Review article

Membrane phospholipid composition, alterations in neurotransmitter systems and schizophrenia

Teresa M. du Bois*, Chao Deng, Xu-Feng Huang

Neuroscience Institute of Schizophrenia and Allied Disorders (NISAD), NSW 2010, Australia
Department of Biomedical Sciences, University of Wollongong, Northfields Avenue, NSW 2522, Australia

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Abstract

This review addresses the relationship between modifications in membrane phospholipid composition (MPC) and alterations in dopaminergic, serotonergic and cholinergic neurotransmitter systems in schizophrenia. The main evidence in support of the MPC hypothesis of schizophrenia comes from post-mortem and platelet studies, which show that in schizophrenia, certain omega-3 and omega-6 polyunsaturated fatty acid (PUFA) levels are reduced. Furthermore, examination of several biochemical markers suggests abnormal fatty acid metabolism may be present in schizophrenia. Dietary manipulation of MPC with polyunsaturated fatty acid diets has been shown to affect densities of dopamine, serotonin and muscarinic receptors in rats. Also, supplementation with omega-3 fatty acids has been shown to improve mental health rating scores, and there is evidence that the mechanism behind this involves the serotonin receptor complex. This suggests that a tight relationship exists between essential fatty acid status and normal neurotransmission, and that altered PUFA levels may contribute to the abnormalities in neurotransmission seen in schizophrenia.

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Keywords: Brain function; Membrane phospholipid composition; Polyunsaturated fatty acid; Schizophrenia; Serotonin; Treatment

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Abbreviations: AA, arachidonic acid; Acb, nucleus accumbens; ACC, anterior cingulate cortex; ADP, adenosine diphosphate; Amg, amygdala; BPRS, brief psychiatric rating score; CA1–3, CA1–3 fields of the hippocampus; CHO, carbohydrate; CPRS, comprehensive psychiatric rating scale; CPUDL, caudate–putamen, dorsolateral; CPUDM, caudate–putamen, dorsomedial; CPUL, caudate–putamen, ventrolateral; CPUVM, caudate–putamen, ventromedial; Cx, cortex; D2, dopamine D2 receptor subtype; DA, dopamine; DAT, dopamine transporter; DG, dentate gyrus; DGLA, dihommo-gamma-linolenic acid; DHA, docosahexaenoic acid; E-E, ethyl eicosapentaenoate; EFA, essential fatty acid; Efamol, evening primrose oil; E-EPA, ethyl eicosapentaenoic acid; EPA, eicosapentaenoic acid; FA, fatty acid; FCx, frontal cortex; GLA, gamma-linolenic acid; HF, hippocampal formation; HPC, hippocampus; 5-HT1A, serotonin 1A receptor subtype; 5-HT2A, serotonin 2A receptor subtype; 5-HT2C, serotonin 2C receptor subtype; 5-HT, serotonin; 5-HTT, serotonin transporter; MPC, membrane phospholipid composition; MUFA, monounsaturated fatty acid; N, number of subjects; n-3, omega 3; n-6, omega 6; PANSS, positive and negative syndrome scale; PET, positron emission tomography; PfCx, prefrontal cortex; PUFA, polyunsaturated fatty acid/s; SANS, scale for the assessment of negative symptoms; SAPS, scale for the assessment of positive symptoms; SF, saturated fat; STR, striatum; VMAT2, vesicular monoamine transporter.

* Corresponding author. Department of Biomedical Science, University of Wollongong, Bldg 41 Room 323, Northfields Avenue, NSW 2500, Australia. Tel.: +61 2 42214300; fax: +61 2 42214096.
E-mail address: tmdbois@uow.edu.au (T.M. du Bois).

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1. Introduction

Schizophrenia is a debilitating mental disorder characterised by both positive and negative symptoms. Positive symptoms include hallucinations, delusions and thought disorganisation; while the negative symptoms include impaired motivation and decreased emotional expression (Lewis, 2000). The aetiology of schizophrenia remains to be elucidated, though researchers are coming to realise that it is probably multifactorial, involving interactions of many neurotransmitter systems and/or other factors. Some recent hypotheses put forward involve membrane phospholipid composition, glycine, vitamin D, noradrenalin and electrical dysfunction. Evidence from new techniques for investigating schizophrenia in vivo, such as positron emission tomography (PET), add new support to some of the more original hypotheses involving dopamine, serotonin and glutamate abnormalities (Farde, 1997).

Arguments have been put forward that in several cases, polyunsaturated fatty acids (PUFA) could be involved in the aetiology of schizophrenia. These are based on findings of reduced omega-3 \((n-3)\) and omega-6 \((n-6)\) PUFA (Yao et al., 1994; Arvindakshan et al., 2003), and of abnormalities in phospholipid metabolism in schizophrenia (Pettegrew et al., 1991; Messamore, 2003). The membrane phospholipid composition (MPC) hypothesis of schizophrenia argues that alterations in MPC of the brain, as a direct result of changes in fatty acid levels, affects aspects of brain function, such as neurotransmitter–receptor interaction (Horrobin, 1998).

Chalon et al. (2001) have inferred a link between MPC, neurotransmission and several neuropsychiatric affections. Studies from this group have focussed primarily on aspects of dopamine neurotransmission in an \(n-3\) PUFA deficient rat model. Dietary manipulation of MPC from this diet has been shown to increase in D2 receptor density in the nucleus accumbens, and reduce D2 receptor density in the frontal cortex. Similar patterns of D2 density have been reported in schizophrenia (Roth and Meltzer, 2000). Studies have also shown that high PUFA diets can affect serotonin and acetylcholine muscarinic receptor expression in the rat brain (Freund et al., 1986; Farkas et al., 2002; Aïd et al., 2003).

This review does not seek to evaluate all literature published on the membrane phospholipid composition hypothesis of schizophrenia. Rather, the objective of this paper is to determine whether a link between fatty acid abnormalities and alterations in neurotransmitter–receptor interactions in schizophrenia can be substantiated. However, it is worth noting that factors such as oxidative stress and altered immune function have also been related to PUFA defects in schizophrenia, which have been recently reviewed elsewhere (Yao et al., 2001; Yao and van Kammen, 2004).

Literature was obtained by searching the online databases 'Science Direct' and 'Ovid' using the keywords: 'membrane phospholipid composition', 'essential fatty acids', 'serotonin', 'dopamine' and 'diet', combined with the term 'schizophrenia' and by collecting references therein.

2. Schizophrenia and the MPC hypothesis

2.1. The role of essential fatty acids in the brain

Essential fatty acids (EFAs) such as arachidonic acid from the omega-6 family and docosahexaenoic acid from the omega-3 family, play important roles in neural membranes, once they are incorporated into membrane phospholipids (Fenton et al., 2000). The phospholipid composition of a membrane can influence the activity of ion channels and enzyme activities including Na\(^+\)/K\(^+\) ATPase, cAMP and cyclic nucleotide (Bourre et al., 1991). The activity of transporters and receptors is also sensitive to changes in lipid environment (Spector and Yorek, 1985), therefore a change in membrane phospholipid composition may affect neurotransmission (Horrobin, 1998). Arachidonic acid and its derivatives can modulate dopamine release and dopamine receptor activity, as well as serotonin and glutamatergic activity (Skosnik and Yao, 2003).

The ratio of \(n-6/n-3\) PUFA incorporated into membrane phospholipids (fluidity) can also influence receptor–ligand interaction, possibly by increasing the availability of binding sites on receptor proteins and/or by increasing receptor concentration in the membrane (Farkas et al.,...
1986), and no change in another (Vaddadi et al., 1996). However not all reports are consistent, as one group (Yao et al., 1994; Khan et al., 2002; Arvindakshan et al., 2003) found increases in DHA levels in one study (Vaddadi et al., 2003). Reduced AA and DHA levels in drug-free first episode patients (Peet et al., 1995; Yao et al., 2000). Confirmation that these abnormalities are present early in the disorder and are not due to the effects of medication come from studies showing reduced AA and DHA in drug-free first episode patients (Yao et al., 1994; Khan et al., 2002; Arvindakshan et al., 2003). However not all reports are consistent, as one group found increases in DHA levels in one study (Vaddadi et al., 1986), and no change in another (Vaddadi et al., 1996).

Skosnik and Yao (2003) note that smoking is more common among patients with schizophrenia, and that oxidative damage to PUFA from smoking may contribute to the decreases in EFAs reported. Addressing this issue, Hibbeln et al. (2003) measured EFA status in red blood cells of schizophrenia patients who smoked or were non-smokers. They report that AA levels were not reduced in schizophrenia patients who smoked compared to those who did not, but that DHA and EPA levels were reduced in the smoker group. Further studies are needed to clarify which EFAs are affected by smoking in schizophrenia.

2.3. Phosphorus magnetic resonance spectroscopy

An increased breakdown and decreased incorporation of fatty acids into neural membranes may be the cause of the reduced AA and DHA levels observed in schizophrenia. This is supported by magnetic resonance spectroscopy studies, pioneered by Pettigrew et al. (1991), showing reduced levels of phosphomonoesters but increased phosphodiester synthesis in the prefrontal cortex of drug naïve schizophrenic patients. Since phosphomonoesters are precursors for phospholipid membrane synthesis while phosphodiesters are the breakdown products, results were interpreted as a decreased incorporation of fatty acids into membranes concomitant with increased membrane catabolism present in schizophrenia.

Other studies of drug-free first episode and chronically ill medicated schizophrenic patients have replicated the finding of decreased phosphomonoesters (Kato et al., 1995; Keshavan et al., 1995; Stanley et al., 1995). The finding of increased phosphodiester synthesis is more difficult to reproduce. While an increase has been reported in drug-free first episode schizophrenic patients (Fukuzako et al., 1999a,b; Stanley et al., 1995) and chronically ill patients (Fujimoto et al., 1992; Deickon et al., 1994), a contrasting study by Volz et al. (1998) reported decreased phosphodiester synthesis in chronically ill medicated schizophrenia patients. They state that chronic neuroleptic treatment has an inhibitory effect on phospholipase A2. So while the finding of decreased phosphomonoesters in schizophrenia is reasonably consistent, the finding of increased phosphodiester synthesis appears more likely to occur in first episode rather than chronically medicated patients.

2.4. The niacin skin flush test

The niacin skin flush test also supports the idea of abnormal fatty acid metabolism in schizophrenia. Oral doses of an excess amount of niacin usually elicit a skin flush response (Stoughton et al., 1960). This response is mediated by the release of vasodilatory prostaglandins from the skin. Absence of the niacin skin flush response reportedly occurs in up to 80% of schizophrenic patients (Tavares et al., 2003), suggesting abnormal prostaglandin signalling in schizophrenia. Since AA is a precursor for prostaglandins, results from the skin flush test are consistent with the reports of decreased AA depletion in schizophrenia. Furthermore, there is the finding that patients with schizophrenia have a reduced prevalence of inflammatory disorders, which Horrobin (1998) believes is related to AA deficiency.

2.5. Increased phospholipase A2 activity

The metabolism of EFA derivatives, the prostaglandins, is regulated by phospholipase A2 (Mathews et al., 2000). Several early studies that investigated phospholipase A2 activity in schizophrenia found it to be increased compared to normal individuals (Gattaz et al., 1987, 1990, 1995). Recent studies have implicated the calcium independent subtype of phospholipase A2 as being decreased as opposed to the calcium dependent subtype (Ross et al., 1997; Tavares et al., 2003). Genetic abnormalities may be associated with the increased phospholipase A2 activity (Gattaz et al., 1990; Horrobin, 1998), although a recent study was unable to demonstrate such an association (Chowdari et al., 2001).

In summary, fatty acids in membranes play a crucial role in regulating enzyme activity, conformation of receptors and transporters as well as neurotransmission. A change in membrane phospholipid composition, such as that proposed to occur in schizophrenia may therefore interfere with these aspects of cell function. Phosphorus magnetic resonance studies and the niacin skin flush test both indicate abnormal fatty acid metabolism underlies the reduced levels of EFAs in schizophrenia.

2.6. Dietary supplementation with PUFA can improve mental health state in schizophrenia

Reductions of EFAs in schizophrenia have led researchers to test the effectiveness of EFA supplementation in schizophrenia patients (Table 1). Studies testing the effec-
tiveness of \( n \approx 3 \) PUFA trials have generally yielded negative results, with 3 out of 4 studies showing no significant improvement in mental health. Two studies did however, report improvements in tardive dyskinesia (Vaddadi et al., 1986; Wolkin et al., 1986).

Results from most \( n \approx 3 \) PUFA trials have demonstrated improvements for patients in mental health rating scales. Fenton et al. (2001) found no improvements in the positive and negative syndrome scale (PANSS) when supplementing with 3 g/day ethyl-eicosapentaenoic acid (E-EPA) and vitamin E. Recently, Arvindakshan et al. (2003) used a similar treatment to Fenton et al. (2001), and reported improvements on the PANSS in patients of around 25%, which were sustained for a 4-month washout period. Peet and Horrobin (2002) identified 2 g/day as being the most effective dose, compared to 1 and 4 g/day. Horrobin (2003) explain that the superior efficacy of the 2 g/day dose may be because this dose gives an increase in red cell EPA without any reduction of red cell arachidonic acid. Indeed, Fenton et al. (2001) reported a large decrease in AA/EPA ratio of patients after treatment, suggesting that treatment was ineffective as it depleted AA. Puri and Richardson (1998) report that in one drug-free patient, supplementation with 2 g/day EPA for 6 months led to 80–85% improvements in standard mental health rating scales.

There is evidence that EPA may be mediating its therapeutic effects through modulation of the serotonin (5-HT)\(_2\) receptor complex. In human platelets, 5-HT amplifies adenosine diphosphate (ADP)-induced platelet aggregation, which is mediated by 5-HT\(_2\) receptors (De Clerk, 1990). A recent study by Yao et al. (2004) investigated 12 patients with chronic schizophrenia following administration of 2 g/day of E-EPA daily for 6 months supplementary to ongoing antipsychotic treatment. EPA markedly enhanced 5-HT responsivity as measured by the magnitude of 5-HT amplification on ADP-induced platelet aggregation. This group previously demonstrated a significant inverse correlation between 5-HT responsivity and psychosis severity in unmedicated patients with schizophrenia (Yao et al., 1996). Reduced serotonin responsivity in schizophrenia patients

<table>
<thead>
<tr>
<th>Study</th>
<th>N</th>
<th>Duration</th>
<th>PUFA dose</th>
<th>Outcomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Holman and Bell, 1983</td>
<td>10</td>
<td>16 weeks</td>
<td>4 g Efamol</td>
<td>No significant changes in BPRS between treatment group or placebo</td>
</tr>
<tr>
<td>Vaddadi et al., 1986</td>
<td>21</td>
<td>12 weeks</td>
<td>(i) 1 g DGLA + neuroleptic</td>
<td>(i) 26% improvement in BPRS—non-significant</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(ii) DGLA + placebo</td>
<td>(ii) 33% improvement in BPRS—non-significant</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(iii) Placebo only</td>
<td>(iii) 8.5% improvement in BPRS</td>
</tr>
<tr>
<td>Wolkin et al., 1986</td>
<td>16</td>
<td>6 weeks</td>
<td>600 mg Efamol</td>
<td>No difference between GLA or placebo groups in BPRS</td>
</tr>
<tr>
<td>Vaddadi et al., 1989</td>
<td>48</td>
<td>32 weeks</td>
<td>12 caps Efamol qd</td>
<td>30% improvement in CPRS</td>
</tr>
<tr>
<td>Peet et al., 1996</td>
<td>20</td>
<td>6 weeks</td>
<td>10 g maxEPA</td>
<td>17% improvement in PANSS, no placebo effect</td>
</tr>
<tr>
<td>Peet and Mellor, 1998</td>
<td>45</td>
<td>–</td>
<td>(i) EPA</td>
<td>(i) 23.8% improvement in PANSS—positive symptoms only</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(ii) DHA</td>
<td>(ii) 3.3% improvement in PANSS</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(iii) Linoleic acid</td>
<td>(iii) 13.7% improvement in PANSS</td>
</tr>
<tr>
<td>Puri and Richardson, 1998</td>
<td>1</td>
<td>24 weeks</td>
<td>EPA</td>
<td>85% improvement in SAPS, 89% improvement in SANS</td>
</tr>
<tr>
<td>Shah et al., 1998</td>
<td>10</td>
<td>12 weeks</td>
<td>2 g EPA</td>
<td>29% improvement in PANSS, no placebo effect</td>
</tr>
<tr>
<td>Fenton et al., 2001</td>
<td>90</td>
<td>16 weeks</td>
<td>2 g/day E-E + vitamin E</td>
<td>No improvement</td>
</tr>
<tr>
<td>Peet et al., 2001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Exp. 1</td>
<td>45</td>
<td>12 weeks</td>
<td>2 g/day EPA or DHA</td>
<td>EPA was significantly greater at improving total PANSS compared to DHA or placebo</td>
</tr>
<tr>
<td>Exp. 2</td>
<td>30</td>
<td>12 weeks</td>
<td>2 g/day EPA</td>
<td>EPA group had significantly lower total PANSS scores compared to placebo</td>
</tr>
<tr>
<td>Peet and Horrobin, 2002</td>
<td>115</td>
<td>12 weeks</td>
<td>1, 2 or 4 g/day of E-E</td>
<td>Significant improvements in PANSS with 2 and 4 g/day E-EPA but placebo effect for patients on antipsychotics. Subjects on clozapine had significant improvements in all scales with no placebo effect. 2 g/day most effective dose.</td>
</tr>
<tr>
<td>Arvindakshan et al., 2003</td>
<td>73</td>
<td>16 weeks</td>
<td>EPA/DHA: 180:120 mg plus 500 mg of vitamin E/C</td>
<td>Significant improvements in PANSS (25%) which were significantly sustained for the 4 month washout period</td>
</tr>
</tbody>
</table>

BPRS: brief psychiatric rating score, CPRS: comprehensive psychiatric rating scale, DHA: docosahexaenoic acid, DGLA: dihomoo-gamma-linolenic acid, E-E: ethyl eicosapentaenoate, Efamol: evening primrose oil, E-EPA: ethyl eicosapentaenoic acid, EPA: eicosapentaenoic acid, Exp.: experiment, GLA: gamma-linolenic acid, N: number of human subjects in study, PANSS: positive and negative syndrome scale, SANS: scale for the assessment of negative symptoms, SAPS: scale for the assessment of positive symptoms.
may relate to a reduction in platelet 5-HT₂ receptor density. Furthermore, Lesch et al. (1993) has shown that the platelet 5-HT uptake site is identical to the brain 5-HT transporter. Therefore changes in platelet serotonin responsivity may reflect that of the brain of schizophrenia, where 5-HT₂ receptor abnormalities have been reported numerous (see Section 4.2).

Alterations in MPC as a result of abnormalities of EFA metabolism in schizophrenia have been explored. To link these fatty acid abnormalities with brain function abnormalities and symptoms of schizophrenia, this requires assessment of the effects of altered MPC on neurotransmitter–receptor interaction. One approach to evaluate this is to manipulate MPC using animal models, and examine the effects on neurotransmitter systems. In the succeeding section, we review experiments which have investigated the effects of fat diets on neurotransmitter systems in rats.

3. The effects of altered MPC on dopaminergic, serotonergic and cholinergic muscarinic receptor density

3.1. Altered cellular MPC affects dopamine levels and receptor binding density

Several studies have investigated the effects of different fat diets on dopamine receptor density in areas of the rat brain including the frontal cortex and nucleus accumbents (Table 2). These have been outlined in Table 2, and are discussed below:

The frontal cortex is part of the mesocortical dopamine system, which has tonic inhibition on the mesolimbic dopamine system (Khan and Davis, 2000). In this area, Chalon et al. (1998) measured monoaminergic neurotransmission in rats following administration of a high $n-3$ PUFA diet. Increased levels of both endogenous dopamine and $D_2$ receptors were evident. Conversely, Delion et al. (1996) examined dopaminergic neurotransmission following an $n-3$ deficient diet. This diet treatment group showed lower levels of endogenous dopamine and significant decreases in $D_2$ receptor levels in the frontal cortex when compared to control rats.

The nucleus accumbens belongs to the mesolimbic dopaminergic system and is important for cognition and emotion (Chalon et al., 2001). In this area, Zimmer et al. (2000) examined dopamine receptor and transporter binding density levels of $n-3$ deficient rats. No change was seen in $D_1$ receptor or dopamine transporter (DAT) densities. However, there was a significant increase in binding density of the $D_2$ receptor compared to control rats. Binding to the vesicular monamine transporter (VMAT₂) was lower in $n-3$ deficient rats compared to controls. In addition, there was a substantial reduction in the amount of dopamine released from the vesicular storage pool, while the cytoplasmic pool remained unchanged.

<table>
<thead>
<tr>
<th>Study</th>
<th>Diet</th>
<th>Study focus</th>
<th>Duration</th>
<th>Receptor levels after diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>Delion et al., 1996</td>
<td>$n-3$ deficient: (mg/100 g FA)</td>
<td>5-HT₂, D₁, D₂ and DAT in the FCx</td>
<td>8, 24, 52 and 104 weeks</td>
<td>5-HT₂: 46, 14, 7 and 18% ↑ binding in the FCx at 8, 24, 52 and 104 months, respectively DA levels: ↓ in FCx of $n-3$ deficient rats at all ages $D_2$/DAT: no difference $D_2$: 7, 18, and 9% ↓ binding in the FCx at 8, 24 and 52 weeks, respectively</td>
</tr>
<tr>
<td></td>
<td>SF: 16.8</td>
<td></td>
<td>Not reported*</td>
<td></td>
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<tr>
<td></td>
<td>MUFA: 61.9</td>
<td></td>
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<tr>
<td></td>
<td>$n-6$: 21.3</td>
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<td></td>
<td>$n-3$: &lt;0.1</td>
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<tr>
<td>Chalon et al., 1998</td>
<td>High $n-3$ PUFA: (mg/100 g FA)</td>
<td>5-HT, 5-HT₂, D₁, and D₂ in the FCx, STR, HPC and CB</td>
<td>8 weeks 43 * Wistar rats, 6–11/group</td>
<td>5-HT: 20% ↑ in FCx DA: 40% ↑ in FCx 5-HT₂/D₂: no difference in any area $D_2$: 10% ↑ in FCx, 7% ↓ in STR</td>
</tr>
<tr>
<td></td>
<td>SF: 44.5</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>MUFA: 35.5</td>
<td></td>
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<td></td>
<td>$n-6$: 2.6</td>
<td></td>
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<tr>
<td></td>
<td>$n-3$: 18.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zimmer et al., 2000</td>
<td>$n-3$ deficient: (mg/100 g FA)</td>
<td>D₁, D₂, DAT and VMAT₂ in the Acb</td>
<td>8 weeks 40 * Wistar rats*, 20/group</td>
<td>$D_2$: 35% ↑ binding in the Acb $D_2$/DAT: no difference In Acb: 90% ↓ in DA released from vesicular storage pool, cytoplasmic pool unchanged. 60% ↓ in VMAT₂</td>
</tr>
<tr>
<td></td>
<td>SF: 16.8</td>
<td></td>
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<tr>
<td></td>
<td>MUFA: 61.9</td>
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<td></td>
<td>$n-6$: 21.3</td>
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<td></td>
<td>$n-3$: &lt;0.1</td>
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</tr>
<tr>
<td>Farkas et al., 2002</td>
<td>High $n-3$ PUFA + vitamins and antioxidants: $n-6$/$n-3$ ratio = 1.54</td>
<td>M1, 5-HT₁A and NMDA in the hippocampus</td>
<td>24 weeks 20 * Wistar rats</td>
<td>NMDA: no difference M1: up to 40% ↑ binding in regions of the hippocampus 5-HT₁A: 12–20% ↑ binding in regions of the hippocampus</td>
</tr>
</tbody>
</table>

* $n$: number of rats in the study. *Indicates that the previous two generations of rats were fed an $n-3$ deficient diet. *: male. 5-HT: serotonin, Acb: nucleus accumbens, CHO: carbohydrate, DA: dopamine, DAT: dopamine transporter, FA: fatty acid, FCx: frontal cortex, HPC: hippocampus, M1: muscarinic receptor, MUFA: monounsaturated fatty acid, NMDA: glutamate receptor, PFCx: prefrontal cortex, PUFA: polyunsaturated fatty acid, SF: saturated fat, STR: striatum, VMAT₂: vesicular monoamine transporter.
Clearly the effects of diet on neurotransmission in these studies are regionally specific. In the nucleus accumbens, rats with \( n - 3 \) deficiency display increased basal levels of dopamine and increased D\(_2\) receptor density (Zimmer et al., 2000). In the frontal cortex, \( n - 3 \) deficient rats display low basal dopamine levels and decreased D\(_2\) receptors (Delion et al., 1996). Zimmer et al. (2000) hypothesize that the reduced amount of dopamine in the frontal cortex of \( n - 3 \) deficient rats might be decreasing the inhibition on the mesolimbic dopamine system, leading to the hyperfunction in this region.

3.2. Altered cellular MPC affects serotonin levels and receptor binding density

Serotonin neurotransmitter–receptor interaction can also be affected by diet. A diet deficient in \( n - 3 \) fatty acids had the effect of increasing 5-HT\(_2\) receptor binding density in the frontal cortex of rats (Delion et al., 1996). Conversely, Chalon et al. (1998) investigated endogenous serotonin levels as well as 5-HT\(_3\) receptor levels in rats fed a diet rich in \( n - 3 \) PUFA. These rats had higher levels of endogenous serotonin in the frontal cortex compared to control rats. However there were no differences in 5-HT\(_2\) receptor-binding densities in any area of the brain. Farkas et al. (2002) also examined a high \( n - 3 \) PUFA diet and reported an increase in 5-HT\(_{1A}\) receptor density in the hippocampus.

Recently, we found that rats fed a high \( n - 3 \) PUFA diet showed only minor differences in 5-HT\(_{2A}\) and 5-HT\(_{3C}\) receptor densities in the rat brain, compared to a low fat control group. However, rats on a high \( n - 6 \) PUFA diet showed a marked increase in 5-HT\(_{2A}\) receptor density in the caudate–putamen, a decrease in 5-HT\(_{3C}\) binding in the mamillary nucleus, and a reduction in serotonin transporter density in the hippocampus (unpublished findings).

3.3. Altered cellular MPC affects acetylcholine muscarinic receptor binding density

Literature concerning the effects of PUFA on cholinergic neurotransmission is somewhat limited. Freund et al. (1986) report that increased fluidity of synaptosomal membranes from an \( n - 3 \) enriched diet can increase binding to cholinergic muscarinic receptors. In agreement with this, Farkas et al. (2002), found up to 40% increases in M1 receptor densities in regions of the hippocampus, in rats on a high \( n - 3 \) PUFA diet.

In contrast, Aid et al. (2003) investigated acetylcholine release and muscarinic receptor binding in \( n - 3 \) PUFA deficient rats. Compared to controls, these rats had a marked increase in basal acetylcholine release in the hippocampus. This diet also caused a reduction in muscarinic receptor binding in the hippocampus. A study limitation is that they used the non-subtype selective muscarinic acetylcholine antagonist [3H]scopolamine, which binds to several muscarinic receptors, possibly masking the differential effects of the diet treatment on individual receptors.

In a recent study we examined [3H]pirenzepine binding to M1/M4 receptors and [3H]AF-DX384 binding to M2/M4 receptors in rats on a high \( n - 6 \) PUFA diet. While no changes in M1/M4 receptor density were observed in the cortex or striatum, these areas showed decreased M2/M4 binding densities (Du Bois et al., 2005). Given that [3H]AF-DX384 labels M2 receptors mainly in the cortex, and M4 receptors mainly in the striatum (Piggott et al., 2002) and since [3H]pirenzepine labels both M1 and M4 in the striatum, it is possible that a reduction in M4 receptor density, as shown by [3H]AF-DX384 binding, is masking increased M1 receptor density in this area.

The review hitherto has identified some of the effects of diet on neurotransmission in the rat brain. High \( n - 6 \) PUFA diets and \( n - 3 \) PUFA deficient diets seem to affect receptor densities to a greater extent than high \( n - 3 \) PUFA diets. Altering MPC through diet produces regionally specific changes that affect multiple receptors differentially. The succeeding section will briefly summarise the main findings of alterations in dopamine, serotonin and muscarinic receptor expression in schizophrenia.

4. Alterations in neurotransmitter receptor expression in schizophrenia

4.1. Dopamine receptor expression in schizophrenia

The evidence implicating dopamine in schizophrenia is based on the observation that all antipsychotics share the property of D\(_2\) receptor blockade (Seeman, 2001). There is also evidence for altered dopamine receptor expression in schizophrenia. Increased D\(_2\) receptor density has been found in post-mortem tissue from patients with schizophrenia (Seeman et al., 1993) as well as in vivo (Wong et al., 1986). A recent study by Dean et al. (2004) suggests D\(_2\) receptors may be decreased in the pituitary. Many studies reporting increased levels of D\(_2\) receptors have used tissue from patients who had been medicated most of their lives. Animal models show that antipsychotic treatment can cause increases in D\(_2\) receptors (Burt et al., 1977), although increases in D\(_2\) receptor levels have also been demonstrated in drug-free patients (Lee and Seeman, 1980; Wong et al., 1986). Farde (1997), who could not replicate the finding of elevated D\(_2\) receptor density in schizophrenia, suggests discrepant results may be due to differences in radioligands used, which often have affinity for more than one receptor.

A new form of the dopamine D\(_2\) receptor, known as D\(_2\) Longer, has been recently discovered. Tallerico et al. (2001) measured levels of this, as well as the other two forms D\(_2\) short and D\(_2\) long in the schizophrenic brain. Results of this study were that levels of D\(_2\) Longer CDNA were higher in the frontal cortex of the schizophrenic brain. Also, levels of D\(_2\) short plus D\(_2\) long were higher in the frontal cortex,
when compared to controls. One patient in this study who was drug-naïve had similar D₂ mRNA to the six other medicated patients.

### 4.2. Serotonin receptor expression in schizophrenia

Serotonin was first implicated in schizophrenia upon findings of its interactions with the hallucinogenic drug LSD (D-lysergic acid diethylamide) (Wooley and Shaw, 1954). Following this, studies using post-mortem brain tissue from patients with schizophrenia showed a marked downregulation of the serotonin 5HT₂A receptor (Bennet et al., 1979; Mita et al., 1986; Arora and Meltzer, 1991; Laruelle et al., 1993; Dean and Hayes, 1996; Gurevich and Joyce, 1997; Pralong et al., 2000; Zavitsanou and Huang, 2002) in cortical and subcortical brain regions (Table 3). However, not all studies report consistent findings. This can partly be explained by the confounding variables of the studies including medication status of subjects, age of subjects, the radioligand used, cause of death and regional area examined. In addition, support for the involvement of serotonin in schizophrenia has emerged from the discovery of “atypical” antipsychotic drugs. These are effective against both positive and negative symptoms, and part of their profile includes antagonism at the serotonin 5HT₂A receptor (Meltzer et al., 2003). Atypical antipsychotic drugs occupy 70–80% of cortical 5-HT₂A receptors at clinically effective doses, while possessing much less occupancy for striatal D₂ receptors. Meltzer et al. (2003) also suggest that 5-HT₁A receptor agonism is an important part of 5-HT₂A/D₂ antagonism in antipsychotics. Atypical antipsychotics may also possess affinity for additional receptors. For example, clozapine has affinity for D₄, histaminergic, cholinergic and noradrenergic receptors (Breier, 1995). Seeman (2001) however, believes that 5-HT₂A receptors have little role in the antipsychotic process, arguing that full blockade occurs at sub-therapeutic doses. Breier (1995) suggests that the effectiveness of atypical antipsychotics may be based on their ability to achieve a balance between serotonin and dopamine, not on their absolute potency for these receptors.

<table>
<thead>
<tr>
<th>Study</th>
<th>5-HT₁A levels</th>
<th>5-HT₂A levels</th>
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<tbody>
<tr>
<td><strong>Post mortem</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mackay et al., 1978</td>
<td>–</td>
<td>No difference in FCx</td>
</tr>
<tr>
<td>Bennet et al., 1979</td>
<td>–</td>
<td>↓ binding in PfCx</td>
</tr>
<tr>
<td>Whitaker et al., 1981</td>
<td>–</td>
<td>No difference in FCx in medicated patients, ↑ binding in drug-free patients</td>
</tr>
<tr>
<td>Reynolds et al., 1983</td>
<td>–</td>
<td>No difference in FCx</td>
</tr>
<tr>
<td>Mita et al., 1986</td>
<td>–</td>
<td>↓ binding in PfCx</td>
</tr>
<tr>
<td>Arora and Meltzer, 1991</td>
<td>–</td>
<td>↓ binding in PfCx</td>
</tr>
<tr>
<td>Hashimoto et al., 1991</td>
<td>↑ binding in PfCx and temporal Cx</td>
<td>–</td>
</tr>
<tr>
<td>Laruelle et al., 1993</td>
<td>↑ binding in PfCx</td>
<td>↓ binding in PfCx</td>
</tr>
<tr>
<td>Joyce et al., 1993</td>
<td>No change in binding in PfCx or temporal Cx</td>
<td>↑ binding in HF, ↑ binding in middle temporal gyrus</td>
</tr>
<tr>
<td>Burnet et al., 1996</td>
<td>↑ binding in the PfCx and ACC, with no change in mRNA</td>
<td>↓ mRNA in the PfCx, ↓ mRNA in superior temporal gyrus, No change in HF</td>
</tr>
<tr>
<td>Dean and Hayes, 1996</td>
<td>–</td>
<td>↓ binding in PfCx</td>
</tr>
<tr>
<td>Simpson et al., 1996</td>
<td>↑ binding in PfCx</td>
<td>–</td>
</tr>
<tr>
<td>Sumiyoshi et al., 1996</td>
<td>↑ binding in PfCx</td>
<td>–</td>
</tr>
<tr>
<td>Gurevich and Joyce, 1997</td>
<td>↑ binding in ACC, PfCx, Broca’s area and association motor Cx</td>
<td>↓ binding in ACC and association motor Cx</td>
</tr>
<tr>
<td>Dean et al., 1999</td>
<td>No difference in dorsolateral PfCx</td>
<td>–</td>
</tr>
<tr>
<td>Pralong et al., 2000</td>
<td>–</td>
<td>↓ binding in planum temporale</td>
</tr>
<tr>
<td>Zavitsanou and Huang, 2002</td>
<td>–</td>
<td>↓ binding in the ACC</td>
</tr>
<tr>
<td>Lopez-Figueroa et al., 2004</td>
<td>No change in mRNA levels in PfCx; ↑ mRNA in HF</td>
<td>↓ mRNA in HF</td>
</tr>
</tbody>
</table>

| **PET**                |               |               |
| Trichard et al., 1998  | –             | No change in whole brain or regional cortical binding |
| Tauscher et al., 2002  | ↑ medial temporal Cx, no change in PfCx | – |

| **Platelet**           |               |               |
| Pandey et al., 1993    | –             | ↓ binding in PfCx in unmedicated schizophrenics compared to control |
| Lewis et al., 1999     | –             | No difference in any brain region |
| Govitrapong et al., 2000| –             | ↑ binding in unmedicated compared to medicated schizophrenics |
| Arranz et al., 2003    | –             | ↑ basal levels |
| –                      |               | ↑ binding in patients on risperidone |

↑=increased, ↓=decreased. ACC: anterior cingulate cortex, Cx: cortex, FCx: frontal cortex, HF: hippocampal formation, PfCx: prefrontal cortex. Studies on medicated as well as drug-free schizophrenia patients are included.
Rather, neurotransmission seems to be sensitive to changes in rats on manipulated diets there are no striking similarities. When comparing receptor expression in schizophrenia and receptor expression have been reported in schizophrenia. Altered dopamine, serotonergic and cholinergic muscarinic neurotransmission in rats. Altered levels of PUFA, as well as altered dopaminergic, serotonergic and cholinergic muscarinic neurotransmission in rats has been suggested to occur in the striatum in schizophrenia (Holt et al., 1999; Crook et al., 1999).

Zavitsanou et al. (2004) examined [3H]pirenzepine binding to M1/M4 receptors in schizophrenia brain tissue. There was a significant reduction in the density of [3H]pirenzepine in the deep and upper laminae of the anterior cingulated cortex in the schizophrenia group compared to the control group. The decrease in M1/M4 muscarinic receptor binding density did not appear to be secondary to previous antipsychotic medication exposure, since they found no correlation between [3H]pirenzepine binding and final recorded antipsychotic drug dose in the schizophrenia group. Furthermore, animal studies suggest that typical or atypical antipsychotic drugs tended to increase or have no effect on the density of [3H]pirenzepine labelled receptors in rat frontal cortex (Crook et al., 2001).

Scarr et al. (2001) have proposed a model explaining their findings of altered muscarinic and serotonergic neurotransmission in the hippocampus of patients with schizophrenia. Firstly, they found a decrease in the affinity of the serotonin transporter in schizophrenic compared to control (Dean et al., 1995). They believe that due to decreased 5-HTT affinity, there is an increased level of serotonin in schizophrenia. Furthermore, because serotonin stimulates release of acetylcholine (Hirano et al., 1995), there is a downregulation of M1/M4 receptors in schizophrenia.

4.3. Acetylcholine muscarinic receptor expression in schizophrenia

A typical antipsychotic drugs also have strong antimuscarinic properties (Raedler et al., 2000). Furthermore, studies on post-mortem brain tissue from patients with schizophrenia suggest that there are regionally specific changes to muscarinic acetylcholine receptors. Findings include increased M2/M4 receptor binding in the orbital frontal cortex and putamen (Owen et al., 1981; Watanabe et al., 1983) and decreased binding in frontal, parietal and temporal cortices (Bennet et al., 1979). More recently, Dean et al. (1996) and Crook et al. (2000, 2001) found decreased M1/M4 receptor binding in the striatum, hippocampal formation and prefrontal cortex in schizophrenia. Also, a deficit in cholinergic interneurons, concomitant with decreased binding to muscarinic M2/M4 receptors, has been suggested to occur in the striatum in schizophrenia (Holt et al., 1999; Crook et al., 1999).

Before firm conclusions can be made linking alterations in MPC with that of altered receptor expression, there is the need for further data on other types of fat diets, such as high saturated fat or high n−6 PUFA diets. Furthermore, the effects of altered MPC on other neurotransmitter receptors implicated in schizophrenia pathology, such as glutamate, could be examined. Abnormalities in glutamate NMDA receptor expression have been reported in several brain regions, including increased expression in the superior temporal gyrus (Grimwood et al., 1999; Nudmamud and Reynolds, 2001), anterior cingulate cortex (Zavitsanou et al., 2002) and prefrontal cortex (Simpson et al., 1991; Ishimaru et al., 1994). NMDA receptors may also be of relevance to the MPC hypothesis since they have been implicated in the production of free radicals, which may alter membrane PUFA.

References


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