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A low omega-6 polyunsaturated fatty acid (n-6 PUFA) diet increases omega-3 (n-3) long chain PUFA status in plasma phospholipids in humans ☆

K.E. Wood [a,c,1], A. Lau [a,1], E. Mantzioris [b], R.A. Gibson [c], C.E. Ramsden [d], B.S. Muhlhausler [c,*]

[a] School of Medicine, Department of Nutrition and Dietetics, Flinders University, Adelaide, SA 5042, Australia
[b] School of Pharmacy and Medical Sciences, University of South Australia, Adelaide, SA 5001, Australia
[c] FOODplus Research Centre, School of Agriculture, Food and Wine, The University of Adelaide, Adelaide, SA 5064, Australia
[d] Laboratory of Membrane Biochemistry and Biophysics, National Institute on Alcohol Abuse and Alcoholism, National Institutes of Health, Bethesda, MD, USA

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ABSTRACT

This study aimed to determine the effect of reducing the dietary linoleic acid (LA) intake from ~5% to < 2.5% energy (%E) on n-3 long chain PUFA (LCPUFA) status in humans. Thirty-six participants followed a < 2.5%E LA diet for 4 weeks. Nutrient intakes were estimated from diet diaries and blood samples were collected for assessment of fatty acid composition in plasma and erythrocyte phospholipids. LA intakes were reduced from 4.6%E to 2%E during the low LA intervention ($P < 0.001$) while n-3 LCPUFA intakes were unchanged. LA and total n-6 PUFA content of plasma and erythrocyte phospholipids were significantly reduced after the low LA diet phase ($P < 0.001$). The n-3 LCPUFA content of plasma phospholipids was significantly increased after the low LA diet compared to baseline (6.22% vs. 5.53%, $P < 0.001$). These data demonstrate that reducing LA intake for 4 weeks increases n-3 LCPUFA status in humans in the absence of increased n-3 LCPUFA intake.

1. Introduction

The fatty acid composition of the typical diet of Western countries, including Australia, has undergone a substantial shift over the past half-century. This shift has been driven primarily by the replacement of animal fats with plant-based oils and spreads in cooking, baking and processed food and has resulted in a significant decrease in the per capita intake of saturated fats and a three-fold increase in the intake of the omega-6 polyunsaturated fatty acid (n-6 PUFA), linoleic acid (LA) [1,2].

The health impacts of this marked change in dietary fatty acid composition are unclear; however concerns have been raised that this substantial increase in LA intake could have negative impacts on cardiovascular and metabolic health [3,4]. This suggestion is based on the biochemical properties of the n-6 PUFA, since the long-chain derivative of LA, arachidonic acid (AA), gives rise to pro-inflammatory and pro-thrombotic compounds [5], and the fact that high circulating concentrations of n-6 PUFA have been implicated in an increased risk of inflammatory and allergic conditions in epidemiological studies [6]. Data from humans directly linking increased LA intake and disease are lacking. However, the findings of the Sydney heart study, recently published by Ramsden and colleagues, provided the first evidence from a randomized controlled trial that a dietary intervention in which saturated fats were replaced by concentrated sources of LA (as opposed to a mixture of n-6 and n-3 PUFA) was associated with an increased risk of death from coronary heart disease, raising renewed concerns about the impact of n-6 PUFA on human health [7].

LA competes with the short-chain n-3 PUFA alpha-linolenic acid (ALA) for the enzymes required for conversion to their respective long-chain derivatives and for incorporation into the plasma membrane [8]. Consequently, high LA diets may limit the capacity of increases in dietary n-3 PUFA intake to improve n-3 PUFA status [9]. Given that several bioactive mediators derived from n-3 LCPUFA are less potent in their inflammatory actions than their AA-derived analogues [10], and others actually have potent inflammation-resolving [11] or neuroprotective properties [12], this competition has the potential to contribute to negative health outcomes.

Current strategies to improve n-3 LCPUFA status of the population have focused almost exclusively on increasing dietary n-3

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* Corresponding author. Tel.: +61 8 8313 0848; fax: +61 8 8303 7135.
E-mail address: beverly.muhlhausler@adelaide.edu.au (B.S. Muhlhausler).
[1] These authors contributed equally to this work.

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LCPUFA intake, via increased consumption of oily fish or through fish oil supplementation [13]. However, many individuals struggle to achieve regular fish intake [14] and dwindling marine resources have raised concerns regarding the long-term sustainability of this approach [15]. The competition that exists between n-6 and n-3 has led to suggestions that lowering the LA content of the diet has the potential to both limit the production of n-6 derived pro-inflammatory mediators and enhance the biological efficacy of n-3 LCPUFA consumed in the diet. However, the ubiquity of LA in the food supply, particularly in pre-prepared and take-away foods, makes any attempt to reduce the n-6 PUFA intake of free-living humans challenging.

We previously designed a low n-6 PUFA diet, in which we reduced the LA content of the diet from ~5% to ~2%E by replacing standard plant-based oils and spreads with low n-6 PUFA alternatives (Macadamia oil and butter), and limiting intake of processed and take-away foods which utilize high n-6 PUFA oils [16]. Importantly, we showed that the reduction in LA intake could be achieved while still adhering to the national dietary recommendations (The Australian Guide to Healthy Eating (AGHE) guidelines) [17] and maintaining a saturated fat intake of less than 10%E [16]. The aim of the present study was to determine whether following this low LA diet for a 4 week period would result in reduced n-6 PUFA and increased n-3 LCPUFA content in plasma and erythrocyte phospholipids in healthy human subjects.

2. Patients and methods

2.1. Participants

Participants were recruited using email advertisements and flyers, and interested individuals were screened by research staff for eligibility. The inclusion criteria were: BMI < 35 kg/m^2 and weight stable, aged 18–65 years, able to eat > 5 meals at home per week and not regularly consuming more than 2–3 fish meals per week. Exclusion criteria included taking high potency fish oil supplements (> 3 g/day EPA/DHA), gastric mal-absorption, vegetarian or vegan diet, pregnant or breastfeeding. All participants gave informed consent. This study was approved by the Human Research Ethics Committees of the University of Adelaide and University of South Australia.

2.2. Study design

This was an open-label clinical trial which consisted of a 2 week control phase, during which participants were instructed to continue their habitual dietary intake, followed by 4 weeks on the low LA diet. No details of the low LA diet were provided at enrolment in order to minimize the potential for this to influence dietary choices during the 2 week control phase. Importantly this study design allowed for each subject to act as their own control which is an important consideration in free-living intervention dietary studies given the potential for considerable variability in dietary intakes between individuals. Baseline demographic, dietary and medical information was collected at enrolment.

2.3. Clinic appointments

All participants attended clinic appointments at enrolment, after the 2 week control phase and at the completion of the 4 week dietary intervention. All appointments were conducted between 7:30 am and 9:00 am after an overnight fast of at least 12 h. During the clinic appointments, weight and height were measured with participants in light clothing and without shoes, using a digital weighing scale (SALTER, Victoria, Australia) and a stadiometer (SECA, New South Wales, Australia) respectively and Body Mass index calculated (weight/height^2). Fasting venous blood samples (8 ml) were collected at each clinic appointment. After collection, blood samples were centrifuged for 15 mins at 3500*g* at 4 °C to separate plasma and erythrocytes for subsequent analysis of fatty acids.

2.4. Dietary information

All participants were asked to maintain a 3-day weighed food diary each week during both the control and dietary intervention periods. Instructions on completing the diary were provided at enrolment, and participants were instructed to record their dietary intake for 2 week days and 1 weekend day in each week of the study. Electronic kitchen scales (SALTER 1021, Victoria, Australia) and standard metric measuring cups (Décor Cook® Measuring Cups/Spoons, Victoria, Australia) were provided to participants to assist them in completing the weighed food diary.

2.5. Low LA diet

The low LA diet was based on the diet previously designed by our group [16] and aimed to achieve an LA intake of < 2.5%E whilst maintaining saturated fat intake at < 10%E and not altering the intake of n-3 LCPUFA. Participants were provided with Macadamia oil (Suncoast Gold Vitality™ Macadamia oil, 1.24 g LA/100 g) and butter (Western Star™ Butter, 1.60 g LA/100 g) and were instructed to use these in place of their usual oils and spreads in food preparation and cooking. Participants were also provided with a list of specific food types and brands to be avoided during the low LA diet phase (based on a cut-off level of < 1 g LA/100 g and/or < 1 g LA/serving size of food product). To facilitate compliance, participants were provided with written materials identifying low LA alternatives for commonly consumed foods to assist them in making appropriate food choices when dining at restaurants or purchasing take away foods. A list of low LA recipes was also provided to participants. These resources were adapted from those produced for participants in a previous low n-6 PUFA trial [18]. All participants were contacted by telephone, email or social networking media every 1–2 weeks for ongoing support and to monitor compliance with weighed food diaries and participants were encouraged to contact study staff at any time during the trial with any questions or concerns.

2.6. Dietary analyses

The diet diaries were analysed for energy intake, macronutrient composition, and fatty acid content including LA, ALA and total n-3 LCPUFA, using the FoodWorks programme (FoodWorks 7 Professional Student, Xyris Software), which uses the latest AUSNUT 2007 food composition tables. These were developed as part of the National Children's Nutrition and Physical Activity Survey, which contains nutrient values for more than 4200 foods, beverages and supplements. The n-3 LCPUFA content of foods was obtained from the AUSNUT 2007 and Australian RMIT Fatty Acids database within the FoodWorks programme and expressed as grams/day and percentage of total energy intake (%E). In cases where information on the LA and ALA content of specific foods was not available on the FoodWorks databases, the LA content of these foods was either estimated based on similar foods, or manually calculated based on the food's main fat source.

2.7. Fatty acid analyses

The fatty acid composition of plasma and erythrocyte phospholipids was analysed using gas chromatography as described in

detail previously [19]. Briefly, total lipids were extracted from plasma and erythrocytes with chloroform/isopropanol and chloroform/methanol (2:1 v/v) [20]. Thin layer chromatography (TLC) on silica gel plates (Silica gel 60H; Merck, Darmstadt, Germany) was then used to separate the phospholipids from total lipid extracts. The lipid classes were visualized under UV light and the phospholipids were transferred into a vial containing 1% (v/v) sulphuric acid (H_2SO_4) in methanol. All phospholipids were transesterified with 1% (v/v) H_2SO_4 in methanol at 70 °C for 3 h. After the samples were cooled, the resulting fatty acid methyl esters (FAME) were extracted in n-heptane and transferred into 2 ml chromatography vials containing 1 to 2 grains of anhydrous sodium sulphate (Na_2SO_4). FAME were then separated and quantified by gas chromatography (GC) (Hewlett-Packard 6890, California, United States of America) using techniques described in detail elsewhere [21]. For statistical purposes, fatty acids at concentrations lower than the limit of detection (0.05%) were allocated a set value of 0.025% (half the limit of detection). All solvents used for extraction and separation contained 0.005% (w/v) of the antioxidant, butylated hydroxyanisol (BHA). All solvents used in this study were purchased from Ajax Firechem Pty Ltd. (Auckland, NZ) or Chem-Supply (South Australia, Australia) and were of analytical grade. Unless specified otherwise, other chemicals and reagents were purchased from Sigma-Aldrich (Missouri, United States of America).

2.8. Statistical analysis

All data are presented as median and interquartile range (IQR). Due to the small sample size and non-normal distribution of a number of dietary intake variables and erythrocyte/plasma fatty acids, data were analysed using non-parametric tests. Dietary intake, plasma and erythrocyte phospholipids fatty acid composition data at baseline and after 4 weeks on the low LA diet were compared using the Wilcoxon signed-rank test for matched pairs. A P value of <0.05 was considered statistically significant.

3. Results

3.1. Subjects

Thirty-six healthy participants (male=12, female=24) were recruited to the study. Of these, three subjects withdrew from the study during the 2 week control phase before the low LA PUFA diet commenced, due either to an inability to provide a blood sample ($n=1$) or because of increased work commitments ($n=2$). A total of 33 participants completed the study, including one participant whose results were excluded due to incomplete dietary records being maintained by the participant. Therefore, the results from the remaining 32 participants were included in the final analysis. The baseline characteristics of the participants are shown in Table 1. There was no change in body weight or BMI after the 4 weeks on the low LA diet compared to weight at baseline (data not shown).

Table 1
Baseline characteristics of participants.

	Sample population
Total number of participants (N)	32
Gender	10 males; 22 females
Age (years)	Range:19–62; Mean ± SD: 31.5 ± 12.3
BMI (kg/m^2)	Range: 17.4–34.7; Mean ± SD: 23.5 ± 3.6

3.2. Dietary data

The mean dietary intakes of energy, macronutrients, saturated fatty acids, monounsaturated fatty acids and n-6 and n-3 PUFA during the 2 week control phase and during the 4-week dietary intervention are presented in Table 2. Participants consumed significantly less total energy and total fat during the low LA diet phase compared to baseline ($P<0.05$). There was no significant difference, however, in the intake of carbohydrate or protein in either g/day or as a percentage of total energy between baseline and the low LA diet phase.

Dietary LA intake decreased from 4.6%E at baseline to 2%E during the low LA diet period ($P<0.001$). Dietary ALA intake also decreased significantly during the low LA intervention compared to baseline ($P<0.001$). There was a small but significant increase in the intake of saturated fat as a percentage of energy during the low LA diet phase compared to baseline ($P<0.05$). MUFA intake did not differ between the baseline and low LA diet periods (Table 2). Importantly, there was no change in n-3 LCPUFA intake during the low LA intervention (Table 2).

3.3. Plasma phospholipid fatty acid profile

Median plasma phospholipid fatty acids at baseline and following 4 weeks on the low LA diet are presented in Table 3. After 4 weeks on the low LA diet, there was a significant decrease in both LA and total n-6 PUFA content of plasma phospholipids compared to the control period ($P<0.001$), but no change in their AA content. There was also no change in dihomo-gamma-linolenic acid (DGLA, 20:3n-6) and saturated fat content of the plasma phospholipids after the low LA intervention (Table 3). There was no change in oleic acid (18:1n-9) content after the low LA intervention, but a small but significant increase in the total monounsaturated content ($P<0.05$, Table 3). The plasma phospholipid content of EPA ($P<0.01$), DPA ($P<0.05$), DHA ($P<0.001$) and total n-3 LCPUFA ($P<0.001$) were all significantly increased after 4 weeks on the low LA diet (Table 3). There was no difference in the ALA content of the plasma phospholipids after 4 weeks on the low LA diet (Table 3).

3.4. Erythrocyte phospholipid fatty acid profile

Median erythrocyte phospholipid fatty acids at baseline and following 4 weeks on the low LA diet are presented in Table 4. Consistent with the plasma phospholipids, after 4 weeks on the low LA diet, there was a significant decrease in both LA and total n-6 PUFA content of erythrocyte phospholipids compared to the

Table 2
Dietary daily intake of energy, macronutrients and fatty acids during the control and low LA diet phase (median interquartile range).

	Control phase	Low LA diet
Total energy (kJ)	7852 (6509–9566)	6734 (5637–8374)*
Protein (g)	82.0 (69.2–99.1)	81.4 (67.3–99.9)
Carbohydrate (g)	205.7 (165.1–243.6)	196.7 (138.4–242.8)
Total fat (g)	63.0 (51.3–83.8)	51.5 (42.1–64.5)*
Monounsaturated fat (%E)	10.8 (9.5–14.4)	11.8 (9.2–14.3)
Saturated fat (%E)	10.8 (8.1–12.7)	11.5 (9.5–13.8)*
n-3 LCPUFA (mg)	235.0 (132.5–365.0)	215.0 (150.0–415.0)
n-3 LCPUFA (%E)	0.12% (0.07–0.20)	0.15% (0.08–0.17)
LA (g)	8.5 (6.6–13.6)	4.2 (9.5–13.8)*
LA (%E)	4.6 (3.5–5.3)	2.0 (1.7–2.6)*
ALA (g)	1.23 (0.81–1.68)	0.57 (0.45–0.81)*
ALA (%E)	0.53 (0.44–0.81)	0.31 (0.27–0.38)*

ALA, α-linolenic acid; LA, linoleic acid.
* $P<0.05$.

Table 3
Plasma phospholipid fatty acid profile at baseline and after 4 weeks on the low LA diet (median interquartile range). All results are expressed as a percentage of the total fatty acid in plasma phospholipids.

		Baseline	Week 4
Monounsaturated fatty acids	Oleic acid (18:1n-9)	10.2 (9.6–11.5)	11.0 (9.6–11.7)
	Total Monounsaturates	13.5 (12.7–14.6)	14.5 (13.1–15.4)*
Saturated fatty acids	Palmitic acid (16:0)	27.5 (26.9–28.1)	27.7 (26.7–28.7)
	Stearic acid (18:0)	13.7 (12.9–14.1)	13.5 (12.7–14.1)
n-6 PUFA	LA (18:2n-6)	21.0 (19.2–22.5)	18.6 (17.3–21.2)***
	DGLA (20:3n-6)	3.05 (2.64–3.44)	3.20 (2.61–3.80)
	AA (20:4n-6)	9.7 (8.6–11.0)	9.9 (9.5–11.3)
	Total n-6 PUFA	35.1 (33.5–36.5)	33.7 (31.8–35.0)***
n-3 PUFA	ALA (18:3n-3)	0.17 (0.14–0.23)	0.19 (0.15–0.22)
	EPA (20:5n-3)	0.87 (0.80–1.18)	1.09 (0.83–1.34)**
	DPA (22:5n-3)	0.88 (0.79–1.01)	0.95 (0.78–1.12)*
	DHA (22:6n-3)	3.47 (3.01–3.90)	3.77 (3.33–4.26)**
	Total n-3 PUFA	5.53 (5.04–6.56)	6.22 (5.57–6.69)***

DGLA, dihomo-gamma-linolenic acid; LA, linoleic acid; DGLA, dihomo-gamma-linolenic acid; AA, arachidonic acid; ALA, α-linolenic acid; EPA, eicosapantaenoic acid; DPA, docosapantaenoic acid; DHA, docosahexaenoic acid.

* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

Table 4
Erythrocyte phospholipid fatty acid profile at baseline and after 4 weeks on the low LA diet (median interquartile range). All results are expressed as a percentage of the total fatty acid in erythrocyte phospholipids.

		Baseline	Week 4
Monounsaturated fatty acids	Oleic acid (18:1n-9)	14.4 (13.5–15.0)	14.2 (14.1–14.6)
	Total Monounsatuates	18.0 (16.6–18.4)	17.9 (17.3–18.4)
	Stearic acid (18:0)	11.4 (10.9–12.3)	11.9 (11.2–12.6)*
n-6 PUFA	LA (18:2n-6)	10.6 (9.71–11.3)	9.41 (8.78–10.5)***
	DGLA (20:3n-6)	1.62 (1.40–1.87)	1.61 (1.37–1.85)
	AA (20:4n-6)	13.4 (12.8–14.1)	13.3 (12.7–14.0)
	Total n-6 PUFA	29.3 (28.7–30.9)	27.9 (27.2–28.7)***
n-3 PUFA	ALA (18:3n-3)	0.14 (0.13–0.15)	0.12 (0.11–0.15)
	EPA (20:5n-3)	0.76 (0.67–0.95)	0.82 (0.67–0.99)
	DPA (22:5n-3)	2.43 (2.19–2.66)	2.40 (2.11–2.61)
	DHA (22:6n-3)	4.67 (4.07–5.50)	4.64 (4.12–5.22)
	Total n-3 PUFA	8.20 (7.67–8.87)	8.01 (7.60–8.66)

DGLA, dihomo-gamma-linolenic acid; LA, linoleic acid; AA, arachidonic acid; ALA, α-linolenic acid; EPA, eicosapantaenoic acid; DPA, docosapantaenoic acid; DHA, docosahexaenoic acid.

* $P < 0.05$.
*** $P < 0.001$.

control phase ($P < 0.001$), but no change in AA or DGLA content. However, in contrast to the plasma phospholipids, there was no difference in erythrocyte phospholipid concentrations of the n-3 LCPUFA, EPA, DPA or DHA following the low LA dietary intervention (Table 4). Stearic acid (18:0) content in the erythrocyte phospholipids increased from 11.4% at baseline to 11.9% ($P < 0.05$) after 4 weeks on the low LA diet. There was no difference in oleic acid or total monounsaturated fatty acid content of erythrocyte phospholipids following the low LA diet (Table 4).

4. Discussion

This study has demonstrated that it is feasible to reduce LA intake in the diet from 4.6%E to 2%E in free-living humans. This is only the second study to demonstrate that n-6 PUFA intakes can be reduced to this extent in an outpatient setting, and the first in which the intervention was based on a combination of dietary advice and provision of study oils and fats, without providing additional study foods. The observed diet-induced reduction in the LA content of plasma and erythrocyte phospholipids without an accompanying decrease in AA content in either blood fraction in the present trial is consistent with previous findings [18,22,23]. Collectively, these results suggest that any positive effects of reduced LA intake on inflammatory pathways are unlikely to be mediated by a reduction in AA. It is possible, however, that lowering LA intake for a 4 week period was insufficient to elicit changes in AA content and that AA content would have decreased had the diet been continued for a more extended period.

Lowering the LA content of the diet for a 4 week period did, however, significantly increase the EPA, DPA and DHA content in plasma, but not erythrocyte phospholipids. Importantly, this occurred in the absence of any increase in n-3 LCPUFA intake, and a reduction in the dietary intake of ALA. The finding of an increase in n-3 LCPUFA status after following a low n-6 LA diet is in agreement with the results of MacIntosh and colleagues who provided participants with a low n-6 PUFA diet for 12 weeks [18], and consistent with the hypothesis that high n-6 diets can limit uptake of dietary n-3 LCPUFA into tissues. The finding that plasma phospholipid n-3 LCPUFA increased despite a significant reduction in dietary n-3 ALA suggests that enhanced incorporation of dietary n-3 PUFA/n-3 LCPUFA, rather than increased conversion from n-3

ALA, likely played the dominant role in increasing n-3 LCPUFA status. This interpretation is supported by the finding that the ALA content of the plasma and erythrocyte phospholipids were not decreased following the 4 weeks on the low LA diet, despite the significant reduction in dietary ALA intake.

In contrast to the effects in plasma, there was no change in EPA, DPA or DHA content of erythrocyte phospholipids after 4 weeks on the low LA diet in this study. This is consistent with the results of two previous 4-week Australian trials [22,23], but differs from that of Macintosh and colleagues, who reported a 51% increase in EPA and 19% increase in DHA content in erythrocyte phospholipids following a low n-6 diet for 12 weeks [18]. Since erythrocytes have a longer half-life than plasma phospholipids, a 4 week period of dietary LA lowering may be insufficient to produce any changes in tissue n-3 status in erythrocyte phospholipids [24]. However, the difference in trial duration cannot completely explain the results, since MacIntosh et al. found substantial increases in EPA and DHA content in erythrocytes after only 4 weeks that were similar in magnitude to those at 12 weeks (Ramsden, unpublished data). Two key differences between the present study and the study conducted by MacIntosh et al. may help explain these results; the Macintosh trial was conducted in a US population which had much higher LA intakes at baseline compared to our Australian population (7.4%E vs. 4.6%E), and the MacIntosh trial maintained baseline dietary n-3 ALA throughout the intervention phase while n-3 ALA intake decreased significantly in the present trial. As a result, reducing LA intake to approximately 2%E in the US trial would be expected to produce a proportionally greater increase in n-3 LCPUFA status due to a more pronounced reduction in competition for esterification into membrane phospholipids and conversion of n-3 ALA to EPA and DHA.

The unintentional decrease in ALA intake in our trial was likely due to the fact that many key dietary sources of ALA (canola oil, nuts and seeds) are also relatively high in LA and were not permitted during the low LA dietary intervention. In addition, the macadamia oil and butter that were used as substitutes for standard vegetable oils and spreads contain very low levels of ALA. This was avoided in the MacIntosh trial by providing a small amount of ground flaxseed to participants during the low n-6 dietary intervention in order to maintain average US ALA intake despite a reduction in high LA vegetable oils that are also major sources of n-3 ALA (e.g. canola, soy) [18].

4.1. Limitations

As with the majority of dietary studies, the accuracy of dietary recording is a potential limitation in the current trial. While weighed food records are generally regarded as one of the higher quality methods of dietary recording, the very act of asking people to record their dietary intake has the potential to alter their dietary choices, both as a result of social desirability bias and potential to favour foods that are easier to weigh/record [25]. However, the significant reduction in LA and total n-6 PUFA content of both the plasma and erythrocyte phospholipids following 4 weeks on the low LA diet provides evidence of good compliance with the diet regimen. The decrease in energy intake and small, but significant, increase in the intake of saturated fat as a percentage of energy during the low LA diet is not unexpected given the restrictions on take-away/processed foods and intake of plant/vegetable based oils, however the potential impact of these changes would need to be considered if a low n-6 LA diet were to be maintained longer term. In the present study, fatty acid status pre- and post- the low LA diet was assessed in the same participant. Thus, while this study provides proof-of-concept that reducing dietary LA intake improves n-3 LCPUFA status, it will be important to confirm these findings in randomized controlled trials which compare changes in separate groups exposed to either a control or low LA diet.

Clearly, the magnitude of change in n-3 LCPUFA status in this 4-week trial is much less than achieved by directly supplementing the diet with n-3 LCPUFA. Therefore, the potential for low LA diets to provide a substitute for n-3 LCPUFA supplementation to raise n-3 LCPUFA status may be limited. However, recent evidence suggests that high n-6 LA intakes have the potential to produce negative health effects through the production of bioactive lipid mediators derived from n-6 LA (e.g. hydroperoxy- and hydroxy-fatty acids), i.e. independent of n-3 LCPUFA status [26–28]. It is therefore possible that low n-6 LA diets may be associated with clinical benefits in spite of relatively modest effects on n-3 LCPUFA status. For example, the bioactive n-6 LA metabolite leukotoxin is reported to contribute to tissue damage [29,30], and the n-6 LA metabolites 9- and 13-HODE are reported to have pro-nociceptive properties [31,32]. Indeed, Ramsden and colleagues recently reported that a combined high n-3 plus low n-6 (H3–L6) dietary intervention reduced the severity of symptoms in chronic headache sufferers [33]. However, the relative importance of lowering dietary n-6 LA and increasing dietary n-3 PUFAs in producing the anti-nociceptive effects is not yet clear. In addition, the issue of sustainability of n-3 LCPUFA supplementation from marine sources has been called into question, and so there is a need to critically examine alternative strategies for maintaining/increasing n-3 LCPUFA status in the population. Thus, low n-6 PUFA diets may indeed have an important role to play in human health.

5. Conclusions

We have demonstrated that it is possible to reduce dietary LA intake to ~2%E in a free-living human population with no provision of foodstuffs apart from a highly monounsaturated oil (macadamia oil) and butter, and without causing an increase in total fat intake. The low LA diet reduced LA and total n-6 PUFA content of plasma and erythrocyte phospholipids. Importantly, the low LA diet also increased the n-3 LCPUFA content of the plasma phospholipids, in the absence of any increase in n-3 LCPUFA intake, almost certainly due to higher incorporation of dietary n-3 LCPUFA. These data support the hypothesis that short-term reductions in dietary LA intake have the potential to improve n-3 LCPUFA status, albeit modestly, without a need to increase dietary n-3 LCPUFA intake. We speculate that the magnitude of the increase in n-3 LCPUFA status would be greater by maintaining or increasing dietary ALA content and/or extending the intervention period beyond 4 weeks, and this remains an important avenue for further studies. In addition, it will be important to determine whether the biochemical changes identified in response to the low LA diet provide clinical benefits, particularly in conditions associated with increased bioactive derivatives of n-6 LA.

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