Fish oil supplementation in patients with and without systemic lupus erythematosus: targeting pro-inflammatory and pro-resolving lipid mediators

Sirs,

Omega-3 fatty acid (ω3FA)-derived "specialised pro-resolving mediators" (SPM) are lipid mediators (LM) central to inflammation resolution. Prior studies suggest SPMs may counter-regulate production of inflammatory mediators and promote inflammation resolution in SLE (1, 2). Fish oil (FO) supplements provide substrates for SPM biosynthesis to enhance endogenous production (3), however, the association of FO supplementation with SPM blood levels in patients with and without SLE, have not been examined.

Within the Massachusetts General Brigham (MGB) Biobank, 16 SLE patients (1997 Updated American College of Rheumatology SLE classification criteria (4)) taking FO were matched (age, sex, race) to 16 non-SLE controls (from outpatient practices, mainly primary care) taking FO (Supplementary Table S1). Another 16 SLE patients not taking FO were matched to 16 non-SLE patients not taking FO. Demographic/clinical data including medication lists recorded by the physicians were obtained by medical record review. MGB Institutional Review Board approved all aspects of this study. Targeted liquid chromatography-tandem spectrometry on plasma quantified 27 LM (Suppl. Fig. S1). Bivariable t-tests and multivariable linear analyses (adjusted for age, race, smoking, body mass index, and medications) examined associations of SLE, FO, and their interactions with LM levels. We adjusted for CRP or ESR and SLEDAI-2K among SLE cases only. Mean SLEDAI-2K for SLE patients on FO *versus* SLE patients not on FO was not statistically different (p=0.27) (Suppl. Table S1). We adjusted for multiple comparisons using false discovery rate (FDR, threshold 0.05, Benjamini and Yekutieli) (SAS v. 9.4 MULTTEST proc).

Higher levels of pro-inflammatory LMs were found in patients with SLE not on FO, *versus* controls not on FO, particularly TXB_2 (p=0.03), an important inflammatory mediator of tissue injury, including renal injury in SLE (5). Of note, AA, TXB_2 precursor, was also higher in SLE not on FO compared to controls not on FO (not statistically significant). It is known that LM formation does not only depend on precursor concentration however. Lower levels of many SPMs were observed among patients with SLE taking FO compared to controls taking FO, including LXB_4 (p<0.01) and 18-HEPE (p=0.04) (Suppl. Fig. S2). It is possible that circulating levels were lower due to consumption by SLE's chronic inflammation. In SLE patient case-only analyses, after adjusting for disease activity and inflammatory markers, 17-HDHA, a potent SPM that reduces pain in murine arthritis models (6), was significantly higher (p=0.05) among patients with SLE taking than not taking FO (Table I), suggesting that high systemic inflammation may decrease FO effects. There were no multiplicative interactions between FO effect and SLE status. After adjustment for multiple comparisons, among control subjects, FO supplementation was a significant predictor for higher EPA (FDR<0.01). The association between FO intake and these LMs in SLE appears less pronounced than in non-SLE patients potentially because SLE patients have chronically depleted pro-resolving SPMs and higher circulating pro-inflammatory SPMs compared to individuals without SLE.

As this study was cross-sectional, we could not directly confirm the causal effect of FO supplements, nor could we examine the effects of different FO preparations, doses, durations of use, or adherence to the supplements as these were not available in the records. It is also possible that some patients were taking FO supplements not listed in the records. The controls may have had other conditions, potentially explaining why few differences were observed between cases and controls.

In conclusion, FO intake did not appear to be associated with LM levels among SLE patients, potentially as a result of a higher burden of inflammation or deficiencies in pro-resolving mechanisms in SLE as suggested by the trend to higher pro-inflammatory eicosanoids and lower pro-resolving SPMs in patients with SLE. Further studies should investigate the effects of ω3FA on SPM production and inflammation in SLE.

Table I. Effects of dietary fish oil supplements on omega-3 fatty acid derived lipid mediators, adjusted for age, race, smoking, BMI, medications (prednisone, hydroxychloroquine and immunomodulators, and statins), elevated ESR/CRP and SLEDAI-2K disease activity among SLE cases.

		Patients with SLE taking *vs*. not taking FO (n=32, 16 of each)		Patients without SLE, taking *vs*. not taking FO (n=32, 16 of each)	
		B coeff. (95% CI)	**p**	**B coeff. (95% CI)**	***p***
Pro-Inflammatory	**AA**	-0.28 (-0.66, 0.09)	0.13	0.33 (-0.02, 0.67)	0.06
	PGD_2	-0.17 (-0.87, 0.53)	0.61	0.46 (-0.12, 1.04)	0.11
	PGE_2	-0.74 (-2.03, 0.55)	0.24	0.31 (-0.34, 0.95)	0.33
	PGF2a	-0.36 (-1.82, 1.11)	0.61	0.63 (-0.64, 1.89)	0.31
	TXB_2	-1.10 (-3.09, 0.88)	0.26	0.07 (-1.42, 1.56)	0.92
	LTB_4	0.43 (-0.23, 1.10)	0.19	0.34 (-0.28, 0.95)	0.27
Pro-Resolving	LXA_4	0.04 (-0.72, 0.80)	0.92	-0.07 (-0.93, 0.79)	0.87
	LXB_4	**-0.73 (-1.41, -0.05)**	**0.04**	0.74 (-0.22, 1.70)	0.13
	EPA	-0.06 (-0.59, 0.48)	0.83	**0.70 (0.40, 1.00)**	**<0.01***
	18-HEPE	0.29 (-0.41, 0.99)	0.40	**0.52 (0.02, 1.02)**	**0.04**
	15-HEPE	0.07 (-0.55, 0.70)	0.81	**0.49 (0.05, 0.94)**	**0.03**
	RvE1	-0.53 (-1.51, 0.46)	0.28	0.34 (-0.35, 1.03)	0.32
	RvE2	-0.05 (-0.65, 0.54)	0.85	-0.53 (-1.52, 0.46)	0.28
	RvE3	0.10 (-0.58, 0.79)	0.75	0.17 (-0.77, 1.11)	0.71
	RvE4	-0.24 (-1.32, 0.85)	0.65	0.83 (-0.41, 2.07)	0.18
	DHA	-0.00 (-0.43, 0.42)	0.99	**0.51 (0.12, 0.89)**	**0.01**
	MaR1	0.13 (-0.51, 0.76)	0.68	**1.07 (0.35, 1.79)**	**<0.01**
	MaR2	-0.29 (-0.63, 0.06)	0.10	-0.07 (-0.40, 0.26)	0.68
	PD1	0.30 (-0.68, 1.28)	0.53	-0.00 (-1.14, 1.13)	0.99
	17-HDHA	**1.11 (0.01, 2.22)**	**0.05**	-0.35 (-1.33, 0.64)	0.47
	14-HDHA	0.16 (-0.68, 0.99)	0.70	**0.57 (0.08, 1.06)**	**0.02**
	RvD1	0.71 (-0.70, 2.12)	0.30	-0.72 (-1.78, 0.33)	0.17
	RvD2	-0.16 (-1.00, 0.69)	0.70	0.29 (-0.10, 0.69)	0.14
	RvD3	-0.25 (-0.80, 0.30)	0.35	0.48 (-0.44, 1.40)	0.29
	RvD4	-0.30 (-1.34, 0.73)	0.55	0.42 (-0.42, 1.26)	0.31
	RvD5	0.37 (-0.32, 1.06)	0.27	0.04 (-0.72, 0.80)	0.91
	RvD6	0.00 (-0.81, 0.82)	0.99	0.25 (-0.73, 1.23)	0.60

*Remained statistically significant after correction for multiple comparisons with false discovery rate (threshold of 0.05). AA: arachidonic acid; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid; LTB4: leukotriene B4; LXA4: lipoxin A4; LXB4: lipoxin B4; MaR1-2: maresin 1-2; PGD2: prostaglandin D2; PGE2: prostaglandin E2; PGF2a: prostaglandin F2 alpha; TXB2: thromboxane B2; PD1: protectin D1; RvD1-6: resolvin D-6; RvE1-4: resolvin E1-4; 14-HDHA: 14-hydroxy-docosahexaenoic acid; 15-HEPE: 15-hydroxyeicosapentaenoic acid; 17-HDHA: 17-hydroxy-docosahexaenoic acid; 18-HEPE: 18-hydroxyeicosapentaenoic acid.

Acknowledgements

We would like to acknowledge the VITAL investigators (Samia Mora, I-Min Lee, JoAnn E. Manson, and Natalya Gomelskaya) and Jessica Lasky-Su for their input on the design and conduct of the study. We would like to thank Emma Stevens for her assistance in data collection, Jack Ellrodt for his technical review of the paper, and Ashley St. Perrier and Denis Reynaud at the Univer-

sity of Toronto (Toronto, Canada) for their technical assistance in the quantification of SPM and LM by targeted LC-MS/MS.

M.Y. Choi[1,2], *MD, FRCPC, MPH*
N.R. Cook[3], *ScD*
G. Kotler[3], *PhD*
C.N. Serhan[4], *PhD, DSc*
R. Tatituri[1], *PhD*
K.H. Costenbader[1], *MD, MPH*

[1]Division of Rheumatology, Inflammation and Immunity, Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, MA, USA,
[2]Division of Rheumatology, Cumming School of Medicine, University of Calgary, Alberta, Canada,
[3]Division of Preventive Medicine, Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, MA, USA; Division of Cardiovascular Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA; Center for Lipid Metabolomics, Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA,
[4]Department of Anesthesia, Perioperative and Pain Medicine, Center for Experimental Therapeutics and Reperfusion Injury, Brigham and Women's Hospital-Harvard Medical School, Boston, MA, USA.

Please address correspondence to:
May Choi,
Division of Rheumatology,
Department of Medicine,
Cumming School of Medicine,
University of Calgary
3230 Hospital Drive NW,
Calgary, NW T2N 4Z6, Canada.
E-mail: may.choi@ucalgary.ca

Funding: this study was supported by NIH R01 AR059086 and NIH K24 AR066109.
Competing interests: M. Choi has received consultancy fees from AstraZeneca, MitogenDx, Mallinckrodt Pharmaceuticals and Glaxo Smith Kline; she is supported by the Lupus Foundation of America Gary S. Gilkeson Career Development Award. The other authors have declared no competing interests.

References

1. NAVARINI L, BISOGNO T, MARGIOTTA DPE *et al.*: Role of the specialized proresolving mediator resolvin D1 in systemic lupus erythematosus: preliminary results. *J Immunol Res* 2018; 2018: 5264195. https://doi.org/10.1155/2018/5264195
2. DAVIS-PORADA J, SERHAN C, NORRIS P, LIPSKY P, SALMON J: 3 Polyunsaturated fatty acids (PUFAs) and specialized pro-resolving mediators (SPMs) are decreased in plasma and serum from SLE patients compared to healthy controls. *Lupus Sci Med* 2019; 6 (Suppl 1): A4-A. http://dx.doi.org/10.1136/lupus-2019-lsm.3
3. WRIGHT SA, O'PREY FM, McHENRY MT *et al.*: A randomised interventional trial of omega-3-polyunsaturated fatty acids on endothelial function and disease activity in systemic lupus erythematosus. *Ann Rheum Dis* 2008; 67: 841-8. https://doi.org/10.1136/ard.2007.077156
4. HOCHBERG MC: Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus. *Arthritis Rheum* 1997; 40: 1725. https://doi.org/10.1002/art.1780400928
5. KELLEY VE, SNEVE S, MUSINSKI S: Increased renal thromboxane production in murine lupus nephritis. *J Clin Invest* 1986; 77: 252-9. https://doi.org/10.1172/jci112284
6. LIMA-GARCIA JF, DUTRA RC, DA SILVA K *et al.*: The precursor of resolvin D series and aspirin-triggered resolvin D1 display anti-hyperalgesic properties in adjuvant-induced arthritis in rats. *Br J Pharmacol* 2011; 164(2): 278-93. https://doi.org/10.1111/j.1476-5381.2011.01345.x