Low fat dietary intervention with ω-3 fatty acid supplementation in multiple sclerosis patients

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ABSTRACT

Objectives: To determine whether a low fat diet supplemented with ω-3 positively affects quality of life (QOL) in relapsing-remitting MS (RRMS) patients. Patients were randomized into a 1-year study comparing two interventions: FO group received a low fat diet (15% fat) with ω-3 fish oils and OO group received the AHA Step I diet (fat ≤ 30%) with olive oil supplements. The primary outcome was the Physical Components Summary Scale (PCS) of SF36. Additional measures using Modified Fatigue Impact Scale (MFIS), Mental Health Inventory (MHI), EDSS and relapse rate were obtained. Results: 31 RRMS patients were enrolled, mean follow up 11 month (SD 2.9m). Marginal significance in PCS/SF36 (p=0.050) and MHI (p=0.050) were seen at 6 months, favoring the FO group. Benefit on the fatigue favoring actually the OO diet was seen at 6 and 12 months (p=0.0591). Decrease in relapse rate was seen in both groups: FO: -0.79 (SD= 1.12) (p=0.0212) vs. OO: -0.69 (SD 1.11) (p=0.044). Trends of decreased levels of proinflammatory cytokines and chemokines mainly in FO group were seen. Conclusions: This pilot study suggests that a low fat diet with supplemental ω-3 was associated with beneficial effects on QOL, clinical, and immunological parameters in RRMS patients.

INTRODUCTION

Multiple sclerosis (MS) is a chronic and disabling disease of the central nervous system with unknown etiology. Converging lines of evidence suggest that the disease is caused by a disturbance in immune function in a genetic predisposed individual as a response to unknown

environmental factors, possibly infections (1, 2). Although MS is treatable with interferon β products (INF β; Avonex®, Betaseron® and Rebif®) and glatiramer acetate (GA; Copaxone®), benefits are only partial (3, 4, 5). Interventions in combination with approved disease modifying drugs are being tested.

The idea that dietary fat may play a role in the etiology of MS has been around for over 50 years (6, 7). However, there has been only minimal evidence to support dietary fat intake as a significant factor, and as mainstream investigators have pursued viral or autoimmune hypotheses.

An increasing body of evidence suggests interactions between the immune system and diet intake, although relationships appear to be complex (8, 9). A low fat diet, supplemented with EFA (essential fatty acids), is advocated for the population in general and has already established benefit in cardiovascular medicine. Similar dietary interventions showed beneficial effects in secondary cancer prevention (i.e. breast and prostate) and other autoimmune diseases (systemic lupus, rheumatoid arthritis, Crohn disease, psoriasis) (10, 11, 12, 13). There are several postulated mechanisms by which EFA may affect immune function including membrane structural changes or alterations in chemical mediators, such as eicosanoids (14). Many studies have shown that the metabolites (eicosanoids) of polyunsaturated fatty acids (PUFA) have immunomodulatory properties, the nature of which depends on the parent fatty acid. Both ω-3 and ω-6 PUFA are essential, being acquired through the diet and one type cannot be converted to the other (15,16). The spectrum of ω-3 PUFA activities is broad and includes suppression of lymphocyte proliferation, cytotoxic T lymphocyte activity, natural killer cell activity, macrophage-mediated cytotoxcity, neutrophil /monocyte chemostasis, major hystocompatibility complex II expression and antigen presentation, functions known to be dysregulated in MS (15; 17, 18). When dietary ω-3 fatty acids are consumed, they are incorporated into cell membranes and compete with ω-6 fatty

acids as substrates for the cyclooxygenase and lypoxygenase pathways (15,19). Some metabolites have primarily pro-inflammatory effects (ω-6 line), whereas eicosanoids from ω-3 line are generally less potent, or are considered to have a direct anti-inflammatory effect (20). Nonetheless, both ω-6 PUFA and saturated fatty acids have been actually associated to a deleterious chemopromotive role in cancer, probably due in part to suppression of apoptosis(21,22).

A few dietary interventions trials in MS using a low fat diet alone or supplemented primarily with linoleic acids (ω-6 family) followed Swank's and other epidemiologists observations of a significant association between high MS prevalence and diets high in meat consumption, diary products and low in fish products (6, 23, 24). Although the initial results were contradictory a reanalysis of these studies, combining the raw data, supported a beneficial effect of linoleic acid supplementation in decreasing the number and severity of relapses (25, 26, 27, 28). Swank' s own study although limited by lack of patient randomization suggested a long standing benefit on clinical parameters such as mortality but also relapses and disability in patients that maintained a very low (≤ 20 gr) fat diet supplemented with cod liver oil, especially if started early at disease onset (29,30). The last large dietary intervention in multiple sclerosis conducted by, Bates in 1989 compared ω-3 fatty acid supplementation to olive oil, both groups of patients had also increased their ω-6 fatty acids intake, based on the prior positive studies. A trend was observed in favor of patients treated with ω-3 fatty acids, fewer of these patients deteriorated compared with the group treated with olive oil supplementation ($p=0.07$) (31). Unfortunately, the authors concluded that ω-3 fatty acid supplementation added only marginally to the effects of ω-6 fatty acids and since, no additional controlled MS dietary interventions were carried out.

The objectives of the present study were to determine whether a low fat diet supplemented with ω-3 long chain polyunsaturated fatty acids (PUFA) positively affects quality of life in patients with

RRMS. Patients could be already on a protective disease modifying therapy (DMT: interferon ß or glatiramer acetate) but they had to be for at least 2 months on the same therapy before entering the study. The dietary effects on disease activity, disability progression as well as on different immunological parameters were also assessed. A decrease in soluble adhesion molecules (sICAM-1, sVCAM), prostaglandin E2 – (PGE2), leukotrienes (LTB4) and proinflammatory cytokines (IL-1β, IL-12, IFN-γ, TNF-α), leptin, and chemokines (IL-8, RANTES, MCP-1) was postulated as an expected beneficial effect from ω-3 PUFA supplementation. The putative therapeutic agent in this study was the ω-3 PUFA and its derivatives, but the potential therapeutic effect related to a low fat diet itself (≤ 15% total fat) and/ or supplementation of OO must be kept in mind.

MATERIALS and METHODS

Our study was a single-blinded randomized study looking for the effect of a low fat diet supplemented with fish oil vs. olive oil in a group of RRMS patients. The MS patients studied were recruited from the Baird MS Center and from an additional suburban neurological facility. The study was approved by the local Institutional Review Board. Subjects, with an age range of 18-60 years old, who have clinically diagnosed MS (Washington Panel criteria) with stable disease for at least 2 months, but with at least one exacerbation or more during the last three years, were enrolled in this study after signing the consent form. The patients could be on disease modifying therapy (i.e: interferon beta (Avonex®, Betaseron®) or Glatiramer acetate (Copaxone®)) on which they had to be for at least 2 months. To be eligible for the study the patients previous diet had to have contained more than 30% of total calories from fat (the average consumption in US is about 35% calories from fat) as determined by a food record. After enrollment patients were randomly assigned into one of two groups: group1 fish oil (FO) and group 2 olive oil- OO. The FO group

received a very low fat diet with ω-3 PUFA supplementation (6 fish oil capsules per day containing 1gr. FO: 65% ω-3; EPA 1.98 gr and DHA 1.32 gr/day) (EPAX 5500 EE, Tishcon corp.).Total fat intake including ω-3 PUFA supplements did not exceed 15% of the total calories consumed. The OO group received the American Heart Association Step I diet which is a controlled low cholesterol diet (total fat not exceeding 30 % of total daily calories and saturated fats < 10%), with "placebo" capsules containing equivalent of olive oil supplements (6 capsules of 1 gr olive oil per day).Patients knew the percentage of dietary fat but did not know if they are getting supplements of fish oil or olive oil. All patients received vitE-400 units, one multivitamin (not containing any PUFA) and calcium at least 500mg per day. No additional supplements or changes in symptomatic therapies were allowed during the study.

Participants were required to meet the dietitian weekly for the first 4 weeks, and then bi-weekly; which was followed by monthly sessions until the end of the study to monitor eating behavior. If the meeting did not occur the patient would be reached by telephone for a dietary consultation.

The patients had a neurological examination including EDSS at baseline, one month, than every 3 month till the end of 1 year and at the time of a new exacerbation, within 7 days of the new symptoms development. The examining practitioner was different than the treating physician, and whom was blinded to the diet the patient was eating. Patients completed a 7-day food-record and a diary for subjective side effects. The food records and diary were reviewed with the dietician and nurse at follow up visits or by telephone.

Patients completed the QOL questionnaires at baseline, 1 month, 6 months and 1 year. Plasma samples for immunological and toxicity monitoring were collected at baseline, 1, 3, 6, 9, and 12 month time points. Laboratory monitoring consisted in CBC/diff, comprehensive metabolic panel, and lipid profile (cholesterol, triglycerides, LDL and HDL). The primary outcome measure was the

Physical Component Scale (PCS) of the Short Form Health Survey Questionnaire (SF-36).

Additional secondary outcome measures included: the Modified Fatigue Impact Scale (MFIS) and the Mental Health Inventory (MHI).

Short Health Status Questionnaire (SF-36) is a health-related quality of life questionnaire used as a standard health survey. It consists of a 100 point scale divided in 8 subgroups that include: physical functioning, role-physical, bodily pain, general health, vitality, social functioning, role-emotional, and mental health. Two summary scales, Physical and Mental, have been derived from the 8 subgroups using factor analytic methods. Scales are set up so that a higher score indicates better health.

Modified Fatigue Impact Scale (MFIS) is a questionnaire designed to probe MS-specific quality of life, particularly perceiving the impact of fatigue on a variety of daily activities. The items of the MFIS can be aggregated into three subscales, the Physical, Cognitive, and Psychosocial subscales, or combined as a total MFIS score. Each of the components of the MFIS is an ordinal rating scale with items scored from 0, indicating no impact of fatigue on the item, to 4, indicating maximal impact of fatigue. The subscales scores can be added to provide a total score ranging from 0-84. Higher score indicate increased perception of fatigue.

Mental Health Inventory (MHI) is a widely-accepted measure of overall emotional functioning. It covers a wide range of negative and positive emotions, not just psychopathology. The 18-item version of the MHI is reasonably brief and reliable. The MHI has 4 subscales (Anxiety, Depression, Behavioral Control, and Positive Affect) and one total score. The subscales and total scores range from 0-100, which higher scores indicating better mental health.

Additional secondary outcomes were the clinical change in EDSS and relapse rate.

Changes in multiple immunological parameters including: soluble adhesion molecules (sICAM-1, sVCAM-1), prostaglandin PGE2, leukotrienes LTB4 and plasma cytokines (IL-1β, IL-4, IL-12, TNF-α, IFN-γ), leptin and chemokines (IL-8, RANTES, MCP-1) levels that were determined by the double sandwich ELISA (Enzyme Linked Immunosorbent Assay) kits, using manufacture protocols (R&D Systems (Minneapolis, MN). Serum fatty acid composition was examined using a gas liquid chromatograph (GLC) after methylation by the method of Lepage and Roy (32). The GLC was equipped with 30 meter EC-Wax capillary column (Alltech, Deerfield, IL). Nitrogen was used as carrier gas. Column temperature was maintained at 190C and the injection port and the detector temperature were set at 250 C and 260 C, respectively. Peaks were identified by their retention times using fatty acid methyl ester standards and quantitated by an integrator. Values were expressed as percentages of total fatty acids.

Statistics: This was a pilot randomized study including 31 patients. Repeated measures analyses were used to analyze the change over time in the Physical Component of the SF-36 as a measure of the change in quality of life. Treatment group, factors related to emotional status, fatigue and EDSS are known to effect quality of life and thus were examined initially for their effects on their primary response variable and statistically modeled accordingly. Next, the remaining factors of interest were considered for inclusion in the model based on the univariate results and on their association with each other. Similar analyses were performed for the secondary outcome measures.

RESULTS

Thirty-one patients were enrolled in this study (see flow chart-Fig 1). Clinical and immunological parameters including adhesion molecules, leukotrienes, and prostaglandins were obtained from 27

patients; additional cytokine/chemokine levels were obtained from 19 patients. Eight patients discontinued the study prematurely: 2 patients could not tolerate the diet (one from each group after one month in the study and two patients were not compliant during the first 2 months- data of these 4 patients was censored); 5 discontinued because active disease (4 were from OO and 1 from FO group, data available for all these patients at 3 and 6 m) and 1 patient became pregnant at 9 months (OO-group).

Baseline characteristics were similar in both groups (gender 85.7% vs. 84.6% female; age-mean 45.1 vs. 39.9; disease duration-6.9 vs. 4.6; EDSS 1.89 vs. 2.04, SF-36/PCS: 43.1 vs. 40.8, MFIS 50.6 vs. 38.3 and MHI 86.3 vs. 79.0) (Table 1). Mean follow up period on the diets was 11 month (SD 2.9 months). Out of 27 patients, 5 were on glatiramer acetate (Copaxone®), one on IFNß-1b (Betaseron®), 20 on IFNß-1a (Avonex®) and one patient was on no disease modifying therapy (DMT). Patients' therapy type did not differ between the 2 groups. Most of patients were recently diagnosed and started on disease modifying therapies with mean time on DMT of 1.11± 0.07 year (range of 2 months - 5 yr). When assessing the weight loss, the FO group lost in avg 5.9 lbs (SD=13.7) while OO group gained an avg 0.5 lbs. (SD=11.2).

A marginal significant difference (p=0.050) in PCS/SF36 between the FO and the OO group was seen at 6 months, suggesting that patients felt healthier in the FO group. Although a higher PCS was maintained at 12 months in the FO group compared to OO group, the difference did not reach statistically significance (Fig 2). The global SF36 remained unchanged in the FO group while in the OO had a tendency to worsen. A similar non-significant trend was seen also in EDSS. A marginal significant difference was seen in the mental health inventory scale- MHI (p=0.050) at the 6 month period between the FO and OO group; however, it was not maintained at 12 months

(Fig 3). A significant difference was seen in the fatigue scale MFIS (p=0.0348) at 6 months that was maintained for 12 months (p=0.059); however, the benefit favored the OO group (Fig 4).

There was a decrease in relapse rate when compared with the 1 yr prior to the study in both groups: FO group: -0.79 (SD= 1.12) (p=0.021) vs. OO group: -0.69 (SD 1.11) (p=0.044). There was no association between change in calories or weight loss and relapse rate or EDSS. Compliance **to the diet** based on individual food records was 69.2% from FO group and 66.7% in OO group, showing both groups complied with the diets. Twenty-five percent of non-compliant patients from the OO group were bellow the required limit, which was similar to what was observed in the FO group fat requirements and only 8.3% were above the limit. No significant clinical or immunological difference was seen based on the compliance status.

The lipid profile did not show significant changes during the 1-yr study, although a trend of increase in HDL was seen only in FO group (FO group: HDL increase of + 1.8 vs. a -11.2 decrease in OO group). The 1-yr HDL difference between the groups did reach significance (p=0.032) (Table.2.).

No significant effect was noticed on the soluble adhesion molecules sICAM and sVCAM during the study and between the two groups. However, a trend for a continuous decrease in PGE2 and LTB4 levels was seen primarily in the FO group (data not shown). Additional cytokines and chemokines data were obtained in 19 of the patients. No clear differences were seen in the pro-inflammatory (IL-1β, IFN-γ and TNF-α) and leptin levels during the study and between the groups. The baseline and study values of IL-4 were not different between the groups, but a trend of increase in IL-4 levels during the initial 6 months was seen only in the FO group. RANTES did not significantly change over the 12 months in neither group, although the FO diet group had an overall decrease of 21% while the OO diet had only a 1% decrease. Similarly MCP-1 did not

significantly change over the 12 months, however comparing the FO to OO diet there was 67% difference between the two groups favoring the FO group.

Fatty acids Chromatography Analysis

Fatty acid analysis chromatography was performed on the serum samples obtained during the study and reported as percentage of total fatty acids. The mean percentage changes between baseline and the last visit are seen in Table 3. Interestingly, the olinic acid (C18:1(9)) percentage decreased in both groups. Only the change in EPA (C20:5(3)) reached statistical significance between the two groups (an increase of 0.36± 0.58 in FO group vs. a decrease of 0.20±0.35 in OO group; p=0.027). The DHA (C22:6 (3)) increased in both groups. A decrease in saturated fatty acids (SFA) was seen in both groups, more in the OO group but the difference did not reach significance (-1.74 (±2.28) in FO group vs. -4.66 (±14.5) in OO group). Spearman correlations were performed between the FA percentage levels at the different time points and the clinical parameters evaluated. The DHA levels at month 6 as well as the difference in DHA from baseline to the last visit correlated with MFIS at the last visit (-067; p= 0.008 and -0.48; p=0.050 respectively) .The EPA levels at 6 and 9 months correlated well with EDSS at the last visit, (r=-0.56; p=0.039 and r=-0.78; p=0.022, respectively). EPA at 9 months correlated with PCS at the last visit (r=0.76; p=0.027). SFA at baseline correlated with the MFIS and the PCS at baseline (r=0.70; p=0.0002 and r=0.49, p=0.017, respectively). SFA at 6 months correlated with the MFIS at last visit (r=0.72; p=0.032), suggesting that higher SFA levels are associated with a higher perceived fatigue. Additional correlation analyses were performed between the EPA/DHA, SFA and the different adhesion molecules and cytokines levels. Most significant correlation were seen between decrease in LTB4, PGE2, ICAM and VCAM and the increase in ω-3 primarily EPA (data not shown).

DISCUSSION

The present study was designed to investigate the effects of two low fat dietary interventions with supplementation of ω-3 (EPA and DHA) vs. olive oil on subjective quality of life measures in MS patients who were on a stable disease modifying therapy (interferon ß or glatiramer acetate for at least 2 months before entering the study). Both interventions represent a significant dietary change compared to the patients pre-study diet with the intention to eliminate any placebo effect that can interfere with subjective QOL assessments. Current clinical outcomes assessment in MS trials relies on disability scales that are heavily influenced by ambulation and thus require very large sample size. Meanwhile these measures are not particularly sensitive to clinical symptoms that are considered meaningful by patients especially during early stages of the disease. Therefore, we've elected an alternative approach for this study - patient-centered outcome measures such as physical functioning quality of life, fatigue level, and emotional well-being. Additional clinical and immunological parameters were assessed prospectively during this one year study. All our patients but one were on a disease modifying therapy for almost one year (one patient only had 2 months since the initiation of IFN ß-1a therapy). It is reasonable to assume the benefit seen during this one year of study is not likely to be related to a delayed effect of DMT, but rather to a combined additive effect.

Patients with a chronic disease like MS often find themselves irresistibly drawn to any therapy that promises some control over their illness (33). Accordingly, dietary modifications appear very attractive although often patients cannot maintain a strict diet for long time periods. Most dietary studies are short (3-6 months) or the longer ones use primarily supplementation without vigorous control on the diet it self. (25-27). Our study was a 1-yr study with strict dietary requirements.

Patient dietary compliance was good, around 70% for both groups (based on their food record), better in the OO group (90%). Some patients even went lower than 30% of total fats suggesting that the diet was not difficult to maintain at least for the1 year period.

The FO group on a very low fat diet (≤15%) showed a significant effect on the primary outcome measure, the PCS/SF 36 at 6 months (p=0.05), compared to the OO group. Even though the FO group maintained higher PCS scores through the 12 months follow-up, the difference was no longer significant at the end of the study. A similar benefit at 6 months was seen using the MHI supporting the premises that MS patients did feel healthier physical and emotional in the FO group. This is probably due to the smaller number of patients that completed the 1-yr QOL assessments and the fact that more patients that discontinued the study were from the OO group and had active disease.

The 15% total fat diet was chosen based on the beneficial effect seen in breast and prostate secondary prevention studies (34, 35). Taking in consideration that our study design could not differentiate the beneficial effect of a low fat diet study vs. PUFA supplementation we decided to apply the most efficient combination for this initial pilot trial. Previous studies used between 1.1 to 5 gr of EPA+DHA per day and taking in consideration the longer period of our study and the combination with other disease modifying therapies we decided to go for 3.3 g daily for safety reasons (increased risk for prolonged bleeding time with higher dosages).

Patients knew their fat diet percent but they were blinded to the type of supplements they took. Although the putative therapeutic agent in this study was considered the FO the low fat diet intake and the beneficial effects of an OO diet must be kept in mind. Recent data indicate that OO a monounsaturated fatty acid (MUFA) rich oil, that is the traditional and essential component of the Mediterranean diet, has well proven cardiac benefits, and may also play an important role in the

modulation of immune system. This effect has been shown in animal models and also human autoimmune diseases as RA (36, 37). However, the effects found in animal models are considered more robust than in human studies. This is likely due to the higher levels of MUFA used in animals studies, which are not readily achievable in human studies (38). Therefore, part of the benefit seen in our OO group could be explained by the anti-inflammatory effects of the OO diet. Interestingly the benefit on fatigue, measured by MFIS, was seen only in the OO group, while the other parameters related to physical and mental status (PCS and MHI) were significantly better in FO group. One simple explanation could be that the 15% fat diet is too low in total calories, making the fatigue worse. However, the total mean daily calories (1466.7 in the FO group vs. 1602.3 in the OO group at 12 months) were not significantly different between the groups, although the FO group lost more body weight. It is difficult to know what role total caloric intake or the loss of body weigh might have played.

The decrease in SFA levels could be another possible beneficial consideration. The SFA levels were positively correlated with MFIS at baseline and at 6 months and the decrease in SFA in OO group was more noticeable than in the FO group although the difference did not reach significance. Although Swank after 50 yrs continue to consider the decrease in SFA as the most important protective element in MS (30), our data primarily supports a beneficial effect of omega-3 PUFA supplementation.

Another explanation for the beneficial effect on fatigue seen in OO group could be related to the antioxidants primarily the phenols present in olive oil that were shown to scavenge free radicals and increase protection against peroxidation an effect that was not seen in the FO supplementation studies(39,40). Although the precise underlying cause of fatigue in MS is not known, an increased oxidative status with free radicals production which has been recently associated with active MS

status could be considered (41,42). Consequently, we may speculate that the benefit seen in the fatigue level in the OO group could be related to the OO antioxidant effects. Although we added for both groups the 400 UI Vit. E supplements in order to prevent the increased PUFA peroxidation tendency, this may not have been a sufficient dose and we did not evaluate the oxidative enzyme status after this supplementation. Therefore, the balance between the ω-3 PUFA and Vit E levels will be important to be evaluated in future studies when determining the overall functional outcome.

The fatty acid data provided an insight on the difficulty monitoring different dietary intervention without an exact measurement of the diets ingested. The increase in EPA-omega-3 levels was indeed significantly higher in the FO group, as it was expected. The EPA levels at 6 and 9 months (seen primarily in FO group) were well correlated with the benefit seen in EDSS and PCS at the last visit, underscoring its beneficial consequence on the disease status. However, the unexplained increase in DHA levels in both groups is difficult to recognize unless indeed the DHA source in the OO group came from fish or other dietary products. Another explanation could be the effect of IFN that was shown to induce specific release of unsaturated fatty acids (i.e. oleic acid, linoleic acid, arachidonic acid and possibly EPA and DHA) from the cell membrane phospholipids without the release of SFA (43). A significant correlation was seen between the DHA levels and the decrease in fatigue measured by MFIS. The increase in DHA levels is clearly an unanticipated but possible explanation for the benefit on fatigue seen, particularly in the OO group.

In regard to the immunological parameters, no significant differences were seen in the cytokines, chemokines and adhesion molecules levels during the study and in between the 2 groups. Several studies have reported higher levels of soluble adhesion molecules (sICAM, sVCAM) in serum and CSF in MS patients, versus controls, with a positive correlation between levels in serum or

cerebrospinal fluid and cranial MRI activity (44, 45). Nevertheless, significant increases in serum levels for sVCAM in patients receiving IFN ß therapy were associated with a favorable treatment response after 1 year and were correlated to decreased MRI activity, whereas by contrast, stable or reduced sVCAM levels occurred more often in non-responders (46). Although FO and OO supplementation was shown to be associated with a beneficial decrease in soluble adhesion molecules levels, in our patients, being on IFN ß for almost one year, the combined dietary/IFN effect on adhesion molecules could have been obscured.

The leukotrienes LTB4 and LTC4 were shown to be increased in the CSF of MS patients, suggesting that the proinflammatory action of these eicosanoids may play a pathogenic role in the MS lesion (47). Leukotrienes increase capillary permeability at the post-capillary venule and may play a role in opening the blood-brain barrier at an early stage in MS lesion formation. In our study a trend for a continuous decrease in PGE2 and LTB4 levels was seen in the FO group, while in OO group a definite trend to a drop in LTB4 and PGE2 serum levels was seen only at the end of the study.

A trend of progressive increase of pro-inflammatory IFN-γ and TNF-α levels till month-6 was seen in OO group possibly reflecting the uncontrolled inflammatory process in the few patients that dropped at the 6 months time point due to active disease. In the FO group a pretty constant level was maintained during the 1 yr study. Interestingly the anti-inflammatory IL-4 cytokine levels showed a mild, but nonsignificant, increase for the first 6 months only in the FO group, returning to baseline at the end of the study. A similar positive trend of decreased pro-inflammatory chemokines RANTES and MCP-1 levels were seen primarily in the FO, and not in the OO group. Recent experimental models and clinical trials support an important role of pro-inflammatory chemokines in the pathogenesis of MS (48).

Conclusions: In summary, our data demonstrated that both FO and OO might have potential to improve the immune dysfunction in MS patients, although the FO effects emerge as a more efficient and more promptly intervention. Both low fat dietary interventions were well tolerated and associated with a decrease in number of relapses and benefit on immunological parameters. However despite the study limitations, including the open label design and the small number of patients, a trend toward an increased benefit on QOL, immunological and lipid profile parameters were seen primarily in the FO group although OO anti-inflammatory and anti-oxidant effects require further evaluations. Future larger studies evaluating the benefit of EPA/ DHA and OO supplementation in MS patients is warranted.

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Table 1. Baseline characteristics

	Group 1 N=14	Group 2 N=13
Gender F	85.7%	84.6%
Age-mean	45.1(7.7)	39.9(10.0)
Disease duration/yr.	6.9(5.9)	4.6 (3.5)
EDSS	1.9 (0.6)	2.04 (1.3)
SF-36/PCS:	43.1(8.4)	40.8 (7.8)
MHI	86.3 (16.2)	79.0 (15.6)
MFIS	50.6 (21.3)	38.3 (15.1)

* No significant difference on baseline clinical parameters

Table 2.

Variable	Diet group = FO		Diet group = OO		p-value
	Mean	SD	Mean	SD	
LDL at 1.visit	120.9	21.6	120.3	38.7	NS
LDL at last visit	125.1	25.9	148.5	93.6	NS
LDL change	4.2	23.1	25.0	93.3	NS
HDL at 1. visit	49.9	10.8	58.0	13.6	NS
HDL at last visit	50.6	10.3	47.1	17.0	NS
HDL change	1.8	7.6	-11.2	19.7	0.0318
Fat at 1. visit	19.6	12.9	21.8	8.5	NS
Fat at last visit	19.0	8.1	25.7	7.6	0.0384
Fat change	-0.6	13.9	3.9	8.5	NS
Calories at 1.visit	1470	437.7	1765	855.8	NS
Calories at last visit	1467	720.7	1602	563.5	NS
Calories change	-2.7	691.6	-162.2	553.1	NS

Table 3. Percent change in fatty acids (chromatography data)

	15% Diet Mean (SD)	30% Diet Mean (SD)	p-value
18:1(9) OO	-0.65 (5.47)	-3.38 (8.94	0.6664
20:5(3) EPA	0.36 (0.58)	-0.20 (0.35)	**0.0270**
22:5(3)	0.39 (0.58)	0.44 (0.92)	0.8852
22:6(3) DHA	0.76 (3.01)	1.45 (2.09)	0.3648
Omega 3's combined	1.51 (2.88)	1.94 (1.74)	0.4916
Sat. fatty acids	-1.74 (2.28)	-4.66 (14.5)	0.6256

Fig 1

Flow Chart

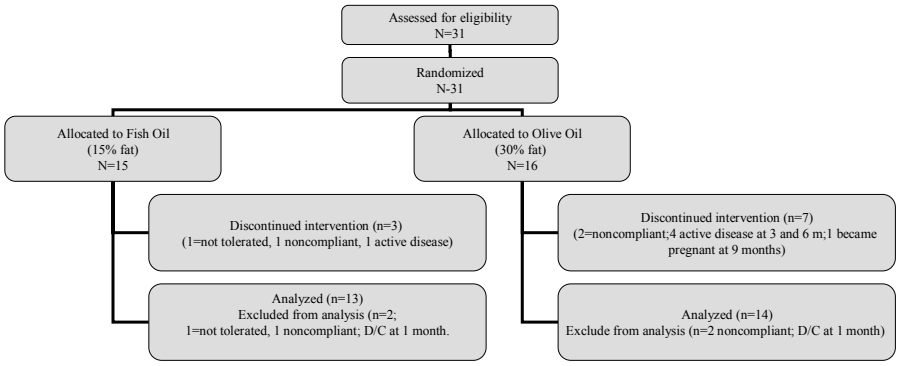
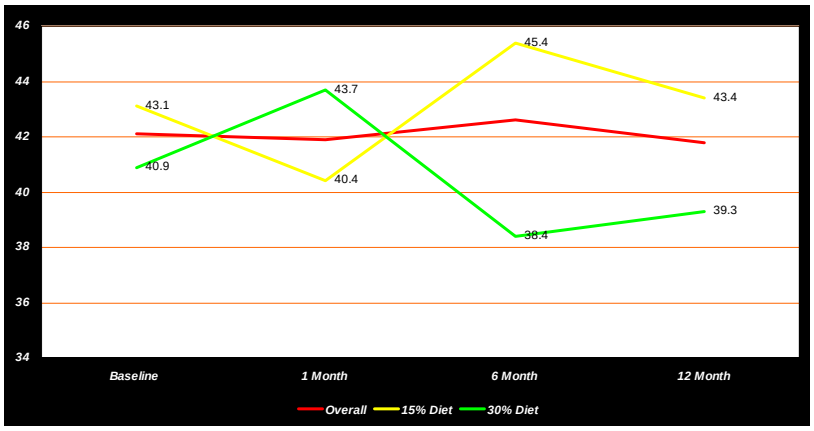


Fig. 2

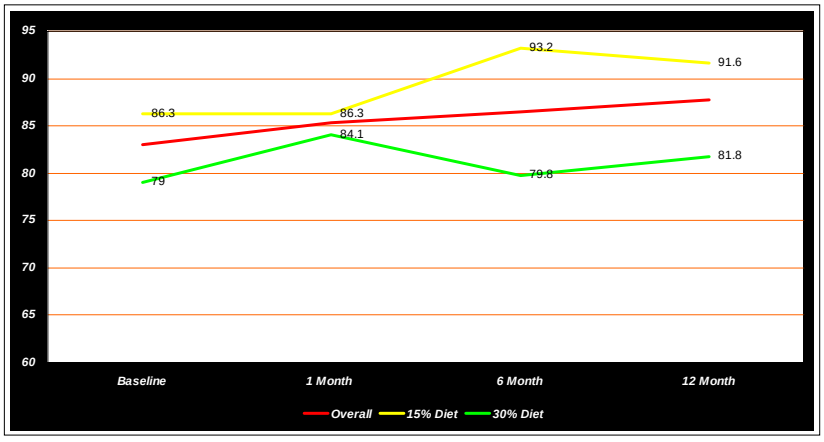
Trend of the PCS



p=0.05 at 6 months

Fig 3

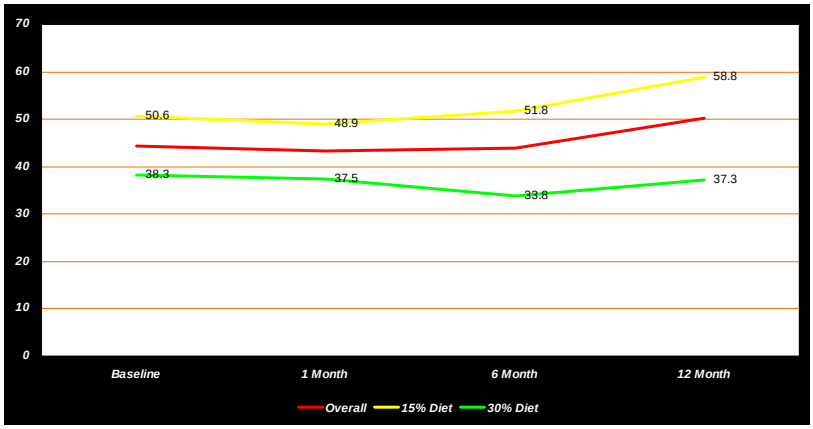
Trend of Mental Health Inventory



P=0.05 at 6 months

Fig. 4

Trend of MFIS



P=0.0348 at 6 months; p=0.0591 at 12 months