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Pro- and anti-inflammatory eicosanoids in psoriatic arthritis

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Abstract

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AUTHOR CONTRIBUTIONS

MG designed and supervised the overall project. Study conception and design: MG, OQ and AK. **Patients recruitment:** AK, DH, TB. **Acquisition of Mass Spectrometry data:** AMA, SDW, OQ. **Analysis and interpretation of data:** RC, AK, BP, SM, MJ, TP, OQ, MG.

DECLARATIONS

ETHICAL APPROVAL

Patients were enrolled following written informed consent. Ethical approval was granted by the Institutional Review Board (IRB) at UCSD.

CONSENT FOR PUBLICATION:

N/A

AVAILABILITY OF DATA AND MATERIALS

All data generated or analyzed during this study are included in this published article

COMPETING INTERESTS

Roxana Coras declares that she has no conflict of interest.

Arthur Kavanaugh declares that he has no conflict of interest.

Tristan Boyd declares that he has no conflict of interest.

Quyen Huynh declares that she has no conflict of interest.

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Introduction: Eicosanoids are biological lipids that serve as both activators and suppressors of inflammation. Eicosanoid pathways are implicated in synovitis and joint destruction in inflammatory arthritis, yet they might also have a protective function, underscoring the need for a comprehensive understanding of how eicosanoid pathways might be imbalanced. Until recently, sensitive and scalable methods for detecting and quantifying a high number of eicosanoids have not been available.

Objective: Here, we intend to describe a detailed eicosanoid profiling in patients with psoriatic arthritis (PsA) and evaluate correlations with parameters of disease activity.

Methods: Forty-one patients with PsA, all of whom satisfied the CASPAR classification criteria for PsA, were studied. Outcomes reflecting the activity of peripheral arthritis as well as skin psoriasis, Disease Activity Score (DAS)28, Clinical Disease Index (CDAI) and Body Surface Area (BSA) were assessed. Serum eicosanoids were determined by LC-MS, and the correlation between metabolite levels and disease scores was evaluated.

Results: Sixty-six eicosanoids were identified by reverse-phase LC/MS. Certain eicosanoids species including several pro-inflammatory eicosanoids such as PGE₂, HXB₃ or 6,15-dk,dh,PGF_{1a} correlated with joint disease score. Several eicosapentaenoic acid (EPA)-derived eicosanoids, which associate with anti-inflammatory properties, such as 11-HEPE, 12-HEPE and 15-HEPE, correlated with DAS28 (Disease Activity Score) and CDAI (Clinical Disease Activity Index) as well. Of interest, resolvin D1, a DHA-derived anti-inflammatory eicosanoid, was down-regulated in patients with high disease activity.

Conclusion: Both pro- and anti-inflammatory eicosanoids were associated with joint disease score, potentially representing pathways of harm as well as benefit. Further studies are needed to determine whether these eicosanoid species might also play a role in the pathogenesis of joint inflammation in PsA.

Keywords

eicosanoids; lipidomics; biomarkers; psoriatic arthritis

Background

Psoriatic arthritis (PsA) is a heterogeneous disorder with activity and severity ranging from mild synovitis to progressive debilitating erosive disease (Gravallese and Schett, 2018; Ritchlin *et al.*, 2017). The pathophysiology of PsA involves chronic inflammation mediated by pro-inflammatory cytokines, including TNF, IL-17 and IL-23 (Gravallese and Schett, 2018; Ritchlin *et al.*, 2017). It has become clear that many of the signaling pathways triggered by inflammatory cytokines that are activated during inflammation have a profound effect on core lipid metabolism of cells, and the study of the lipidome/metabolome has been successful in identifying new pathogenic targets (Johnson *et al.*, 2016).

Although they represent only a small fraction of the total fatty acids in plasma, free fatty acids are highly metabolically active lipids and they also include the polyunsaturated fatty acids (PUFA): arachidonic acid (AA), linoleic acid (LA) and the nutritionally essential α -linolenic acid (α -LA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA)

(Quehenberger *et al.*, 2010; Quehenberger and Dennis, 2011). Eicosanoids are a class of bioactive lipid mediators derived from the metabolism of AA and related PUFAs by cyclooxygenases (COX), lipoxygenases (LOX), cytochrome P450s, or non-enzymatic pathways (Buczynski *et al.*, 2009). In general, AA gives rise to pro-inflammatory eicosanoids whereas α -LA, EPA and DHA give rise to anti-inflammatory eicosanoids (Buczynski *et al.*, 2009). This class of lipids has been extensively studied over the years due to its implication in the inflammatory response. Some studies suggest that an alteration in AA metabolism is seen in the form of increased formation of pro-inflammatory eicosanoids and decreased production of PUFA-derived anti-inflammatory lipoxins, resolvins, protectins and maresins in several inflammatory diseases (Dennis and Norris, 2015; Serhan, 2014).

Until recently, sensitive and scalable methods for detecting and quantifying a large number of eicosanoids have not been available. Advances in mass spectrometry (MS) analytics now allow for high-throughput rapid and reliable quantification of more than 150 eicosanoids, both pro-and anti-inflammatory, in human plasma. This new technology addresses the question of how eicosanoid pathways may be active and involved in PsA pathogenesis. More important, it allows a more comprehensive, and detailed understanding of how eicosanoid pathways are active, imbalanced, and perturbed.

In this study, we describe the eicosanoid profiling in patients with PsA and its association with joint inflammation. We show that certain pro-inflammatory eicosanoids, such as PGE₂, HXB3 or 6,15-dk,dh,PGF_{1a}, but also some EPA-derived anti-inflammatory eicosanoids, such as 11-HEPE, 12-HEPE and 15 -HEPE, correlated with joint disease. Of interest, resolvin D1, a DHA-derived anti-inflammatory eicosanoid, was down-regulated in patients with high disease activity. These results suggest that an imbalance of pro- and anti-inflammatory eicosanoids might be associated with clinical inflammation in PsA patients and provide a foundation for more focused investigations into novel eicosanoid-based interventions to reduce PsA-related morbidity.

Patients and methods

Patient selection and assessment.

41 adult patients with PsA fulfilling the classification for PsA (CASPAR) criteria were approached in University of California San Diego (UCSD) Arthritis Clinics for opportunistic sampling. The study was approved by the UCSD Institutional Review Board. Clinical assessment included the following: evaluation of the number of tender (TJC) and swollen joints (SJC) (out of 28); the number of tender enthesal sites; the percentage of the body surface affected by psoriasis; functional status as assessed by Health Assessment Questionnaire (HAQ); and assessments of pain; fatigue; global disease severity by patients; and a global assessment of disease by physicians, using a Visual Analogue Scale ranging from 0 to 10. Composite measures of peripheral arthritis were calculated using the above measures: Disease Assessment Score using a 28 joint count and C reactive protein (DAS28-CRP), Clinical Disease Activity Index (CDAI) and Simple Disease Activity Index (SDAI). Non-fasting blood samples were collected in the clinic by research personnel into 10 ml BD Vacutainer Blood Collection Tubes containing spray-coated silica and a polymer gel for serum separation. After 30 min incubation at room temperature, tubes were centrifuged for

10 min at 2000×g and sera were transferred into 1.7ml tubes and immediately frozen and stored at –80 °C until analysis.

Lipid extraction

All sera samples were stored at –80°C, thawed once, and immediately used for free fatty acid and eicosanoid isolation as described (Wang *et al.*, 2014). Briefly, 50 µl sera was spiked with a cocktail of 26 deuterated internal standards that also included some selected PUFAs (individually purchased from Cayman Chemicals, Ann Arbor, MI) and brought to a volume of 1 ml with 10% methanol. The samples were then purified by solid phase extraction on Strata-X columns (Phenomenex, Torrance, CA), using an activation procedure consisting of consecutive washes with 3 ml of 100% methanol followed by 3 ml of water. The eicosanoids were then eluted with 1 ml of 100% methanol, and the eluent was dried under vacuum, dissolved in 50 µl of buffer A (consisting of water-acetonitril-acetic acid, 60:40:0.02 (v/v/v)), and immediately used for analysis).

LC-MS measure of eicosanoids

Eicosanoids in sera were analyzed and quantified by LC/MS/MS as previously described (Quehenberger *et al.*, 2010; Wang *et al.*, 2014). The methods used are in line with the proposed minimum reporting standards published in 2007 by Sumner et al (Sumner *et al.*, 2007). Briefly, eicosanoids were separated by reverse-phase chromatography using a 1.7 µm 2.1 × 100 mm BEH Shield Column (Waters, Milford, MA) and an Acquity UPLC system (Waters). The column was equilibrated with buffer A, and 10 µl of sample was injected via the autosampler. Samples were eluted with a step gradient starting with 100% buffer A for 1 min, then to 50% buffer B (consisting of 50% acetonitril, 50% isopropanol, and 0.02% acetic acid) over a period of 3 min, and then to 100% buffer B over a period of 1 min. The LC was interfaced with an IonDrive Turbo V ion source, and mass spectral analysis was performed on a triple quadrupole AB SCIEX 6500 QTrap mass spectrometer (AB SCIEX, Framingham, MA). Eicosanoids were measured using electrospray ionization in negative ion mode and multiple reaction monitoring (MRM), using the most abundant and specific precursor ion/product ion transitions to build an acquisition method capable of detecting 158 analytes and 26 internal standards. The ionspray voltage was set at –4,500 V at a temperature of 550°C. Collisional activation of the eicosanoid precursor ions was achieved with nitrogen as the collision gas with the declustering potential, entrance potential, and collision energy optimized for each metabolite. Eicosanoids were identified by matching their MRM signal and chromatographic retention time with those of pure identical standards.

Eicosanoids and free fatty acids were quantitated by the stable isotope dilution method. Briefly, identical amounts of deuterated internal standards were added to each sample and to all the primary standards used to generate standard curves. To calculate the amount of eicosanoids and free fatty acids in a sample, ratios of peak areas between endogenous metabolite and matching deuterated internal standards were calculated. Ratios were converted to absolute amounts by linear regression analysis of standard curves generated under identical conditions (Loomba *et al.*, 2015). Eicosanoid levels are shown as pmol/ml.

Data Analysis

The data, consisting of 41 patient samples measured across clinical outcomes were processed using R, Version 3.4.1. (www.r-project.org). Continuous variables were expressed as mean $\pm$ standard deviation (SD) and the categorical variables as percentage. Pearson correlation was used to check correlation between disease activity scores. T-test was used to analyze statistically significant difference between the means of two groups, and the comparisons were then adjusted for BMI, therapy, and disease activity, by including the latter as covariates in a logistic regression model. Hierarchically clustered heatmaps were generated for correlations between each eicosanoid and clinical outcomes. Dendrograms were divided into flat clusters using a cophenetic distance metric. Linear regression was performed between each eicosanoid metabolite- clinical outcome pair, controlling for patient BMI, therapy, and disease activity using the Ordinary Least Square method. Normally distributed independent variables were standardized to a mean of 0 and a standard deviation of 1. Bejamini-Hochberg method was used to adjust for multiple comparisons.

Results

Patient demographics and disease characteristics

Patient characteristics are summarized in Table 1. Of the 41 PsA patients included in this study, 61% were males (N=25) and the mean age was 49.2 ± 11.1 years. The mean number of tender joints and swollen joints were 1.7 ± 3 (range 0 to 10) and 2.1 ± 3.2 (range 0 to 10) respectively. The average DAS28-CRP score was 2.72 ± 1.3 (range 1–5.2). Twenty-seven patients (71%) had active skin disease, with an average BSA of 7.3 ± 15.7 (range 0–95%). Eight patients had enthesitis. The average body mass index (BMI) was 27.4 ± 5.5 . Sixty-five percent were receiving biological therapy, 56.5% of them in association with a synthetic disease modifying antirheumatic drug (sDMARDs), mostly methotrexate. 14.6% received sDMARDs as monotherapy, and 29.2% were on daily non-steroidal anti-inflammatory drugs (NSAIDs). Comorbidities are also summarized in Table 1.

Eicosanoid profiling and clustering

Sixty-six eicosanoids, which are derived from AA, EPA, LA, DGLA, α LA and DHA, were identified by Reverse-phase LC/MS. Among these, 18 (22.38%) belong to the cyclooxygenase (COX) pathway, 12 (11.94%) belong to the 5 lipoxygenase (LOX) pathway, 13 (23.88%) belong to the 15LOX pathway, 7 (13.43%) belong to the 12LOX pathway, 14 (20.89%) belong to the CYP pathway and the rest (2) are synthesized by non-enzymatic pathways. Twenty-seven of the detected eicosanoids are considered pro-inflammatory and 39 are anti-inflammatory species. Figure 1A shows the pro- (in red) and anti-inflammatory (in blue) eicosanoids and the different enzymatic eicosanoid pathways of the eicosanoids detected in our samples. We also analyzed eicosanoid clustering (Figure 1B, C). Concentrations of the circulating eicosanoids detected in our samples are shown in Table 2.

Linear regression analysis between grouped serum eicosanoids and clinical parameters of PsO and PsA.

Linear regression was performed between each eicosanoid-clinical parameter pair, controlling for BMI, NSAIDs and biological therapy, by including these factors as covariates in the model. We also adjusted joint disease scores (TJC, SJC, DAS28, CDAI and SDAI) for BSA, and BSA score was also adjusted for CDAI. The regression coefficients for each eicosanoid-clinical parameter pair were used to form a clustered heatmap to lend insight into which groups of eicosanoids were correlated with which clinical parameters (Figure 2A). p-values and q-values of the regression analysis of metabolite levels are displayed in Figure 2B and Supplementary Figure 1A, where the row and column order are preserved from Figure 2A. Several eicosanoids positively correlated with parameters of joint disease activity (tender joint count -TJC-, swollen joint count -SJC-, CDAI, SDAI and DAS28-CRP) including several pro-inflammatory eicosanoid such as PGE2, 12-oxo-HETE, HXB3 or 6,15-dk,dh,PGF1a, but interestingly also several anti-inflammatory eicosanoids such as 11-HEPE, 12-HEPE and 15-HEPE among others. Interestingly, we also detected some eicosanoids that negatively correlated with joint disease activity including some anti-inflammatory eicosanoids such as 8,9-diHETrE, 11,12-diHETrE,14,15-diHETrE, 19,20-diHDPA and 7,17 DHDPA. Other clinical scores that cluster together were patients' clinical scores (pain, HAQ, fatigue and global patient score), with a similar eicosanoid profiling as CDAI, TJC and TJC.

Logistic regression analysis between grouped serum eicosanoids and DAS28CRP

We further analyzed whether some eicosanoids could differentiate between high and low disease activity scores. Figure 3A, B and Supplemental Table S1 and Figure S1B show logistic regression analysis adjusted by BMI, BSA, NSAIDs and biological therapy of each eicosanoid in patients in remission or with low disease activity (DAS28CRP ≤ 2.32), compared to patients with high disease activity (DAS28CRP > 2.32). Figure 3C shows the significant eicosanoids within the different PUFA and enzymatic pathways. Of interest, while most of the EPA-derived anti-inflammatory eicosanoids were elevated in the high disease activity, some of the DHA- anti-inflammatory eicosanoid, including resolvin D1 and 17-HDoHE, were down-regulated in these group of patients.

Discussion

Data regarding the role of eicosanoids in PsA is very scarce, as most of the research is associated with rheumatoid arthritis (RA) pathogenesis. Evidence that the COX pathway might be involved in the pathogenesis of RA dates back to the 1970s, when elevated prostaglandin levels were reported in synovial fluid from patients with RA (Korotkova and Jakobsson, 2014). Since then, studies in animals and patients have established a pivotal and complex role of prostaglandins and other eicosanoids in RA. For instance, expression of 5LOX and 15LOX was also increased in rheumatoid synovium (Gheorghe *et al.*, 2009). LTB₄, a metabolite of AA via 5LOX, has been related to inflammation in arthritis for a long time. Two independent studies demonstrated that LTB₄ and BLT1 receptors are indispensable for the development of arthritis in the K/BxN model of RA (Chen *et al.*, 2006; Kim *et al.*, 2006). Another study demonstrated that inhibition of 5LOX in fibroblast-like

synoviocytes (FLS) and knocking out 5LOX gene in a mouse model of RA decreased inflammatory cytokines expression and paw inflammation (Lin *et al.*, 2014). More importantly, in RA, despite the wide range of sDMARDs and biological therapies that potentially suppress specific inflammatory mechanisms, the COX and LOX pathways remain overexpressed in inflamed tissue in some patients with RA, which might contribute to subclinical inflammation and disease relapse (Korotkova and Jakobsson, 2014).

In PsA, pharmacological therapies begin with NSAIDs, suggesting that at least COX pathway is critical in PsA pathogenesis. However, the role of LOX and cytochrome P450 (CYP) enzymes, which catalyze several AA-derived pro-inflammatory eicosanoids and most of the PUFA-derived anti-inflammatory eicosanoids, is unknown in this disease. In our study, several eicosanoids were positively correlated with parameters of joint disease activity (tender joint count -TJC-, swollen joint count -SJC-, CDAI, SDAI and DAS28CRP) including several AA-derived pro-inflammatory eicosanoids such as PGE₂, HXB3 or 6,15-dk,dh,PGF_{1a}, being HXB3 and tetranor 12-HETE, 12LOX derived eicosanoids. More interestingly several EPA-derived anti-inflammatory eicosanoids, such as 11-HEPE, 12-HEPE and 15-HEPE, were also upregulated in active patients, suggesting that EPA-derived anti-inflammatory eicosanoids amount might be increased to try to balance the inflammation induced by AA-derived pro-inflammatory eicosanoids. Yet, we also detected some DHA-derived anti-inflammatory eicosanoids that did negatively correlate with joint disease activity, including anti-inflammatory eicosanoids such as resolvin (Rv)D1 and 17-HDoHE. RvD1 is one of the pro-resolving lipid mediators shown to actively contribute to the resolution of several inflammatory processes (Serhan, 2014). The injection of RvD3 reduced joint leukocytes as well as paw joint eicosanoids, clinical scores and edema in the K/BxN serum transfer inflammatory arthritis model (Arnardottir *et al.*, 2016). Resolvins are also potential analgesics: systemic treatment with 17-HDoHE, the precursor of RvD, reduced inflammatory pain in an adjuvant-induced arthritis model (Xu and Ji, 2011). The fact that we detected a negative correlation between these pro-resolving lipids mediators and joint activity could suggest that a disbalance between pro- and anti-inflammatory eicosanoid species might play a role in the pathogenesis of joint inflammation in PsA and that these could be used for the treatment of inflammatory arthritis.

Although these findings are certainly promising, this study is not without limitations, the heterogeneity of our PsA patients being the main limitation. Patients had long-standing disease and were exposed to various therapies prior to the study which could represent activated pathways resistant to current DMARDs and biological therapies. Confirmation of our results with a larger sample size from prospective cohorts of patients with new onset inflammatory arthritis before treatment initiation is necessary to strengthen our conclusions. Comparison with other arthritides would help to determine if the described eicosanoid changes are specific to PsA or secondary to systemic inflammation. Yet, we believe that this work can improve our limited understanding of role of eicosanoids beyond arachidonic acid metabolites and leukotrienes in PsA and may lay the groundwork for a more targeted investigation of novel eicosanoid- and lipidomics-based studies in PsA.

Conclusions

Sensitive and scalable methods for detecting and quantifying a high number of eicosanoids are now available. In this study 66 eicosanoids were identified, and both pro- and anti-inflammatory eicosanoids were associated with joint disease score, potentially representing pathways of harm as well as benefit. Further studies are needed to determine whether these eicosanoid species might also play a role in the pathogenesis of joint inflammation in PsA.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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ABBREVIATIONS

(PsA)	Psoriatic arthritis
(RA)	Rheumatoid arthritis
(DAS)28	Disease Activity Score
(CDAI)	Clinical Disease Index
(BSA)	Body Surface Area
(EPA)	Eicosapentaenoic acid
(PUFA)	Polyunsaturated fatty acids
(AA)	Arachidonic acid
(LA)	Linolenic acid
(α-LA)	α -linolenic acid
(DHA)	docosahexaenoic acid
(COX)	cyclooxygenases
(LOX)	lipoxygenases
(SD)	standard deviation
(sDMARDs)	Synthetic disease modifying antirheumatic drug
(NSAIDs)	Non-steroidal anti-inflammatory drugs

(TJC)	Tender joints
(SJC)	Swollen joints
(HAQ)	Health Assessment Questionnaire
(FLS)	Fibroblast-like synoviocytes
(CRP)	C-reactive protein

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Fig. 1. Pro-inflammatory and anti-inflammatory eicosanoids.

A) Pro-inflammatory and anti-inflammatory eicosanoids detected in our patients, divided by pathways and origin PUFA. In grey, metabolites not identified by MS COX – cyclooxygenase; LOX – lipoxygenase; CYP – cytochrome P450; NE – non-enzymatic; PGFS – prostaglandin F synthase; PGES – prostaglandin E synthase; PGDS – prostaglandin D synthase; PGIS – prostaglandin I synthase; TXAS – thromboxane A2 synthase; LTAH – leukotriene A4 hydrolase; MDB – membrane dipeptidase; HEDH - hydroxyeicosanoid dehydrogenase; PGDH - hydroxyprostaglandin dehydrogenase; 13-PGR – 15-ketoprostaglandin₁₃ reductase; sEH – soluble epoxide hydrolase. B) Heat map and hierarchical cluster analysis indicate positive relationships between circulating eicosanoids identified in blood from PsA patients (the heatmap was created using regression coefficients adjusted for BMI, BSA, NSAIDs and biological therapy. C) Eicosanoid regression p-values are displayed in C, where the row and column order are preserved from B

Fig. 2. Clinical score correlations with eicosanoids.

A) Linear regression was performed between each clinical score – eicosanoid pair, controlling for BMI, NSAIDs and biological therapy. We also adjusted joint disease scores (TJC, SJC, DAS28, CDAI and SDAI) for BSA, and BSA score was also adjusted for CDAI. Other factors, including comorbidities, gender and age were not found to influence eicosanoid levels and were not included in the model. The regression coefficients for each pair were used to form a clustered heatmap, to lend insight into which clinical scores were correlated with which groups of eicosanoids. Row clusters have been identified by cophenetic cutting of the row dendrogram. B) Metabolite regression p-values are displayed in Figure 2B, where the row and column order are preserved from Figure 2A TJC: tender joint count; SJC: swollen joint count; CDAI: clinical Disease Activity Index; SDAI: simple Disease Activity Index; HAQ: Health Assessment Questionnaire; GMD: physician global assessment; GIPT: patient global assessment; BMI: body mass index; BSA: body surface

area; DAS28CRP: disease assessment score 28; NSAIDS: non-steroidal anti-inflammatory drugs

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A

Pathway	PUFA	Eicosanoid	Mean (SD) pmol/ml DAS28CRP ≤ 2.32	Mean (SD) pmol/ml DAS28CRP > 2.32	P	p adjusted
COX	AA	PGF2a	1.6 (1.3)	0.7 (0.7)	0.01	0.03
		PGE2	0.7 (0.6)	1.4 (1.5)	0.08	0.04
		6,15dk-,dh-PGF1a	3.1 (3.6)	7.8 (8.8)	0.03	0.05
		dhk PGD2	15.1 (6.9)	11.5 (3.6)	0.05	0.06
		12-HHTrE	83.7 (81.9)	62.2 (74.4)	0.39	0.04
12LOX	EPA	TXB3	0.2 (0.3)	0	0.02	0.05
	AA	tetranor 12-HETE	9.4 (36.2)	12.4 (22.5)	0.76	0.03
		HXB3	4.1 (5.4)	8.6 (7.6)	0.04	0.06
Non-enzymatic	AA	11-HETE	4.6 (9.8)	5.4 (4.9)	0.74	0.02

B

Pathway	PUFA	Eicosanoid	Mean (SD) pmol/ml DAS28CRP ≤ 2.32	Mean (SD) pmol/ml DAS28CRP > 2.32	P	p adjusted
COX	AA	PGA2	0.4 (0.7)	0.1 (0.2)	0.11	0.02
		PGJ2	0.1 (0.2)	0 (0.1)	0.07	0.02
5LOX	DHA	20-HdoHE	1.3 (3.8)	2.9 (3.8)	0.21	0.08
12LOX	AA	12-HEPE	15.7 (26.2)	38.7 (46.6)	0.06	0.05
	DHA	14-HdoHE	59.9 (114.3)	129.1 (165)	0.13	0.03
15LOX	EPA	15-HEPE	0.5 (0.8)	0.7 (0.8)	0.34	0.09
	LA	13-HODE	48.1 (50.3)	43.7 (58.4)	0.8	0.07
	ALA	13-HoTrE	1.9 (3.6)	4.1 (5.6)	0.16	0.05
		13-HoTrE(y)	0.5 (1.1)	0.7 (1.3)	0.6	0.04
	DHA	17-HdoHE	3 (12.6)	2.9 (6.3)	0.97	0.08
CYP	AA	Resolvin D1	0.3 (0.2)	0.2 (0.3)	0.52	0.04
		11,12 EET	0.1 (0.1)	0 (0.1)	0.29	0.04
		5,6 diHETrE	1.7 (2.1)	0.7 (0.8)	0.07	0.04
		DHA	19,20-diHDPA	1.7 (0.5)	1.3 (0.8)	0.12
Non-enzymatic	EPA	18-HEPE	0.3 (0.5)	0.5 (0.6)	0.19	0.05
		11-HEPE	0.3 (0.7)	0.7 (0.7)	0.09	0.02
	DHA	10-HdoHE	1.2 (2.5)	2.2 (2.7)	0.24	0.03
		13-HdoHE	0.4 (0.8)	0.2 (1)	0.49	0.06

C

Fig. 3. Pro- and anti-inflammatory eicosanoids associated with DAS28-CRP.

Logistic regression was performed between each eicosanoid (pmol/ml) in patients with DAS 2.32 compared with patients with DAS > 2.32. A) Pro-inflammatory eicosanoids with p value < 0.1 after adjusting for BMI, BSA, NSAIDs and biological therapy. B) Anti-inflammatory eicosanoids with p value < 0.1 after adjusting for BMI, BSA, NSAIDs and biological therapy. Other factors, including comorbidities, gender and age were not found to influence eicosanoid levels and were not included in the model. C) Significant eicosanoids (p < 0.1) are circled in green if upregulated in patients with DAS > 2.32, and in red if downregulated in these patients. DAS28: disease activity score; NSAIDs: non-steroidal anti-inflammatory drugs

Table 1

Demographic and disease characteristics of patients included in our study

Characteristic	Percentage/Mean (Standard Deviation)
Male	61%
Age	49.2 (11.1)
BMI	27.4 (5.5)
Duration of PsA in years	11.3 (10)
Duration of psoriasis in years	18.4 (14.1)
Number of tender joints	1.7 (range 0–10)
Number of swollen joints	2.09 (range 0–10)
Pain	2.8 (2.8)
Fatigue	3.3 (3.3)
HAQ	0.45 (0.55)
DAS28-CRP	2.72 (1.28)
CDAI	9.1 (9.6)
SDAI	10.84 (10.25)
Enthesitis	19.5%
Body surface affected by psoriasis (BSA)	7.3 (range 0–95)
Treatment with TNF inhibitors	56%
Diabetes Mellitus type 2	17%
High Blood Pressure	44%
Dyslipidemia	29.2%

Table 2.

Mean (SD) of each circulating eicosanoid detected in 41 PsA patients

Eicosanoid	Mean (SD) pmol/ml
TxB2	27.332 (32.049)
PGF2a	1.269 (1.286)
PGE2	1.029 (1.232)
6,15dk-,dh-PGF1a	5.435 (6.886)
dhk PGF2a	0.813 (1.018)
dhk PGD2	13.341 (5.663)
tetranor-PGFM	1.609 (3.016)
tetranor-PGEM	26.919 (63.712)
9-HETE	9.747 (13.861)
TXB1	0.278 (0.424)
TXB3	0.133 (0.244)
LTB4	0.31 (0.455)
LTE4	0.139 (0.182)
5-HETE	5.242 (6.475)
5-oxoETE	0.203 (0.505)
9-HODE	21.767 (20.02)
9-oxoODE	5.177 (4.807)
12-HETE	581.357 (988.653)
12-oxoETE	3.246 (7.153)
HXB3	7.054 (7.689)
8-HETE	7.764 (12.175)
8,15-diHETE	0.619 (1.171)
15-oxoETE	0.349 (0.852)
9,10 EpOME	4.258 (2.809)
12,13 EpOME	5.052 (4.786)
9,10 diHOME	8.081 (8.241)
12,13 diHOME	8.804 (7.916)
PGA2	0.263 (0.595)
PGJ2	0.127 (0.229)
12-HHTrE	71.954 (76.621)
11-HETE	5.961 (8.577)
9-HEPE	3.316 (5.152)
11-HEPE	0.67 (1.049)
13 HDoHE	0.43 (0.993)
5-HEPE	1.059 (2.126)
8-HEPE	2.999 (2.862)
9-HOTrE	1.656 (1.477)

Eicosanoid	Mean (SD) pmol/ml
5-HETrE	0.176 (0.463)
16 HDoHE	4.736 (6.834)
20 HDoHE	2.585 (4.27)
tetranor 12-HETE	14.622 (38.727)
12-HEPE	32.511 (45.631)
11 HDoHE	6.285 (9.566)
14 HDoHE	109.937 (162.784)
15-HETE	14.395 (21.968)
15-HEPE	0.877 (1.484)
13-HODE	53.761 (64.177)
13-HOTrE	3.588 (5.313)
13-HOTrE(y)	0.795 (1.427)
8-HETrE	0.841 (1.184)
15-HETrE	1.903 (2.789)
4 HDoHE	0.529 (1.323)
17 HDoHE	4.201 (11.148)
Resolvin D1	0.273 (0.269)
5,6-EET	0.113 (0.276)
8,9-EET	2.294 (5.015)
11,12-EET	0.127 (0.335)
14,15-EET	0.215 (0.397)
5,6-diHETrE	1.894 (3.549)
8,9-diHETrE	0.983 (1.393)
11,12-diHETrE	0.822 (0.337)
14,15-diHETrE	1.181 (0.41)
19,20 DiHDPA	1.55 (0.735)
7,17 dHDPA	1.073 (0.782)
18-HEPE	0.546 (0.768)
10 HDoHE	2.03 (3.004)