that seen in arthritis and diabetes mellitus [4]. Moreover, functional impairments remain after the remission of a depressive episode [5]. Unsurprisingly, the World Health Organization (WHO) cites MDD as a leading cause of disability worldwide [6]. However, despite decades of research, the etiology of the illness remains largely unknown.

Lipidomic alterations have been reported in numerous psychiatric disorders, including schizophrenia [7], autism [8,9], and bipolar disorder [10–12]. In particular, changes in the lipidome (the complete lipid profile of an organism) have been most consistently associated with MDD [13]. The first indication of this association came from early trials of statins, statins are

cholesterol-lowering drugs prescribed to individuals with increased lipid levels [14]. During the statin trials, the lipid-lowering benefits of statin therapy (i.e. reduced cardiovascular disease risk) were offset, in some cases, by an increase in suicidality [15–20]. Though, it should be noted that others have reported beneficial effects of statins on depressive symptomatology when combined with anti-depressant medications including SSRIs [21,22]. The obvious overlap between suicidality and MDD led some to propose a direct link between lipids and MDD. Indeed, subsequent studies have reported differences between depressed and healthy subjects in the concentrations of fatty acids in both animal models of depression [23–27] and also in clinical populations of humans [28–31]; and also alterations in lipid classes including phospholipids (e.g., phosphatidylcholines [PCs], lysophosphatidylcholines [LPCs], lysophosphatidylethanolamine [LPEs], phosphatidylethanolamines [PEs], sphingolipids, and cholesterol esters) [32–35]. However, despite strong evidence linking lipid concentrations and MDD, it is currently unclear whether the lipidomic alterations observed in MDD are secondary to the manifestation of the illness or its treatment, or whether lipid concentrations are related to the genetic predisposition for depression. If the latter supposition were true, lipids could be considered a promising diagnostic and/or treatment target for MDD.

In the present study, we aimed to provide evidence for a shared etiology between lipidomic concentrations and MDD, and determine which lipid classes, and which species within those classes, might be most informative when attempting to isolate potential diagnostic and treatment targets for MDD. To achieve these aims we completed three steps:

- we ranked sum concentrations of twenty-three lipid classes by their genetic overlap with MDD and isolated those classes with the greatest degree of overlap;
- we took the top-ranked lipid classes and investigated the structure of the species within them using cluster analysis;
- we evaluated the degree of genetic overlap between each species cluster and MDD in an attempt to characterize the relationships between the lipids and MDD at the species level.

2. Methods

2.1. Participants

Lipidomic and psychiatric data were available from a total 567 participants from 37 families (average family size = 13.57, range = 3–80) the sample was 64% female and had a mean age of 49.47 years (SD = 13.31, range = 27–97). The lipidomic data was collected as part of the San Antonio Family Study (SAFS), diagnostic data were also available in these same individuals as part of assessments conducted in overlapping individuals as part of the Genetics of Brain Structure and Function (GOBS) study. GOBS data collection occurred between 2006 and 2016. Individuals from the SAFS cohort have actively participated in research for over 18 years. Participants were randomly selected from the community with the constraints that they were of Mexican American ancestry, part of a large family, and lived in the San Antonio, TX, region. All participants provided written informed consent in compliance with the institutional review board at the University of Texas Health Science Center of San Antonio [36].

2.2. Continuous index of MDD

All participants received the Mini-International Neuropsychiatric Interview (MINI) [37], a semi-structured interview augmented to include items on lifetime diagnostic history. Masters- and doctorate-level research staff, with established reliability for diagnosing affective disorders ($\kappa \geq 0.85$), conducted the interviews. All subjects with possible psychopathology were discussed in case conferences that included licensed psychologists or psychiatrists. Lifetime consensus diagnoses were determined based on available medical records, the MINI interview, and the interviewer's narrative. Consistent with previous work [38], all items from the Past Major Depressive Episode (A3a-g) section of the MINI were entered into a confirmatory factor analysis with a single factor, and maximum-likelihood estimates of the latent factor scores were used as the dimensional scale of MDD. In our previous study, we demonstrated that this continuous index conferred multiple advantages for gene-finding efforts over the conventional dichotomous (present-absent) diagnosis of MDD (for details, see [38]). Using conventional diagnoses, 216 individuals endorsed a major depressive episode in their lifetime while 115 had experienced two or more episodes (recurrent MDD).

2.3. Lipid extraction and analysis procedure

The lipid extraction procedure used in this sample has been described in detail elsewhere (see [39,40]). Briefly, the San Antonio Family study is part of an ongoing longitudinal observational investigation comprising four phases of data collection during a 23-year period. The plasma samples used for lipidomic analysis in the present study were collected during the first phase, between the years 1992–1996. The order of the plasma samples was randomized prior to lipid extraction and analysis. Quality control plasma samples were included at a ratio of 1:18. Total lipid extraction from a 10 mL aliquot of plasma was performed by a single phase chloroform:methanol (2:1) extraction after the addition of 15 μL of internal standard mix containing 16 non-physiological or stable isotope lipid standards (Supplementary Table 1) [41].

Lipid analysis was performed by liquid chromatography, electrospray ionisation-tandem mass spectrometry using an Agilent 1200 liquid chromatography system combined with an Applied Biosystems API 4000 Q/TRAP mass spectrometer with a turboionspray source (350 °C) and Analyst 1.5 data system [41]. Liquid chromatography was performed on a Zorbax C18, 1.8 μm, 50 × 2.1 mm column (Agilent Technologies) using the following gradient conditions (300 μL/min) 0% solvent B to 100% solvent B over 8.0 min, 2.5 min at 100% solvent B, a return to 0% solvent B over 0.5 min then 10.5 min at 0% solvent B prior to the next injection. Diacylglycerol (DG) and triacylglycerol (TG) species (1 μL injection) were analyzed in a separate chromatographic run using an isocratic flow (100 μL/min) of 85% solvent B over 6 min. Solvents A and B consisted of tetrahydrofuran:methanol:water in the ratio (30:20:50) and (75:20:5) respectively, both containing 10 mM ammonium formate. Columns were heated to 50 °C and the auto-sampler regulated to 25 °C. All other lipid species (5 μL injection) were separated under gradient conditions.

Multiple reaction monitoring (MRM) experiments were used to analyses lipid species in the following classes and subclasses: dihydroceramide (dhCer), ceramide (Cer), monohexosylceramide (MHC), dihexosylceramide (DHC), trihexosylcermide (THC), GM3 ganglioside (GM3), sphingomyelin (SM), phosphatidylcholine (PC), alkylphosphatidylcholine (PC(O)), alkenylphosphatidylcholine (plasmalogen, PC(P)), lysophosphatidylcholine (LPC), lysoalkyl-phosphatidylcholine (lysoplatelet activating factor, LPC(O)), phosphatidylethanolamine (PE), phosphatidylinositol (PI), phosphatidylserine (PS), phosphatidylglycerol (PG), cholesterol ester (CE), free cholesterol (COH), diacylglycerol (DG) and triaclyglycerol (TG) [41–43]. A total of 65 diacylglycerol and triacylglycerol species and 257 other lipid species were analyzed. The mass spectrometry conditions are shown in Supplementary Table 1. The listed abbreviations are used to refer to individual lipid species e.g.

LPC 22:6, which defines a lysophosphatidylcholine with a fatty acid containing 22 carbons and six double bonds. A number of lipids contain two fatty acid chains, for these the mass spectrometry based measurements reflect the sum of the number of carbons and the sum of the number of double bonds across both fatty acids, rather than directly determining the constituent fatty acids. In accordance with this, for these species we denote the combined length and number of double bonds (e.g. PC 36:4). However, it is of note that the identity of at least the major fatty acids making up such a species in plasma may be reasonably inferred. Relative lipid amounts were calculated by relating the peak area of each species to the peak area of the corresponding stable isotope or non-physiological internal standard. Total lipid classes were calculated from the sum of the individual lipid species within each class [39].

2.4. Quantitative genetic analyses

All genetic analyses were performed in SOLAR [39]. SOLAR implements maximum-likelihood variance decomposition to determine the contributions of genetic and environmental influences to a trait by modeling the covariance among family members as a function of expected allele sharing given the pedigree (see [40] for a detailed description of the variance components methods). The genetic analysis was done at the class levels rather than at the species level in the first instance. We did this because regulation of the lipid metabolic pathway occurs at the class level, and within each class regulation occurs at the level of the fatty acid. Thus by focusing at the class level we hoped to constrain the search space of the lipidome to a set of species and fatty acids in which we could search for associations with MDD. The genetic analyses at the class level were conducted in two steps.

First, univariate polygenic analysis was applied to the individual lipid class sum scores and the MDD index, as part of this step all traits were converted to ranks and were normalized using an inverse Gaussian transformation in addition to being residualized for relevant covariates. Age, age^2, sex and their interactions were included as covariates for all traits while some additional covariates were included only for either the lipid classes or for MDD. For the lipid classes, some combination of the following metabolic covariates, collected at the time of blood sampling as part of the SAFS assessment [41], were included: BMI; antilipid (statin) medication; diabetes status; heart attack; heart surgery; smoking status; hypertension status. Inclusion of the metabolic covariates was dependent on the significance of the covariate with the lipid class in question, a liberal threshold of $P < 0.10$ was applied in order to increase confidence that important covariates were included. For MDD we included any alcohol and any substance use disorder.

Second, bivariate polygenic analysis was applied to each residualized lipid class sum score combined with the residualized MDD index, wherein the phenotypic covariance between the lipid score and MDD was decomposed into its genetic and environmental constituents to determine the extent to which they were influenced by shared genetic effects. Parameter estimates from the bivariate analyses were used to calculate ERVs for each MDD/lipid class pairing.

2.5. ERV calculation

The ERV statistic has been described in detail elsewhere [40], but briefly the ERV for the *i*th lipid class and MDD is given by:

$$ERV_i = \left| \sqrt{h_i^2} \sqrt{h_{MDD}^2} \rho_g \right|$$

where h_i^2 denotes the heritability of the *i*th lipid class, h_{MDD}^2 denotes the heritability of the MDD index, and ρ_g denotes the genetic correlation between the two traits. The ERV is simply an effect size bounded between zero and one, it is useful for prioritizing phenotypes in terms of their shared genetic overlap with a disease of interest. In the present study we ranked lipid classes by their genetic overlap with MDD. After ranking was performed, we tested the statistical significance of the genetic correlation between the top-ranked class and MDD. This approach involved only one null-hypothesis significance test, because we did not test (and indeed, it was never our intention to test) whether each lipid class was associated with MDD or not. Instead, we treated this as a parameter-estimation problem, with the ERV associated with each lipid class as the parameters of interest.

2.6. Cluster analysis of top-ranked lipid class

This set of analysis was done using the lipid species encapsulated by the top-ranked lipid classes revealed by the above analysis step. This species-level analysis was done to more finely investigate the genetic overlap of the top-ranked lipid class and MDD. In order to do this we first applied bivariate polygenic models to all pairs of lipid species and then, using the genetic correlations estimated from these models, created a genetic correlation matrix of all species. Next we applied hierarchical cluster analysis, as implemented in R [42] to the genetic correlation matrix in order to establish clusters of genetically related species. In more detail, the genetic correlation matrix was converted into a matrix of dissimilarity scores by subtracting the absolute value of each correlation from 1. Agglomerative clustering was then applied to this matrix of distance scores. This method of clustering begins with *n* clusters where each cluster represents a single item then, at each step, two clusters are fused together in accordance with the distance values. This analysis was interpreted using a dendrogram plot where similar traits are on the same limb of the tree and distinctly different traits are placed on other limbs [43,44]. Scores for the resultant clusters were derived using principal components analysis where, for each of the clusters all lipid classes were entered into PCA and the first unrotated principal component scored was extracted for bivariate polygenic analysis with MDD.

2.7. Assignment of fatty acids to phosphatidylcholine species

Fatty acid assignments were performed on the quality control pooled plasma sample (*n* = 6 healthy volunteers) used during the lipid analysis for this cohort. The pooled plasma samples were extracted in the same conditions replacing 10 mM ammonium formate with 200 μM lithium acetate in the process. Assignments were made based on the fragmentation patterns of the lithium adducts as described by Hsu et al. [45] using the same chromatography system as described but with 200 μM lithium acetate instead of 10 mM ammonium formate. Scheduled MRMs for the possible fatty acid specific fragments for each phosphatidylcholine species were used over several injections, resulting in qualitative data of possible combinations of fatty acids for each of the species. PC(O) species of very low abundance were not able to be characterized using this approach. Throughout the present manuscript we follow the naming convention of lipid classes and species outlined by the LIPID MAPS consortium [46].

3. Results

3.1. Heritability of MDD

As has been previously reported the dimensional scale of MDD was deemed to be significantly heritable ($h^2 = 0.20$, se = 0.06, $P = 2.6 \times 10^{-5}$) [38].

3.2. ERV: ranking of lipid classes by genetic overlap with MDD

The endophenotype ranking results are presented in Table 1, which includes a list of the metabolic covariates that were included in the analysis of each class. The top-ranked lipid class was PC(O) for which ERV = 0.13 (h^2 = 0.39, se = 0.06, $P = 1.99 \times 10^{-16}$). The second best ranked lipid class using ERV was the PC(P). The genetic correlation between PC(O) and PC(P), which are both phosphatidylcholine lipid classes, was high and significant ($\rho_g = -0.75$, $P = 1.4 \times 10^{-7}$), consequently we elected to sum the two and treat them as a single trait. This sum score of PC(O) and PC(P) was significantly heritable (h^2 = 0.39, se = 0.06, $P = 4.89 \times 10^{-15}$), and the genetic correlation between this score and MDD was significant ($\rho_g = -0.51$, $P = 0.01$). Therefore the lipid classes exhibiting the greatest degree of genetic overlap with MDD were the ether-phosphatidylcholine classes PC(O) and PC(P), a sum score of which shared a significant genetic correlation with MDD.

3.3. Clustering of PC(O) and PC(P) lipid species

In order to identify clusters of genetically related species we applied cluster analysis to the genetic correlation matrix of all ether-phosphatidylcholine species in our top-ranked lipid classes, PC(O) and PC(P), for MDD. Fig. 1 shows the results of the hierarchical cluster analysis applied to the genetic correlation matrix of all lipid species within the PC(O) and PC(P) classes. This analysis revealed three primary clusters, one of which is primarily characterized by those PC(O) and PC(P) species with a relatively lower number of carbon atoms and double bonds, shown in purple, cluster 1. Cluster 2, shown in orange, encapsulates those species with a relatively higher number of carbon atoms and double bonds. Finally, PC(O-40:7), PC(O-36:0), PC(O) 34:0 and PC(O) 36:1 were deemed to be outliers, belonging to neither cluster, given their position on a separate branch of the dendrogram.

3.4. Genetic overlap between clusters 1 and 2 and MDD

Both clusters 1 and 2 were shown to be significantly heritable (cluster 1: h^2 = 0.3684, se = 0.06, $P = 4.68 \times 10^{-15}$; cluster 2: h^2 = 0.3965, se = 0.06, $P = 2.16 \times 10^{-15}$). In order to determine which species might be driving the relationship between MDD and

Table 1
Ordered endophenotype ranking values (ERVs), Heritability estimates, genetic correlations and included covariates for all lipid classes tested against MDD.

Lipid class	h^2	ρ_g	ERV	Metabolic covariates included
PC(O)	0.39	−0.46	0.13	Smoking
PC(P)	0.42	−0.40	0.12	Diabetes; smoking; BMI
SM	0.30	−0.45	0.11	BMI
COH	0.38	−0.39	0.11	BMI
DHC	0.25	−0.43	0.10	None
MHC	0.49	−0.26	0.09	None
PC	0.54	−0.25	0.08	BMI
PE(P)	0.28	−0.34	0.08	Smoking
LPC(O)	0.38	−0.28	0.08	BMI
THC	0.51	−0.21	0.07	BMI
dhCer	0.49	−0.21	0.07	Diabetes; BMI
Cer	0.30	0.20	0.05	Diabetes
PS	0.38	−0.18	0.05	None
GM	0.35	−0.17	0.05	Diabetes; BMI
PE(O)	0.37	−0.14	0.04	None
LPC	0.34	−0.13	0.04	Diabetes; BMI
CE	0.32	−0.12	0.03	Diabetes; BMI
LPE	0.34	−0.09	0.03	Diabetes; BMI
DG	0.31	−0.09	0.02	Diabetes; BMI
TG	0.41	0.05	0.01	Diabetes; BMI
PI	0.40	0.04	0.01	Diabetes; BMI
PE	0.34	−0.03	0.01	Diabetes; BMI
PG	0.40	0.01	0.00	Smoking

the PC(O) and PC(P) lipid classes we applied bivariate polygenic analysis. This analysis revealed that only cluster 2 ($\rho_g = -0.4852$, $P = 0.01$) shared significant genetic overlap with MDD, while the overlap with cluster 1 was not significant ($\rho_g = -0.3015$, $P = 0.1035$).

3.5. Fatty acid assignment of phosphatidylcholine species in cluster 2

Given that cluster 2 exhibited a significant genetic overlap with MDD we performed fatty acid assignments for all ether-phosphatidylcholine species in the cluster. In general, phosphatidylcholine species consist of 3 different classes; diacyl, alkyl and alkenyl, with ether-lipids consisting of the latter two. Our initial experiments only determined the total chain length of the phospholipid (i.e. the sum of carbons and double bonds) as is represented in Fig. 1. The subsequent reanalysis of a pooled plasma sample allowed us to determine the acyl/alkyl and alkenyl chains and their relative abundance. The majority of the species observed in cluster 2 contained either a 20:4 (eicosatetraenoic acid; ETA), a 22:5 (docosapentaenoic acid; DHA) or a 20:5 (eicosapentaenoic acid; EPA) as their sn2 side chain with as 16:0, 18:0 or 18:1 alkyl/alkenyl chain in the sn1 position (Table 2). It is well established that in humans ETA (20:4) exists mainly as the omega-6 fatty acid (or, arachidonic acid) while EPA (20:5) is an omega-3 fatty acid. DHA (22:5) however exists as both forms, with the majority existing as an omega-3 [47]. This means that cluster 2 represents alkyl- and alkenyl phosphatidylcholine species containing omega 6 and omega 3 fatty acids in the sn2 position. While arachidonic acid is represented by only three of the eight species in the cluster, in terms of lipid concentration these species represent approximately 75% of the total lipids within this cluster. Therefore, cluster 2 is mostly characterized by those ether-phosphatidylcholine species containing arachidonic acid.

4. Discussion

The aims of the present study were to provide evidence for shared genetic overlap between lipidomic concentrations and MDD, and to determine which lipid classes, and species, in particular, might be most informative when attempting to isolate potential biomarkers for MDD. Numerous studies have highlighted an association between MDD and the lipidome [32,34,35], indeed it has been previously shown that reductions phosphatidylcholine (and sphingomyelin) concentrations are associated with symptoms of depression [33]. However, previous research has not shown whether the lipid alterations observed in MDD are secondary to the manifestation of the illness or its treatment, or whether lipid concentrations are related to the genetic predisposition for depression. Therefore, the present study extends those findings by showing that: (1) the majority of lipid classes share at least some degree of genetic overlap with MDD; (2) the classes exhibiting the greatest degree of genetic overlap with MDD were phospholipid classes PC(O) and PC(P) which are ether-phosphatidylcholines; and (3) of those top-ranked ether-phosphatidylcholine classes the species which appeared to be driving the genetic overlap with MDD were mostly those containing arachidonic acid. These findings are intriguing because they imply that rather than alterations in phospholipids being secondary to the manifestation of MDD, they might have a shared etiology with the illness, and as such these lipids, their fatty acids, and their molecular pathways, might be fruitful candidates when looking to improve diagnostic and treatment efforts in MDD. Moreover, because the pathways underling arachidonic acid synthesis and metabolism are well characterized, the present study provides an empirically testable set of hypotheses for MDD risk, namely the utility of those genes
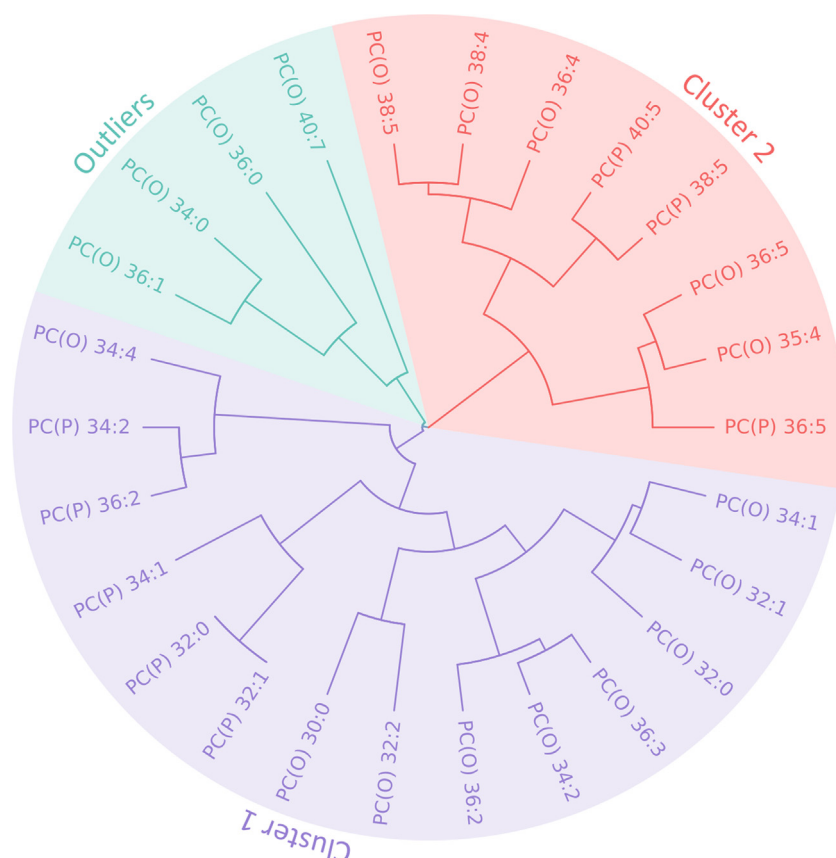
Fig. 1. Dendrogram of the cluster analysis of all lipid species contained in the PC(O) and PC(P) classes. Two main clusters emerged: cluster 1 (purple), and cluster 2 (orange), plus two outliers (green).

Table 2
Fatty acid assignments of the phosphatidylcholine species in cluster 2.

Class	Species	Assignment	Fatty acid
PC(O)	36:4	20:4	Arachidonic acid
PC(O)	38:4		
PC(O)	38:5	>70% 20:4	
PC(P)	38:5	Mix (20:4/22:5/20:5)	Arachidonic/ docosapentaenoic/ eicosapentaenoic acid
PC(P)	40:5	22:6	Docosahexaenoic acid
PC(P)	36:5	20:5	Eicosapentaenoic acid
PC(O)	35:4	Likely 20:4[a]	Arachidonic acid
PC(O)	36:5	Likely 20:5[a]	Eicosapentaenoic acid

[a] No product ions observed in mass spectra due to low abundance.

and proteins encapsulated by arachidonic acid pathways in diagnosing and treating the illness.

Lipids fulfill a plethora of biological functions [48]; they can be stored as forms of energy (as fats and oils), they play a key role in membrane structure and scaffolding, and they may actively influence metabolic traffic via roles in cellular regulation, signaling, and intracellular messaging [49–51]. Lipids make up 50% of the weight of the brain, in fact, the lipid concentration of the brain is second only to adipose tissue [52]. Phospholipids can be broken down into two broad categories, glycerophospholipids (which include phosphatidylcholines) and sphingolipids, both of which are critical in membrane structure [49]. By virtue of their amphiphilic nature these classes of lipids are able to form a semipermeable bilayer around a cell and its contents, which consists of a hydrophobic core of fatty acid tails facing each other and the phospholipid head groups pointing outwards towards the

cell surfaces [49]. Thus, these lipid classes are perfectly placed to modulate signal transduction, molecular recognition processes, and the transportation of ions across the cell membrane [53]. Moreover, the length of the acyl lipid tails affects bilayer width which unsurprisingly influences properties such as ion permeability in addition to the structure and function of membrane proteins [54,55]. Thus ether-phosphatidylcholines, the class of lipids most strongly associated with MDD in the present study, have an established role in cell structure and function in the brain.

The ether-phosphatidylcholine cluster that showed the greatest degree of genetic overlap with MDD is characterized by those species that contain omega 6 and omega 3 fatty acids (arachidonic acid, EPA and DHA), but the majority of fatty acids at the sn2 position of these contains an arachidonic acid. In addition, ERV estimates for the individual species in cluster 2 show that the species sharing the greatest genetic overlap with MDD is one containing arachidonic acid, species PC(O) 38:4 (Table S2). Arachidonic acid is an omega-6 fatty acid that is a precursor to a number of eicosanoids (e.g. prostaglandins), which are crucial for the progression and resolution of inflammatory responses [56]. Inflammation has been linked to the onset of many diseases including, for example, diabetes [57], heart disease [58] and cancer [59]. A number of meta-analyses have implicated the role of inflammation, and specifically pro-inflammatory cytokines (e.g. IL-6, IL-1β, TNF-α, and CRP), in the etiology of MDD [60–65]. Cytokines feature upstream in the immune response to phosphatidylcholines and arachidonic acid. Specifically, eicosanoid production may be triggered when a cell is activated via the release of cytokines, this in turn triggers the release of a phospholipase (e.g. cytosolic phospholipase A2; cPLA2) at the cell

membrane, which liberates arachidonic acid from the cell membrane phospholipid, rendering the fatty acid available for eicosanoid production via cyclooxygenase-2 (COX-2) (Fig. S1). Thus, the association of AA containing PC(O) and PC(P) species with MDD may relate to an underlying chronic inflammation as suggested by the previous literature on cytokines and depression, and potentially highlights a downstream event underlying the relationship between inflammation, cytokines and MDD, namely the release of arachidonic acid from the cell membrane and subsequent eicosanoid synthesis.

Much attention has been paid to the relationship between dietary intake of omega-3 fatty acids and depressive symptoms [65–67], and to a lesser extent with a focus specifically on omega-6 fatty acids [68]. For omega-3 fatty acids the results have been largely positive, although some controversy remains regarding the clinical subgroup for which omega-3 fatty acids are most beneficial (i.e. sub-clinical versus severe) [69–71]. Moreover, two meta-analyses suggest that EPA, as opposed to an alternative fatty acid DHA, which ameliorates depressive symptoms [72,73]. As highlighted by Kiecolt-Glaser and colleagues in their recent review [65] this is consistent with the greater anti-inflammatory properties of EPA. Arachidonic acid is synthesized from dietary intake of linoleic acid (Fig. S1) [74]. Our findings suggest that arachidonic acid shares a genetic overlap with MDD, which might seem contradictory as dietary intake of fatty acids is an environmental factor. However, there might exist a gene-environment interaction. Or, alterations in the linoleic/arachidonic acid pathway might disrupt the downstream metabolism of arachidonic acid. Such alterations might be revealed by a focused search of genetic variation within those pathways.

The findings of the present manuscript rely on a peripheral index of lipid levels in the form of extractions performed on plasma samples. This allows us only to speculate on the ways in which these findings might be interpreted in the brain. Phosphorous-31 magnetic resonance spectroscopic (31P MRS) imaging is a method that allows non-invasive measurement of biological compounds (e.g. phospholipids) in vivo. Alterations in peripheral lipids, including in phosphatidylcholines, have been noted in psychiatric disorders other than MDD, including in schizophrenia and bipolar disorder [75,76]. Studies employing MRS techniques have documented brain-based alterations in membrane phospholipid characteristics in schizophrenia [77], bipolar disorder [78], and in MDD [79,80]. Future work might follow up on the findings in the present manuscript using similar methods.

It is possible that lipidomic abnormalities in relation to affective disorders may be characterized differently in other ethnic populations. For example, non-Hispanic populations exhibit altered lipidomic profiles and associated risk for myocardial infarction relative to Hispanics [81]. Thus it is important that the generalisability of the findings in the present manuscript should be further tested in future research.

In summary, the findings presented here highlight ether-phosphatidylcholines, and in particular those species containing arachidonic acid, as having a sizeable genetic overlap with MDD. While it has been previously demonstrated that those with MDD exhibit altered levels of phospholipids, and also arachidonic acid, this is the first study to highlight a shared genetic etiology between the two. When taken within the context of previous research demonstrating the role of phospholipids and their fatty acids (and in particular arachidonic acid) in inflammation, and the wealth of literature linking inflammation and MDD; the present study, at the very least, highlights the potential utility of ether-phosphatidyl-cholines and their biochemical pathways as potentially interesting avenues of research for MDD. Going further than that, the findings of the present study generate a tentative but testable hypothesis, which is that the well-characterized linoleic/arachidonic acid

inflammation pathway is a potential diagnostic marker and/or treatment target for MDD.

Disclosure of interest

The authors declare that they have no competing interest.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.eurpsy.2017.02.479.

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