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Phospholipase A2 and Cyclooxygenase 2 Genes Influence the Risk of Interferon- α -Induced Depression by Regulating Polyunsaturated Fatty Acids Levels

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Abstract

Background—Phospholipase A2 (PLA2) and cyclooxygenase 2 (COX2) are the two key enzymes in the metabolism of polyunsaturated fatty acids, which in turn play an important role in cytokine-induced depression and sickness behavior.

Methods—Patients with chronic hepatitis C viral infection ($n = 132$) were assessed to examine the effects of seven single nucleotide polymorphisms in COX2 and PLA2 genes on the development of depression during interferon (IFN)- α treatment; a subsample ($n = 63$) was assessed for the erythrocyte levels of the three main polyunsaturated fatty acids, docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), and arachidonic acid. An independent “replication” sample of patients with major depression unrelated to cytokine treatment ($n = 82$) was also examined.

Results—Twenty-eight percent of participants developed INF- α -induced depression. Participants with the PLA2 BanI GG or the COX2 rs4648308 AG genotypes had a higher risk of IFN- α -induced depression (odds ratio = 3.1 and 3.5, respectively). The “at risk” PLA2 genotype was associated with lower EPA levels, and the “at risk” COX2 genotype was associated with lower DHA levels, during IFN- α treatment. The PLA2 BanI GG polymorphism was also associated with more somatic symptoms of depression, both in patients with INF- α -induced depression and in the replication sample of patients with major depression.

Conclusions—Genetic variations in the COX2 and PLA2 genes increase the risk of IFN- α -induced depression, possibly by affecting the levels of EPA and DHA. Moreover, PLA2 genotype is associated with somatic symptoms in depression. Our study confirms the role of inflammatory mechanisms in major depression.

Keywords

Cyclooxygenase 2 (COX-2); cytokine; depression; immune; inflammation; interleukin (IL)-6; phospholipase A2 (PLA2); polyunsaturated fatty acids (PUFAs); prostaglandins; psychoneuroimmunology

There is a great interest in psychiatric symptoms induced by the cytokine interferon- α (IFN- α) in patients with chronic hepatitis C virus (HCV) infection: first, HCV infection is a major public health concern, and psychiatric symptoms can affect the clinical outcome (1); and, second, inflammation has been proposed as one of the molecular mechanisms underlying major depression, and these patients are an ideal clinical sample to examine such a mechanism (2).

Interferon- α is the only effective treatment for chronic HCV infection. Unfortunately, this cytokine treatment is associated with severe psychiatric symptoms, including depression, fatigue, anxiety and irritability (3). In turn, these symptoms result in poor compliance or discontinuation of IFN- α treatment and in poor viral response (4,5). Because of the similarity between IFN- α -induced psychiatric symptoms and the symptoms of major depression, especially the somatic symptoms (6), this model has been used to study the role of inflammation in depression (7,8) and in particular in the mechanisms leading to somatic symptoms (9,10).

Several candidate genes have been examined to identify at baseline, before starting IFN- α , those who will develop depression. For example, we have recently found that the “G” allele of a functional G > C single nucleotide polymorphism (SNP) (rs1800795) in the promoter region of the interleukin (IL)-6 gene—associated with higher plasma concentrations of IL-6 during immune activation (11)—increases the risk of IFN- α -induced depression (12). Moreover, we have found that the “s” allele of a functional insertion/deletion polymorphism (SLC6A4; 5-HTTLPR) in the promoter region of the serotonin transporter (5HTT) gene—a “risk allele” for depression (13)—also increases the risk of IFN- α -induced depression (12). This latter finding has been replicated by a second (14) but not a third study (15). The 5/5 or 5/14 genotype of a GT repeat dinucleotide microsatellite in the IFN- α receptor (16) and the 1019 polymorphism in the serotonergic receptor 1A (5HT1A) (15) have also been shown to modulate the risk of developing IFN- α -induced depression. Interestingly, the link between serotonergic genes and IFN- α -induced depression is consistent with biomarker studies showing that depression in these patients is driven, at least in part, by changes in tryptophan and serotonin metabolism (17,18). Clearly, other genetic markers in biological systems relevant to inflammation can also be important for IFN- α -induced depression.

The n-3 (or omega-3) polyunsaturated fatty acids (PUFAs) seem to protect against major depression (19,20) as well as against cytokine-induced behavioral changes in animals (also known as “sickness behavior”) (21). For example, major depression is associated with low levels of n-3 PUFAs in peripheral plasma erythrocytes (22-24) and in postmortem brain (25). In addition, n-3 PUFAs have been found to have antidepressant effects in some although not all placebo-controlled trials (26,27). Moreover, we (28) and others (29) have reported that n-3 PUFAs protect against the development of depressive-like behavior induced by the forced swimming test in animals. Finally, high n-3 PUFAs in the diet have been reported to reduce brain prostaglandin E₂ (PGE₂) levels and to attenuate cytokine-induced sickness behavior in animals (21,30,31).

Phospholipase A2 (PLA2) and cyclooxygenase-2 (COX2) are the two key enzymes of the PUFA metabolism and PGE₂ synthesis. These two genes are arranged in a head-to-head

configuration in chromosome 1q25 and hence possibly share a common regulatory region (32). In 1998, Peet *et al.* (33) found that the G variant of the BanI SNP in the PLA2 locus is associated with schizophrenia. In another independent population, Wei and Hemmings (32) systematically analyzed six SNPs in the COX2 and PLA2 loci and again found that the G allele of the PLA2 BanI polymorphism is associated with schizophrenia. Pae *et al.* (34) also found that this same G allele of the PLA2 BanI polymorphism increases the risk of developing depression, in a Korean population. To our knowledge, there are no studies examining the association between SNPs in the COX2 gene and depression.

On the basis of this evidence, our aim was to investigate whether polymorphisms in PLA2 and COX2 genes have an effect on the occurrence of IFN- α -induced depression in patients taking IFN- α . Moreover, we wanted to test whether these putative genetic effects are mediated by changes in the levels of the three main PUFAs, docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), and arachidonic acid (AA). An independent replication sample of patients with major depression was also examined, to examine the effects of genotypes on specific clusters of depressive symptoms. Finally, a secondary aim of this study was to replicate, in this Chinese sample, our previous study examining IL-6 and 5-HTTLPR SNPs in a Caucasian population (see preceding text) (12).

Methods and Materials

Subjects

The study was approved by the Institutional Review Board of the China Medical University Hospital in Taichung, Taiwan. Subjects with chronic HCV infection were recruited between July 2005 and June 2007, at the Hospital Liver Centre. The eligible patients were scheduled to receive peg-IFN- α -2beta (1.5 μ g/kg of body weight, subcutaneously, once weekly) and daily ribavirin (800–1200 mg) for at least 24 weeks. Of the original sample ($n = 149$), 3 had major depression at baseline and were excluded, 11 withdrew consent or were lost to follow-up, and 3 did not provide blood samples. The final sample included 132 patients (82 men and 50 women; mean age $\pm$ SD = 49 ± 12 years) who provided blood samples for genetic analysis and completed the follow-up psychiatric assessments during IFN- α treatment. Among them, 63 had baseline and follow-up blood samples for PUFAs analysis. At baseline, none of the enrolled subjects had depression or were receiving antidepressants, antipsychotics, or mood stabilizers. We also excluded patients with problems of substance abuse, including excessive consumption of alcohol and nicotine dependence. If the patients developed an episode of IFN- α -induced depression, the antidepressant treatment was prescribed by the treating physicians according to their clinical judgment and was not controlled by our study protocol.

The independent replication sample ($n = 82$) consisted of medication-free patients with major depression (21 men and 61 women), collected at the psychiatric outpatients clinic of the China Medical University Hospital. The mean age of this group was 37 ± 12 years, and the mean total Hamilton Depression Rating Scale (HDRS) score was 28 ± 6 .

Experimental Design

This was a 24-week prospective cohort study. Patients had to fully understand and then sign the informed consent before enrollment. Patients were evaluated at baseline (week 0) and after 2, 4, 8, 12, and 24 weeks of IFN- α treatment. We have previously used this design successfully in similar populations (35–37). At each study visit, patients were assessed with the structured Mini-International Neuropsychiatric Interview (38) to ascertain the diagnosis of a major depressive episode and with the 21-item Beck Depression Inventory (BDI) to monitor depressive symptoms. For HCV patient that developed IFN- α -induced depression,

the semi-structured version of the 21-item HDRS was used to assess the severity and the different clusters of depressive symptoms. The same HDRS was also used in the 82 patients of the replication sample with major depression. The HDRS items were divided into the following published clusters (39): Core (items 1, 2, 7, 8, 10, 13), Sleep (items 4, 5, 6), Activity (items 7, 8), Anxiety (items 9, 10, 11, 12, 13), Psychic Anxiety (items 9, 10), Somatic Anxiety (items 11, 12, 13), and Delusion (items 2, 15, 20).

Laboratory Methods and Choice of Markers

The Genomic DNA was extracted from peripheral blood with DNA extraction kits (Qiagen, Hilden, Germany). Three cPLA2 markers (rs10798059 [BanI], rs4651330, rs3736741) and three COX2 markers (rs4648308, rs2745557, rs689466), which had been identified and published when we designed our study (32,40), were chosen for genotyping. These six SNPs in the PLA2/COX2 loci are not in linkage disequilibrium (32,40). In addition, the rs20417 (COX2 – 765G > C) marker was genotyped (41), together with one functional polymorphism in the IL-6 gene, (rs1800795) and one functional polymorphism in the 5HTT gene (5-HTTLPR) (12). We performed the Hardy Weinberg equilibrium (HWE) test separately in cases and control subjects; the only polymorphism that violated the HWE test was the COX2 rs4648308 in cases.

Fatty acid composition of erythrocyte membranes was analyzed by thin-layer chromatography, and the level of individual fatty acid was measured with gas chromatography of methyl esters (Lipid Standards, FAMES, Sigma, St. Louis, Missouri). The detailed step-by-step procedures have been published and described elsewhere (42-44). The levels of PUFAs were presented as percentage of total fatty acids.

Statistic Analysis

Patients who developed IFN- α -induced depression during the treatment were defined as “cases,” whereas those who never developed depression were defined as “control subjects.” The independent-samples *t* test was used to compare means of continuous variables between case and control subjects. Changes in PUFA levels as a function of time were assessed with repeated-measures analysis of variance with mixed linear modeling. All comparisons of genotype and allele frequencies were conducted with the χ^2 test, with Yates' correction when appropriate. Odds ratios (OR) were also calculated, along with 95% confidence interval, to measure the genetic risk of developing IFN- α -induced depression. The *p* values < .05 were considered statistically significant. Data are reported as mean $\pm$ SD, whereas the error bars in Figure 1 represent SEM.

Results

Of the total sample (*n* = 132), 37 patients (28%) developed an IFN- α -induced major depressive episode (“cases”) at some point during the 24-week treatment, whereas 95 (72%) patients did not develop IFN- α -induced depression (“control subjects”). Table 1 presents sociodemographic and clinical data for cases and control subjects. There were no significant differences in age, gender distribution, education, and the mean BDI scores at baseline. Five subjects had a previous history of major depression and were all in the “cases” group. Of the 37 subjects who developed depression, 20 cases (54%) received antidepressants after the diagnosis.

Effects of Genotypes on IFN- α -induced Depression

Table 2 describes the effects of allele types and genotypes on the development of IFN- α -induced depression. Both the G allele (OR = 1.8) and the GG genotype (OR = 3.5) in the PLA2 BanI polymorphism significantly increased the risk of developing IFN- α -induced

depression. Moreover, both the A allele (OR = 2.3) and the AG genotype (OR = 3.1) in the COX2 rs4648308 polymorphism significantly increased the risk of developing IFN- α -induced depression (there were no subjects with the COX2 rs4648308 AA genotype). Excluding the five subjects with a previous history of major depression did not affect the findings (data not shown).

Interestingly, these genetic effects were not simply due to the different genotypes being associated with more depressive symptoms at baseline (before starting IFN- α), because there were no differences between the genotypes in the baseline BDI scores: for the PLA2 BanI, 6.9 ± 10.6 in the “at risk” GG genotype versus 4.0 ± 6.2 in the AA/AG genotype ($p = .3$); for the COX2 rs4648308, 4.5 ± 6.4 in the “at risk” AG genotype versus 4.2 ± 6.7 in the GG genotype ($p = .8$). However, a contribution of the baseline depressive symptoms to these genetic effects cannot be completely excluded, because there was no difference between the groups in the “delta” increase from baseline BDI to the maximum BDI scores during the treatment (data not shown).

There were no significant effects of other polymorphisms on the development of IFN- α -induced depression. All subjects had the IL-6 (rs1800795) GG genotype, and therefore no further analyses were conducted on this gene.

Effects of the PLA2 BanI Polymorphism on Specific Clusters of Depressive Symptoms

We investigated the effects of the PLA2 BanI and the COX2 rs4648308 polymorphisms—the genotypes shown to influence the risk of IFN- α -induced depression—on the HDRS clusters of symptoms at the time of the diagnosis of IFN- α -induced depression. Patients with the PLA2 BanI GG genotype (the “at risk” genotype) developed significantly more somatic symptoms than the grouped AG/AA genotypes ($p = .002$) (Table 3), whereas there were no differences between the genotype groups in the total HDRS score or in the scores of the other symptom clusters. There was no effect of the COX2 rs4648308 genotype on any of the symptom clusters (data not shown).

We further examined the effects of the PLA2 BanI GG genotype on somatic symptoms in the replication sample of patients with major depression. The frequencies of alleles and genotypes of the PLA2 BanI polymorphism in this group were 71% for A and 29% for G; 51% for AA, 39% for AG, and 10% for GG. Again, the BanI polymorphism had a significant effect on the somatic cluster ($p = .01$) but not on the total HDRS score or on the scores of the other symptom clusters (Table 3).

Effects of PLA2 and COX2 Genotypes on PUFA Levels

A subgroup of 63 patients had at least one baseline and one follow-up sample for measurement of PUFA levels. Of these 63, 21 (33%) patients developed IFN- α -induced depression at some point during the 24-week treatment (“cases”), and 42 (67%) patients did not (control subjects). Of the 21 who developed depression, 13 (62%) received antidepressants after they developed depression.

There were no significant differences between cases and control subjects in sociodemographic characteristics, including age, education, and the mean BDI scores at baseline (Table 4). In this subsample, however, the occurrence of IFN- α -induced depression was significantly higher in women than in men. Two subjects had a previous history of major depression, both in the “cases” group. Interestingly, cases had lower levels of DHA ($p = .024$) but not of EPA or AA at baseline (i.e., before starting IFN- α treatment).

We examined the effects of the PLA2 BanI and the COX2 rs4648308 polymorphisms—the two genotypes shown to influence the risk of IFN- α -induced depression—on PUFA levels at baseline and during IFN- α treatment.

At baseline, subjects who had the PLA2 BanI GG genotype (the “at risk” genotype) had lower EPA levels than those who had the AG/AA genotype ($p = .013$) (Figure 1A, week 0), but there were no differences in DHA ($3.33 \pm .25\%$ vs. $3.33 \pm .30\%$, $p = 1.0$) or AA ($5.58 \pm .16\%$ vs. $5.49 \pm .28\%$, $p = .4$) levels. During IFN- α treatment, subjects with PLA2 BanI GG also had significantly lower EPA levels [mixed model analysis for repeated measures; $F(52) = 8.9$, $p = .004$] (Figure 1A), but again there were no differences in DHA [$F(77) = .5$, $p = .5$] or AA [$F(130) = .2$, $p = .6$] levels (data not shown). However, the difference in EPA levels during IFN- α treatment was not significant after adjusting for the baseline difference [$F(7) = 5.5$, $p = .053$] or for the presence of antidepressant treatment after the development of depression [$F(62) = .6$, $p = .5$]. Interestingly, there was also no main effect of depression on EPA levels during IFN- α ($F = 1.6$, $p = .2$).

At baseline, subjects who had the COX2 rs4648308 AG genotype (the “at risk” genotype) had lower DHA levels than those who had the GG genotype ($p = .049$) (Figure 1B, week 0), but there were no differences in EPA ($.81 \pm .03\%$ vs. $.82 \pm .05\%$, $p = .4$) or AA ($5.44 \pm .26$ vs. $5.54 \pm .26$, $p = .1$) levels. During IFN- α treatment, subjects with AG genotype also had significantly lower DHA levels [mixed model analysis for repeated measures; $F(95) = 13.4$, $p < 0.001$] (Figure 1B), but again there were no differences in EPA [$F(66) = 3.3$, $p = .08$] or AA [$F(142) = .1$, $p = .7$] levels (data not shown). The difference in DHA levels during IFN- α treatment remained significant after adjusting for the baseline difference [$F(101) = 13.1$, $p < .001$] or for the presence of antidepressant treatment after the development of depression [$F(101) = 14.6$, $p < 0.001$]. Moreover, there was a main effect of depression, with DHA levels being lower in cases than in control subjects across the genotypes, even after adjusting for the baseline difference ($F = 4.7$, $p = .033$).

Discussion

We find that the PLA2 BanI and the COX2 rs4648308 polymorphisms influence the risk of developing IFN- α -induced depression. Moreover, the PLA2 BanI polymorphism is also associated with higher severity of somatic symptoms of depression (but not of other symptoms), in patients with IFN- α -induced depression and in a replication sample of patients with major depression unrelated to cytokine treatment. Finally, these genetic effects seem to be mediated by the regulation of EPA and DHA levels.

The fact that PLA2 BanI polymorphism influenced the risk of IFN- α -induced depression is consistent with previous evidence. The G variant in the BanI polymorphism—the same genotype that increases the risk of IFN- α -induced depression in the present study—has been previously reported to be associated with depression (34). Moreover, the BanI GG genotype has been reported to be associated with higher PLA2 enzyme activity in platelets, suggesting that this polymorphism is functional (45). In our study, we have evaluated PUFA levels rather than PLA2 enzyme activity, but the results further support the functional significance of this BanI polymorphism. In fact, subjects who had the PLA2 BanI “at risk” GG genotype had lower EPA levels before IFN- α treatment. Because the PLA2 enzyme deacylates EPA from membrane phospholipids (46,47), the lower EPA levels in those with the GG genotype could be explained by the higher PLA2 enzyme activity. In turn, the lower EPA levels could explain the higher risk of developing IFN- α -induced depression: in several case-control studies, lower EPA levels are associated with increased risk of depression and with increased severity of depressive symptoms (48,49).

The PLA2 BanI GG genotype was also associated with more somatic symptoms of depression, both among those who developed IFN- α -induced depression and in a replication sample of patients with major depression. These shared genetic effects across the two samples are consistent with previous reports showing that patients with IFN- α -induced depression have similar neuroendocrine and neuroimaging abnormalities as those described in patients with major depression unrelated to cytokine treatment (50–52). The lower EPA levels found in our subjects with the PLA2 BanI GG genotype are a possible explanation for this high risk of specifically developing somatic symptoms. In fact, somatic symptoms of depression are similar to symptoms of “sickness behavior” induced by cytokine administration (6) and are observed in up to 80% of patients with major depression (53). Indeed, somatic symptoms have been described by Dantzer (54) as “the outward manifestation of sensitization of the brain cytokine system that is normally activated in response to activation of the innate immune system and mediates the subjective, behavioral, and physiological components of sickness.”

The other main finding of this study is that the COX2 rs4648308 polymorphism is also associated with IFN- α -induced depression. Specifically, patients with the AG genotype had a higher risk of developing depression during IFN- α treatment. In addition, the “at risk” AG genotype was also associated with lower DHA levels before and during IFN- α treatment, implying a functional significance of this polymorphism. Indeed, lower baseline DHA levels were a risk factor for the development of depression independently from the genotype, further supporting the notion that lower baseline DHA levels are indeed a risk factor for the development of depression. This is consistent with studies showing lower DHA levels in patients with major depression (23,49,55,56).

Subjects with lower PUFA levels before starting IFN- α could be at an increased risk of developing depression by having lower PUFA levels or failing to mount a compensatory PUFA increase during the immune challenge or both. As shown in Figure 1, erythrocyte PUFA levels vary during the 24 weeks of IFN- α , with an initial decrease during the first 2 weeks and an increase in the following weeks. A possible explanation for the initial decrease in PUFA levels is that IFN- α increases the activation of PLA2, leading to the release of PUFAs from the erythrocyte membrane phospholipids (57–58). This initial decrease in membrane PUFAs might then result in an increase in free-form PUFAs, which in turn could exert an “inhibitory feedback” and thus attenuate the IFN- α -induced PLA2 activity, leading to the increase in the erythrocyte PUFA levels in the later weeks of treatment (59).

Higher depressive symptoms at baseline (before starting IFN- α) are considered one of the strongest factors predicting the IFN- α -induced depression (3). However, for both PLA2 and COX2 genes, there were no differences in the baseline BDI scores between the “at risk” and the “low risk” genotypes. This indicates that the main effect of these “immune” genes is to increase the risk of developing depression specifically in response to IFN- α , probably by modulating PUFA levels during the treatment. This is consistent with our previous findings showing that the effects of IL-6 gene on IFN- α -induced depression are also independent from baseline depression scores (12).

We also attempted to replicate, in this Chinese sample, our previously published findings showing an effect of the IL-6 and 5-HTT genes on IFN- α -induced depression in a Caucasian population (12). Specifically, we have previously found that the IL-6 “G” and the 5-HTTLPR “s” genotypes increase the risk of IFN- α -induced depression, compared with the “CC” and the “ll” genotypes, respectively (12). However, there were only IL-6 GG subjects in the present Chinese sample, and thus we could not test the “at risk” effects of this allele. The lack of C genotype in a Chinese sample is a novel finding, although it is consistent with recent HapMap data showing a G allele frequency of 100% in Asians (60).

Interestingly, we also did not find an effect of the 5-HTTLPR “s” polymorphisms in the present study. In this regard, it is important to emphasize that the effects of the 5-HTTLPR genotype in our previous study are only present in those with the IL-6 CC genotype (i.e., in those with the lower immune activation genotype) (12). Therefore, our negative findings in this Chinese sample (all GG) are actually replicating our previous negative findings in Caucasian GG subjects. Interestingly, the “at risk” effects of the 5-HTTLPR genotype have been replicated by a second (14) but not a third study (15), thus supporting the notion that the effects of 5-HTTLPR genotype on IFN- α -induced depression might be dependent on other genes and on the inflammatory status. We did not assess, in this or in our previous study, any other SNPs in the 5HTT gene. Interestingly, Lotrich *et al.* (14) also examined a polymorphism within the “l” allele that might result in lower transcription efficiency, functionally comparable with the “s” allele, as well as a variable number of tandem repeats polymorphism in the second intron. They found that the polymorphism within the “l” allele also influences the risk of developing depression, whereas the variable number of tandem repeats polymorphism has no effect (14).

To our knowledge, this is the largest published study examining genetic predictors of IFN- α -induced depression, the only one assessing genes as well as biomarkers in the same sample, and the first one examining biological markers of IFN- α -induced depression in Han Chinese people. Moreover, our findings are strengthened by the fact that all subjects were antidepressants-free before the development of IFN- α -induced depression. Nevertheless, there are a few important methodological considerations and study limitations. The selection of the polymorphisms from PLA2 and COX2 genes was not completed systematically, because of the lack of comprehensive bioinformatic tools when this project started in 2005. Moreover, the distribution of the COX2 rs4648308 in the case group violated HWE, although some authors have argued that this is acceptable, because “cases” in case-control studies cannot be considered “general population” (61). Furthermore, we did not adjust for the number of statistical comparisons. Many genetic studies, especially those where the choice of genes is hypothesis-driven (like ours), do not apply such adjustment. For example, none of the previous studies investigating genetic predictors of IFN- α -induced depression adjusted for the number of statistical comparisons (12,14–16). Finally, only a subset (48%) of the genetic sample was available for the analysis of PUFA levels. Clearly, our findings will need to be replicated independently.

In conclusion, we describe two polymorphisms in genes relevant for PUFA metabolism that increase the risk of IFN- α -induced depression, together with the putative molecular mechanism (lower EPA and DHA levels) explaining these genetic effects. Moreover, we find a regulatory effect of PLA2 gene on somatic symptoms in major depression unrelated to cytokine treatment. Because n-3 PUFAs have been found to be a safe and effective treatment for depression, these findings provide the rationale to conduct a clinical trial to test the prophylactic effects of treatment with n-3 PUFAs for IFN- α -induced depression. Furthermore, these results confirm the role of inflammatory mechanisms in the pathogenesis of major depression.

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Figure 1.

The association between phospholipase A2 (PLA2) BanI and eicosapentaenoic acid (EPA) levels and between cyclooxygenase 2 (COX2) rs4648308 and docosahexaenoic acid (DHA) levels, during interferon (IFN)- α treatment. **(A)** EPA levels in patients with GG and AG/AA BanI PLA2 genotypes during IFN- α treatment. The mixed model analyses showed significant effects on groups [$F(52) = 8.9, p = .004$] and time [$F(203) = 15.2, p < .001$] but no group $\times$ time interaction [$F(203) = 1.5, p = .2$]. **(B)** DHA levels in patients with AG and GG rs4648308 COX2 genotypes during IFN- α treatment. The mixed model analyses showed significant effects on groups [$F(95) = 13.4, p < .001$], time [$F(97) = 2.6, p = .028$], and group by time interaction [$F(97) = 3.5, p = .006$].

Table 1

Characteristics of 132 Study Participants Available for Genotyping Analysis

	Depressed	Nondepressed	<i>p</i>
<i>N</i>	37 (28%)	95 (72%)	—
Age, yrs (mean $\pm$ SD)	51 $\pm$ 11	48 $\pm$ 12	.2
Gender (Male/Female, <i>n</i>)	21/16	61/34	.4 ^a
Education (yrs)	8 $\pm$ 3	9 $\pm$ 5	.1
Marriage, <i>n</i>			
Married	28	74	.8 ^a
Single/divorced/widow or widower	9	21	.8 ^a
Baseline BDI scores (mean $\pm$ SD)	6 $\pm$ 7	4 $\pm$ 6	.2

The *p* values from independent-samples *t* test, unless χ^2 test result.

BDI, Beck Depression Inventory.

^a χ^2 test result.

Table 2
Allele and Genotype Frequencies and ORs of IFN- α -Induced Depression on the Basis of Allele and Genotyping Groups

SNP	Frequency (%)				ORs	95% (L) ^a	95% (U) ^a	<i>p</i> ^b	
	Cases	Control	Subjects						
Alleles									
rs10798059 (BamI)	G	38	25	1.8	1.0		3.2	.043	
rs4651330	C	19	21	.9	.5		1.8	.8	
rs3736741	G	19	16	1.2	.6		2.4	.6	
rs4648308	A	28	15	2.3	1.2		4.4	.010	
rs2745557	A	5	4	1.5	.4		5.3	.8	
rs689466	G	43	51	.8	.4		1.3	.3	
COX-2-765G > C	G	97	95	1.8	.4		8.5	.7	
5HTTLPR	S	75	76	1.0	.5		1.9	1.0	
Genotypes									
	GG	19	6	3.5	1.1		11.1	.029	
rs10798059 (BamI)	AA	38	38	.6	.3		1.3	.2	
	AG	43	56	1.0	.5		2.2	1.0	
	CC	8	3	2.7	.5		14.1	.4	
rs4651330	AA	22	35	1.4	.6		3.3	.4	
	AC	70	62	.5	.2		1.3	.1	
	GG	0	3	×	×		×	.7	
rs3736741	AA	38	26	.7	.3		1.6	.4	
	AG	62	71	1.7	.7		3.9	.2	
rs4648308	AG	57	29	3.1	1.4		6.9	.004	
rs2745557	AG	11	7	1.5	.4		5.6	.8	
	GG	19	24	.7	.3		1.9	.5	
rs689466	AA	49	53	1.6	.7		3.7	.3	
	AG	32	23	.9	.4		1.8	.7	
COX-2-765G > C	GG	95	92	1.6	.3		8.0	.8	
	CC	0	1	×	×		×	.6	
	CG	5	7	.7	.1		3.6	1.0	

SNP	Frequency (%)					ORs	95% (L) ^a		95% (U) ^a		<i>p</i> ^b
	Cases	Control	Subjects								
5HTTLPR	SS	54		58	.9	.4	1.8	.7			
	LL	43		36	.4	.1	3.6	.4			
	LS	3		6	1.4	.6	3.0	.5			

OR, odds ratio; INF, interferon; COX-2, cyclooxygenase 2; 5HTTLPR, serotonin-transporter-linked polymorphic region.

^aUpper (U) and lower (L) 95% confidence interval.

^bYates's correction as appropriate.

Table 3

The Effect of PLA2 BanI Genotypes on the HDRS Symptom Clusters in Groups of Patients with IFN- α -Induced Depression and Patients with Major Depressive Disorder

HDRS Clusters Scores ^a	IFN- α -Induced Depression			Major Depressive Disorder		
	GG	AG/AA	p	GG	AG/AA	p
Core	11.1 $\pm$ 1.3	10.7 $\pm$ 2.2	.6	15.6 $\pm$ 2.8	15.1 $\pm$ 2.3	.5
Sleep	4.0 $\pm$ 1.3	3.7 $\pm$ 1.9	.7	3.4 $\pm$ 2.4	3.5 $\pm$ 1.6	.8
Activity	3.1 $\pm$ 1.5	2.9 $\pm$ .7	.7	3.9 $\pm$ 1.6	3.8 $\pm$ 1.4	.8
Psychic Anxiety	2.0 $\pm$ 1.3	2.0 $\pm$ 1.0	.8	4.1 $\pm$ 1.6	4.5 $\pm$ 1.6	.5
Somatic Anxiety	5.9 $\pm$ 1.1	4.2 $\pm$ 1.2	.002 ^b	5.8 $\pm$ .7	4.5 $\pm$ 1.7	.01
Delusion	2.1 $\pm$ .9	2.0 $\pm$ 1.2	.8	4.8 $\pm$ 2.3	3.8 $\pm$ 1.8	.2
Total HDRS	24.0 $\pm$ 5.0	21.3 $\pm$ 4.1	.1	29.3 $\pm$ 7.1	27.3 $\pm$ 5.4	.3

PLA2, phospholipase A2; HDRS, Hamilton Depression Rating Scale; INF, interferon.

^aThe HDRS items were divided into the six clusters: Core (items 1, 2, 7, 8, 10, 13), Sleep (items 4, 5, 6), Activity (items 7, 8), Anxiety (items 9, 10, 11, 12, 13), Psychic Anxiety (items 9, 10), Somatic Anxiety (items 11, 12, 13), and Delusion (items 2, 15, 20) (40.62).

Table 4

Characteristics of 63 Study Participants Available for PUFA Levels Analysis

	Depressed	Nondepressed	<i>p</i>
<i>n</i>	21 (33%)	42 (67%)	—
Gender (Male/Female, <i>n</i>)	10/11	32/10	.023 ^a
Age, yrs (mean ± SD)	49 ± 11	46 ± 11	.3
Education (yrs)	8 ± 3	9 ± 4	.2
Marriage, <i>n</i>			
Married	16	33	.8 ^a
Single/divorced/widow or widower	5	9	.8 ^a
Baseline BDI scores (mean ± SD)	5 ± 7	3 ± 6	.3
DHA levels (%; mean ± SD)	3.24 ± .09	3.38 ± .34	.024
EPA levels (%; mean ± SD)	.81 ± .04	.82 ± .04	.4
AA levels (%; mean ± SD)	5.55 ± .27	5.48 ± .26	.3

The *p* values from independent-samples *t* test, unless χ^2 test result.

PUFA, polyunsaturated fatty acids; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; AA, arachidonic acid; BDI, Beck Depression Inventory.

^a χ^2 test result.