



Contents lists available at ScienceDirect

## Psychiatry Research: Neuroimaging

journal homepage: [www.elsevier.com/locate/psychresns](http://www.elsevier.com/locate/psychresns)

Brief report

## Neuroprotective effects of ethyl-eicosapentaenoic acid in first episode psychosis: A longitudinal T<sub>2</sub> relaxometry pilot study

Stephen J. Wood<sup>a,\*</sup>, Luca Cocchi<sup>a</sup>, Tina-Marie Proffitt<sup>b</sup>, Mirabel McConchie<sup>b</sup>, Graeme D. Jackson<sup>c,d</sup>, Tsutomu Takahashi<sup>a,e</sup>, Christos Pantelis<sup>a</sup>, Patrick D. McGorry<sup>b</sup>, Gregor E. Berger<sup>b,f</sup>

<sup>a</sup>Melbourne Neuropsychiatry Centre, University of Melbourne, Australia

<sup>b</sup>Orygen Youth Health Research Centre, University of Melbourne, Australia

<sup>c</sup>Brain Research Institute, Australia

<sup>d</sup>Florey Neurosciences Institute, Melbourne, Australia

<sup>e</sup>Department of Neuropsychiatry, University of Toyama, Toyama, Japan and Core Research for Evolutional Science and Technology, Japan Science and Technology Corporation, Tokyo, Japan

<sup>f</sup>Department of Research & Education, The Schloessli Clinic, Zurich, Switzerland

## ARTICLE INFO

## Article history:

Received 22 October 2009

Received in revised form 9 December 2009

Accepted 10 December 2009

## Keywords:

Neuroprotection

Schizophrenia

Hippocampus

MRI

## ABSTRACT

We used magnetic resonance imaging to examine the effect of ethyl-eicosapentaenoic acid (E-EPA) on hippocampal T<sub>2</sub> relaxation time in first episode psychosis patients at baseline and after 12 weeks of follow-up. There was an increase in T<sub>2</sub> in the placebo group but not in the E-EPA group, suggesting a neuroprotective effect of E-EPA treatment. In addition, the smaller the increase in T<sub>2</sub>, the greater the improvement in negative symptoms.

© 2010 Elsevier Ireland Ltd. All rights reserved.

### 1. Introduction

Omega-3 fatty acids (essential fatty acids or EFA) are critical to normal brain functioning (Bazan, 2005; Bazan et al., 1995), and are potential adjunctive treatments for psychotic disorders (Berger et al., 2003). These effects may arise via a number of mechanisms, such as through the incorporation of EFAs into brain cell membranes (Tappia et al., 1997), EFA-induced alteration of neurotransmission (Yao et al., 2004) and EFA-driven reduction of oxidative stress (Lonergan et al., 2002).

Previous neuroimaging studies have shown that treatment with ethyl-eicosapentaenoic acid (E-EPA, an omega-3 fatty acid) may result in significant increases in a marker of neuronal integrity (Frangou et al., 2007) and increased brain antioxidant levels (specifically glutathione Berger et al., 2008). Although the former report was in bipolar disorder and the latter in first episode psychosis, they both suggest a neuroprotective effect of E-EPA mediated by reducing oxidative stress.

One limitation of these studies is that they did not examine the effect of E-EPA on membrane fluidity, or look for subtle regional

effects. One way to do this using magnetic resonance imaging is to measure brain water proton transverse relaxation times (T<sub>2</sub>). T<sub>2</sub> is an index of the water present in neuronal tissues, and T<sub>2</sub> increases are thought reflective of reduced neuronal health. The method has been extensively used to quantify hippocampal sclerosis in patients with epilepsy (Van Paesschen et al., 1997) and to detect abnormalities in normal-appearing white matter (Neema et al., 2009). Previous work in bipolar disorder showed that treatment with EFAs was associated with T<sub>2</sub> reductions across the whole brain (Hirashima et al., 2004) indicative of a neuroprotective action.

Here we present a pilot placebo-controlled study of the effect of E-EPA on hippocampal T<sub>2</sub> measures in first episode psychosis patients (FEP). There were two reasons for selecting the hippocampus as our region of interest. First, the hippocampus is known to be involved in schizophrenia, the region has already been shown to be sensitive to EPA in a spectroscopy study (Berger et al., 2008), and the hippocampus may be particularly sensitive to neuroprotective agents because it is one of the few brain areas in which adult neurogenesis occurs. Second, measurement of T<sub>2</sub> in the hippocampus is simplified by the fact that the region has clear boundaries, enhancing reliability, and a number of other relevant areas are not measurable because of the sequence design (e.g. posterior regions, orbitofrontal cortex).

\* Corresponding author. c/o National Neuroscience Facility, 161 Barry Street, Carlton Victoria 3053, Australia. Tel.: +61 3 83441877; fax: +61 3 94380469.

E-mail address: [sjwood@unimelb.edu.au](mailto:sjwood@unimelb.edu.au) (S.J. Wood).

## 2. Method

### 2.1. Demographic data

All participants were patients of the Early Psychosis Prevention & Intervention Centre (Orygen Youth Health Clinical Program, Melbourne). The Mental Health Research and Ethics Committee (Melbourne Health) approved the study, and inclusion criteria have been reported elsewhere (Berger et al., 2008). Participants were part of a larger randomized, double-blind, placebo-controlled clinical trial (Berger et al., 2007) investigating the augmenting effects of E-EPA [(5Z,8Z,11Z,14Z,17Z)-eicosa-5,8,11,14,17-pentaenoic acid] in 80 drug-naïve or early-treated FEP patients, and were also included in our recent report of the effect of EPA on brain spectroscopy measures (Berger et al., 2008). Participants were included in this study if they had T<sub>2</sub> relaxometry maps at both baseline and after 12 weeks. Seventeen FEP subjects, eight in the placebo arm (25% female, 75% neuroleptic-naïve) and nine taking a daily dose of 2 g E-EPA (33% female, 44% neuroleptic-naïve), met these criteria and were included in the study. The two groups were similar in almost all demographic and clinical measures ( $P > 0.12$ , see Table 1), but did differ in the proportion diagnosed with an affective psychosis.

### 2.2. Neuroimaging

#### 2.2.1. Hippocampal T<sub>2</sub> relaxometry

All scans were performed on a 3 T GE LX Horizon scanner (GE Healthcare, Milwaukee, USA) at the Brain Research Institute, Victoria, Australia. Eight tilted, coronal T<sub>2</sub> weighted images (perpendicular to the long axis of the hippocampus) were obtained over a range of echo times (29 to 231 ms; TR = 4 s, slice thickness = 6 mm with 1.5 mm gap, matrix 256 × 256, FOV 24 cm). Single exponentials were fitted to the image data of corresponding voxels from these eight echoes using iBrain (Brain Research Institute, Melbourne). This created a series of T<sub>2</sub> maps, one for each coronal slice. The brightness of each individual voxel on this map represents its calculated T<sub>2</sub> relaxation time.

All images were coded for blind analysis. Regions of interest were traced for two hippocampal regions in each hemisphere using ANALYZE 7.2 software (Mayo clinic, Rochester, Minn). The head was traced on the most caudal slice on which the uncus was still visible. The body was traced 15 mm (2 slices) back from the head.

Regions were made as large as possible whilst avoiding partial volume effects (the result of the inclusion of CSF). T<sub>2</sub> relaxation times (ms) were recorded by averaging the relaxation time for all voxels contained in the region of interest. The tracing was undertaken by a single researcher (TT). To assess reliability, hippocampal T<sub>2</sub> relaxation times were compared to those obtained by another researcher (SJW) on

10 randomly selected brains. Inter and intra-rater reliability (assessed with intraclass correlation coefficients) ranged from 0.82 to 0.93.

## 3. Results

A repeated-measures group × laterality × region × time ANOVA showed a significant main effect of time ( $F_{1,15} = 6.19$ ,  $P = 0.025$ ; follow-up > baseline) and region ( $F_{1,15} = 24.69$ ,  $P < 0.001$ ; head > body), and a trend interaction between time and group ( $F_{1,15} = 3.49$ ,  $P = 0.083$ ). There was no significant effect of laterality ( $F_{1,15} = 0.05$ ,  $P = 0.820$ ), and no three- or four-way interactions (all  $P > 0.2$ ).

*Post hoc* analyses were conducted to investigate the interaction trend between time and group (see Table 1). Because of the main effect of region but not of hemisphere, we analyzed the head and the body of the hippocampus separately, but combining data from both hemispheres. Paired *t*-tests contrasting T<sub>2</sub> at baseline and follow-up showed a non-significant increase in the E-EPA group for both hippocampal region (head:  $t(8) = 0.331$ ,  $P = 0.749$ , Cohen's  $d = 0.14$ ; body:  $t(8) = 0.348$ ,  $P = 0.737$ ,  $d = 0.15$ ). In contrast, a significant increase in T<sub>2</sub> was found in the placebo group for the body ( $t(7) = 2.405$ ,  $P = 0.047$ ,  $d = 0.86$ ), and a large, near-significant, increase for the head ( $t(7) = 2.214$ ,  $P = 0.062$ ,  $d = 0.85$ ).

### 3.1. Correlational analyses

No significant correlations were found between change in T<sub>2</sub> and change in the GAF score or total positive symptoms. When both groups were combined, a significant positive correlation was found between change in T<sub>2</sub> and change in total negative symptoms for the head but not the body (head  $r = 0.49$ ,  $P = 0.045$ ; body  $r = 0.41$ ,  $P = 0.103$ ).

## 4. Discussion

In this pilot study we examined the effects of E-EPA on hippocampal T<sub>2</sub> relaxation time in a group of young FEP patients. Although not reaching significance (most likely because of small groups), effect sizes clearly indicated a large increase in the placebo group that was not seen in the E-EPA group. These findings support a general neuroprotective effect of E-EPA (e.g. Frangou et al., 2007), perhaps through an increase in the antioxidant glutathione and subsequent oxidative stress reduction (Berger et al., 2008). Our results also indicate that when both groups were combined, a significant positive correlation was found between change in T<sub>2</sub> (head) and change in total negative symptoms. In other words, the smaller the increase in T<sub>2</sub>, the greater the improvement in negative symptoms.

**Table 1**

Sociodemographic and clinical information for placebo and E-EPA groups. (SD = standard deviation, PANSS = Positive and Negative Symptom Scale, GAF = General Assessment of Function).

	E-EPA	PLACEBO	Statistic
Sex (n)	6 m, 3 f	6 m, 2 f	$\chi^2 = 0.091$ ; $P = 0.70$
Age at baseline, years (SD)	18.7 (2.1)	21.6 (5.1)	$t(15) = 1.576$ ; $P = 0.13$
Median duration of treatment, days (range)	1 (0–10)	0 (0–11)	$t(15) = 0.690$ ; $P = 0.50$
Cumulative dose of chlorpromazine equivalents during the period of the study, mg (SD)	15809 (9513)	20730 (7338)	$t(15) = 1.182$ ; $P = 0.26$
Median duration of untreated psychosis at entry to the study, months (range)	3.25 (0.25–7.0)	9.0 (0.25–36)	$t(15) = 1.749$ ; $P = 0.12$
Schizophrenia spectrum disorder diagnosis (%)	56	100	$\chi^2 = 0.523$ ; $P = 0.03$
Tobacco smokers (subjects)	3	3	
Mean PANSS total at intake (SD)	84.0 (15.1)	79.8 (18.7)	$t(15) = 0.507$ ; $P = 0.62$
Mean PANSS total at 12 weeks (SD)	66.9 (13.5)	60.8 (15.2)	$t(15) = 0.887$ ; $P = 0.39$
Mean GAF at intake (SD)	42.3 (12.7)	46.1 (8.8)	$t(14) = 0.674$ ; $P = 0.51$
Mean GAF at 12 weeks (SD)	58.1 (10.3)	61.6 (10.0)	$t(14) = 0.692$ ; $P = 0.50$
Mean of hippocampus T <sub>2</sub> head baseline (SD)	100.8 (3.0)	98.3 (3.5)	$t(15) = 1.570$ ; $P = 0.14$
Mean of hippocampus T <sub>2</sub> head follow-up (SD)	101.4 (5.9)	102.0 (5.1)	$t(15) = 0.252$ ; $P = 0.80$
Mean of hippocampus T <sub>2</sub> body baseline (SD)	97.3 (3.4)	94.9 (2.9)	$t(15) = 1.578$ ; $P = 0.13$
Mean of hippocampus T <sub>2</sub> body follow-up (SD)	97.8 (2.8)	98.7 (5.5)	$t(15) = 0.436$ ; $P = 0.67$

This suggests that treatment with E-EPA may be associated with clinical improvement at least in part through preventing additional hippocampal damage, and is similar to our previous finding of a relationship between glutathione and negative symptoms (Berger et al., 2008).

In our clinical trial, patients with a non-affective psychosis responded best to E-EPA (Berger et al., 2007). Given that only half of the E-EPA group in this study had such a diagnosis (see Table 1), this may represent a significant confound. Indeed, examination of the data by diagnostic subgroup showed that the non-affective psychosis patients in the E-EPA group had a mean  $T_2$  decrease of 1.1%, compared with an increase of 2.5% in the affective group and an increase of 4% in the placebo group. The reason for this difference is unclear, but suggests greater neuroprotective effect of E-EPA in non-affective psychosis. It is possible that this larger beneficial effect in non-affective psychosis subjects is related to a greater level of oxidative stress at baseline (see Wood et al., 2009). Antipsychotic medication (i.e. olanzapine, quetiapine and risperidone) was not standardized but distribution during the study was similar in both groups. Cumulative antipsychotic dose was also similar, although slightly higher in the placebo group. While no significant correlation between cumulative dose and change in  $T_2$  (body and head) was found, there is evidence for an effect of antipsychotics on  $T_2$  signal, at least for white matter (Barnes et al., 1991).

Considering the pilot nature of this study, results need to be interpreted very cautiously. The small sample size, the different diagnoses and the low number of females represent important limits. Despite these limits our results provide a solid rationale to justify further investigations on the neuroprotective effect of E-EPA in psychotic populations and support recent findings showing that E-EPA treatment in pre-psychotic individuals can reduce the risk of transition to frank psychotic disorder (Amminger et al., 2010).

#### Acknowledgements

This work was supported by two NH&MRC project grants (ID: 145627; 209062), a NH&MRC program grant (ID: 350241), and the Colonial Foundation, Melbourne, Australia. A/Prof Wood is supported by a Clinical Career Development Award from the NH&MRC, and Dr Cocchi by a Swiss National Science Foundation young investigator award (PBLAB3-119622). A Margaret & Walter Lichtenstein Foundation award (University of Basel) supported Dr Berger. These funding sources had no further role in study design; in the collection, analysis and interpretation of data; in the writing of the report; and in the decision to submit the paper for publication. None of the authors has a

conflict of interest with any manufacturer or distributor of omega-3 fatty acids. The presented data were part of an investigator-initiated trial. Laxdale Ltd (now owned by Amarin Corporation) provided the study medication for free. None of the research centres received any financial support from Laxdale Ltd.

#### References

- Amminger, P.G., Schäfer, M.R., Papageorgiou, K., Klier, C.M., Cotton, S.M., Harrigan, S.M., Mackinnon, A., McGorry, P.D., Berger, G.E., 2010. Long-chain omega-3 fatty acids for indicated prevention of psychotic disorders: a randomized, placebo-controlled trial. *Archives of General Psychiatry* 67, 146–154.
- Barnes, D., Harvey, I., McDonald, W.L., Ron, M.A., Moore, S., 1991. The effect of chlorpromazine and methylprednisolone on NMR relaxation times in the normal cat brain. *Magnetic Resonance in Medicine* 18, 232–236.
- Bazan, N.G., 2005. Lipid signaling in neural plasticity, brain repair, and neuroprotection. *Molecular Neurobiology* 32, 89–103.
- Bazan, N.G., Rodriguez de Turco, E.B., Allan, G., 1995. Mediators of injury in neurotrauma: intracellular signal transduction and gene expression. *Journal of Neurotrauma* 12, 791–814.
- Berger, G.E., Wood, S., McGorry, P.D., 2003. Incipient neurovulnerability and neuroprotection in early psychosis. *Psychopharmacology Bulletin* 37, 79–101.
- Berger, G.E., Proffitt, T.M., McConchie, M., Yuen, H., Wood, S.J., Amminger, G.P., Brewer, W., McGorry, P.D., 2007. Ethyl-eicosapentaenoic acid in first-episode psychosis: a randomized, placebo-controlled trial. *Journal of Clinical Psychiatry* 68, 1867–1875.
- Berger, G.E., Wood, S.J., Wellard, R.M., Proffitt, T.M., McConchie, M., Amminger, G.P., Jackson, G.D., Velakoulis, D., Pantelis, C., McGorry, P.D., 2008. Ethyl-eicosapentaenoic acid in first-episode psychosis. A 1H-MRS study. *Neuropsychopharmacology* 33, 2467–2473.
- Frangou, S., Lewis, M., Wollard, J., Simmons, A., 2007. Preliminary in vivo evidence of increased N-acetyl-aspartate following eicosapentaenoic acid treatment in patients with bipolar disorder. *Journal of Psychopharmacology* 21, 435–439.
- Hirashima, F., Parow, A.M., Stoll, A.L., Demopoulos, C.M., Damico, K.E., Rohan, M.L., Eskesen, J.G., Zuo, C.S., Cohen, B.M., Renshaw, P.F., 2004. Omega-3 fatty acid treatment and T(2) whole brain relaxation times in bipolar disorder. *American Journal of Psychiatry* 161, 1922–1924.
- Lonergan, P.E., Martin, D.S., Horrobin, D.F., Lynch, M.A., 2002. Neuroprotective effect of eicosapentaenoic acid in hippocampus of rats exposed to gamma-irradiation. *Journal of Biological Chemistry* 277, 20804–20811.
- Neema, M., Goldberg-Zimring, D., Guss, Z.D., Healy, B.C., Guttman, C.R., Houtchens, M.K., Weiner, H.L., Horsfield, M.A., Hackney, D.B., Alsop, D.C., Bakshi, R., 2009. 3 T MRI relaxometry detects T2 prolongation in the cerebral normal-appearing white matter in multiple sclerosis. *Neuroimage* 46, 633–641.
- Tappia, P.S., Ladha, S., Clark, D.C., Grimble, R.F., 1997. The influence of membrane fluidity, TNF receptor binding, cAMP production and GTPase activity on macrophage cytokine production in rats fed a variety of fat diets. *Molecular and Cellular Biochemistry* 166, 135–143.
- Van Paesschen, W., Revesz, T., Duncan, J.S., King, M.D., Connelly, A., 1997. Quantitative neuropathology and quantitative magnetic resonance imaging of the hippocampus in temporal lobe epilepsy. *Annals of Neurology* 42, 756–766.
- Wood, S.J., Yucel, M., Pantelis, C., Berk, M., 2009. Neurobiology of schizophrenia spectrum disorders: the role of oxidative stress. *Annals of the Academy of Medicine, Singapore* 38 (5), 396–401.
- Yao, J.K., Magan, S., Sonel, A.F., Gurklis, J.A., Sanders, R., Reddy, R.D., 2004. Effects of omega-3 fatty acid on platelet serotonin responsiveness in patients with schizophrenia. *Prostaglandins Leukotrienes and Essential Fatty Acids* 71, 171–176.