Angiogenic cytokines can reflect synovitis severity and treatment response to biologics in rheumatoid arthritis

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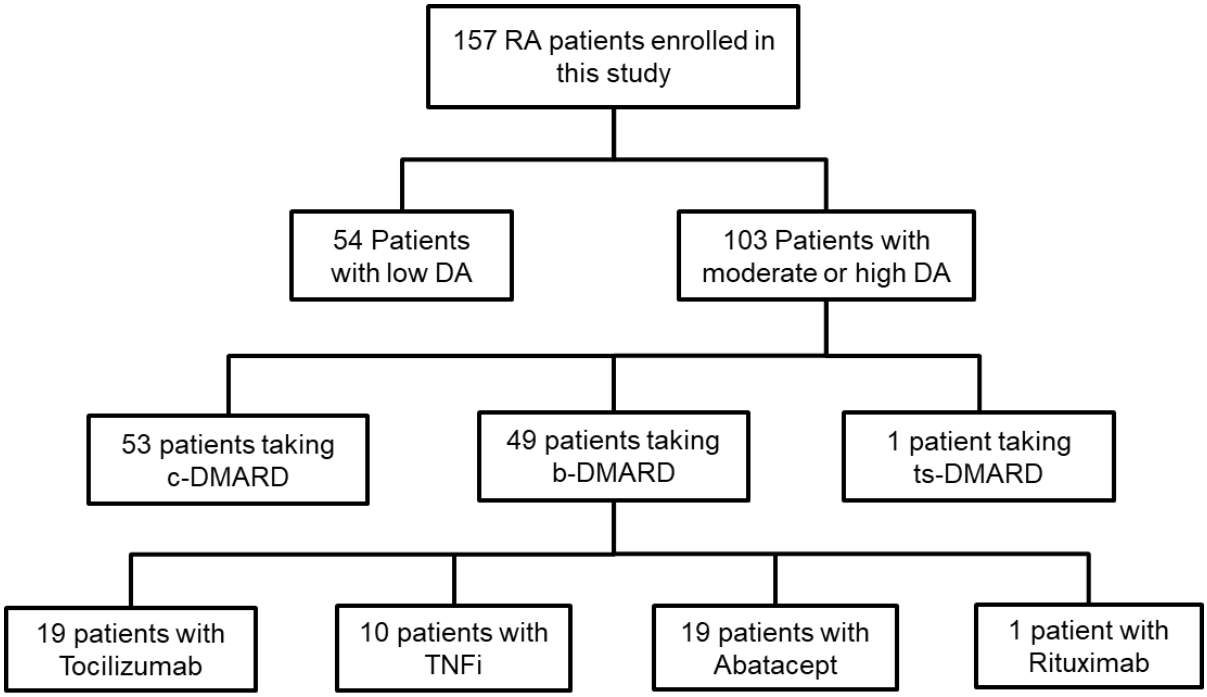
Supplementary figure legends

Supplementary Fig. 1. Flow chart describing treatment regimen of RA patients enrolled for serum sampling. DA, disease activity; c-DMARD: conventional disease-modifying anti-rheumatic drug; b-DMARD: biologic disease-modifying anti-rheumatic drug; ts-DMARD: targeted synthetic disease-modifying anti-rheumatic drug; and TNFi: tumor necrosis factor inhibitor.

Supplementary Fig. 2. A representative of ultrasonographic findings demonstrating synovial hypertrophy and hyper-vascularity. (**a** and **b**) Grey scale US images showing synovial hypertrophy in proximal interphalangeal (PIP) joints. (**c** and **d**) Power-Doppler US images showing vascularity of the synovium in PIP joints.

Supplementary Fig. 3. Correlations of △DAS28 with △ESR, △CRP, △VEGF, and △IL-6 levels in patients treated with c-DMARD (a), b-DMARD (b), Tocilizumab (TCZ) (c), or non-TCZ biologics (d).



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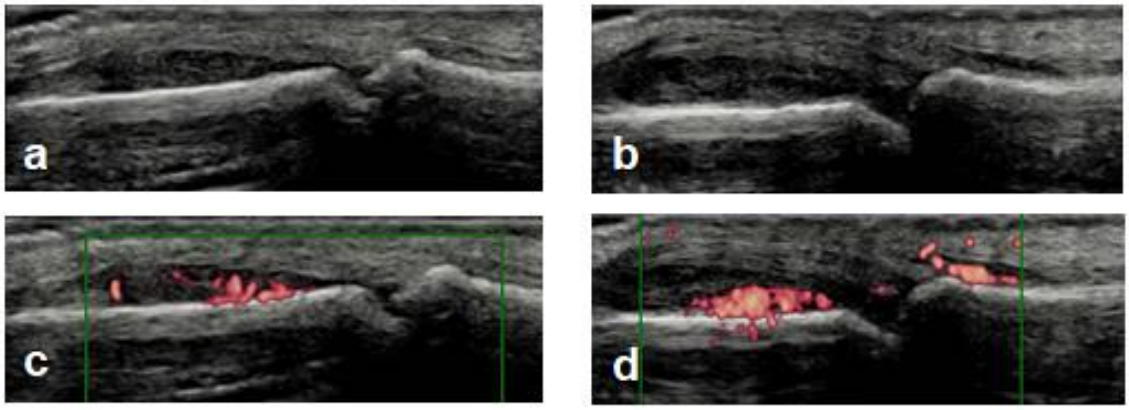

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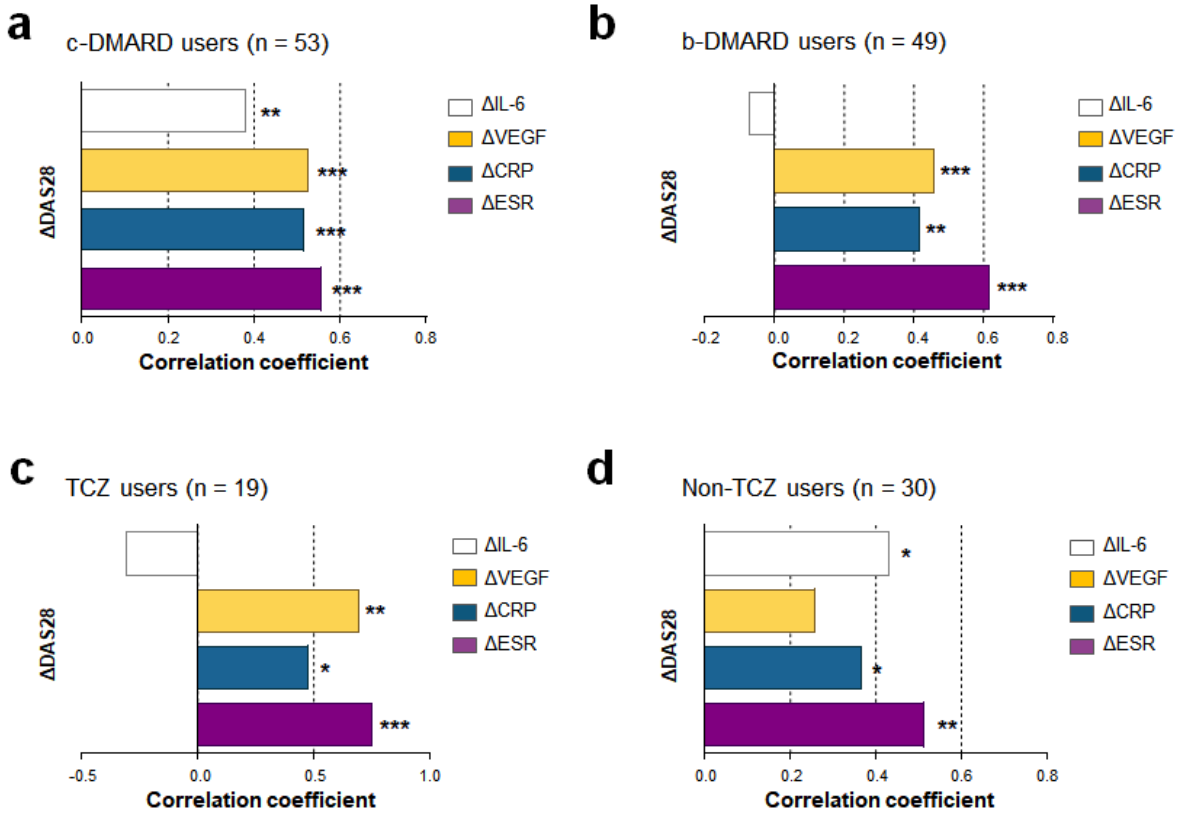

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Supplementary Table 1. Characteristics of RA patients whose synovial fluids were obtained

Variable	Total patients (n=54)
Age, years	59.3 (12.4)
Female, n (%)	51 (94.4)
Serum RF, IU/mL	59.6 (7.5-173.7)
Serum ACPA, U/mL	84.0 (5.4-291.3)
Blood WBC count, /μL	7,390 (6,373-8,958)
Blood % neutrophil	65.1 (10.6)
Hemoglobin, g/dL	12.3 (1.4)
ESR, mm/hour	22.5 (12.8-39.3)
CRP, mg/dL	0.89 (0.20-2.08)
WBC count in the SF, /μL	11,700 (4,275-17,200)
% neutrophil in the SF	63.0 (43.0-75.5)
Glucocorticoid use, n (%)	46 (85.2)
Glucocorticoid dose, PD equivalent, mg/day	5.0 (2.5-7.5)
Methotrexate use, n (%)	28 (51.9)
GSUS grade ≥2, n (%)	31 (57.4)
PDUS grade ≥1, n (%)	22 (40.7)
Active synovitis (GSUS ≥2 or PDUS ≥1), n (%)	32 (59.3)

Data are expressed as mean (standard deviation) or median (interquartile range) for continuous variables. RF: rheumatoid factor; ACPA: anti-citrullinated peptide antibodies; WBC: white blood cell; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; SF: synovial fluid; PD: prednisone; GSUS: grey scale ultrasound; and PDUS: power-Doppler ultrasound.

Supplementary Table 2. Baseline characteristics of RA patients whose sera were obtained

Variable	Total (n=157)	Low DA (n=54)*	Moderate or High DA (n=103)*	p value*
Age, years	56 (11.6)	55.9 (11.8)	56 (11.6)	0.951
Female, n (%)	136 (86.6)	45 (83.3)	91 (88.3)	0.380
BMI, kg/m^2	23 (20.8-24.4)	22.8 (20.5-23.9)	23.1 (21-24.9)	0.125
Duration of disease, years	5 (1-13.5)	7 (3-12.3)	4 (0-15)	**0.026**
ESR, mm/hour	26 (13-44)	12 (3.8-21)	36 (20-52)	**<0.001**
CRP, mg/dL	0.42 (0.08-1.42)	0.06 (0.02-0.21)	0.90 (0.34-2.0)	**<0.001**
RF, IU/mL	99 (33.7-362.7)	69.7 (19.9-156.2)	121 (40.4-476.7)	**0.015**
ACPA, U/mL	171.3 (37-340)	200 (42.3-340)	163.5 (34.9-340)	0.949
Glucocorticoid use, n (%)	110 (70.1)	39 (72.2)	71 (68.9)	0.669
PD equivalent, mg/day	2.5 (0-5)	2.5 (0-5)	2.5 (0-5)	0.772
Previous c-DMARD use, n (%)	135 (86.0)	51 (94.4)	84 (81.6)	**0.027**
MTX dose, mg/week	7.5 (0-10)	7.5 (0-10)	7.5 (0-10)	0.532
Previous b-DMARD use, n (%)	28 (17.8)	19 (35.2)	9 (8.7)	**<0.001**

Data are expressed as mean (standard deviation) or median (interquartile range) for continuous variables. *Comparison of characteristics between patients with low DA and those with moderate or high DA by student's unpaired *t* test. DA: disease activity; BMI: body mass index; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; RF: rheumatoid factor; ACPA: anti-citrullinated peptide antibodies; PD: prednisone; c-DMARD: conventional disease-modifying anti-rheumatic drug; MTX: methotrexate; and b-DMARD: biologic disease-modifying anti-rheumatic drug.

Supplementary Table 3. Baseline characteristics of RA patients treated with c-DMARD versus b-DMARD

Variable	c-DMARD users (n=53)	b-DMARD users (n=49)	p value
Age, years	55.9 (11.9)	56.0 (11.5)	0.960
Female, n (%)	46 (86.8)	44 (89.8)	0.638
BMI, kg/m^2	23.1 (21.3-25.1)	23.1 (20.8-24.5)	0.467
Duration of disease, years	2 (0-14.5)	5 (1-16)	0.082
ESR, mm/hour	32 (19-46)	43 (25-60)	**0.024**
CRP, mg/dL	0.78 (0.33-1.84)	1.03 (0.37-2.53)	0.269
Baseline DAS28	4.6 (0.9)	5.0 (1.1)	**0.030**
RF, IU/mL	129.9 (28.3-484.6)	138.1 (47.8-528.6)	0.350
ACPA, U/mL	165 (32.3-340)	152 (35.2-340)	0.525
vdH Sharp score	8 (0-33)	17 (5-38.5)	0.090
Glucocorticoid use, n (%)	31 (58.5)	40 (81.6)	**0.011**
PD equivalent, mg/day	2.5 (0-5)	5 (2.5-5)	**0.019**
MTX dose, mg/week	7.5 (0-10)	10 (7.5-12.5)	**0.045**

Data are expressed as mean (standard deviation) or median (interquartile range) for continuous variables. c-DMARD: conventional disease-modifying anti-rheumatic drug; b-DMARD: biologic disease-modifying anti-rheumatic drug; BMI: body mass index; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; DAS28: disease activity score 28; RF: rheumatoid factor; ACPA: anti-citrullinated peptide antibodies; vdH: van der Heijde; PD: prednisone; and MTX: methotrexate.

Supplementary Table 4. Baseline characteristics of RA patients according to their treatment response to DMARD

Variable	Good or moderate response (n=82)	No response (n=21)	p value
Age, years	57.3 (11.0)	50.9 (12.8)	**0.022**
Female, n (%)	70 (85.4)	21 (100)	0.120
BMI, kg/m^2	23.2 (21.1-25.1)	22.3 (20.8-24.9)	0.466
Duration of disease, years	4 (0-15)	2 (0-13)	0.340
ESR, mm/hour	36 (19-52.3)	38 (23.5-50.5)	0.863
CRP, mg/dL	0.84 (0.34-2.09)	1.09 (0.36-1.86)	0.797
Baseline DAS28	4.9 (1.0)	4.5 (1.0)	0.087
RF, IU/mL	119.2 (37.9-499.8)	129.9 (49.5-405.1)	0.925
ACPA, U/mL	147 (28-340)	198.5 (141.9-340)	0.130
vdH Sharp score	10 (4-35)	12 (3-42)	0.937
Treatment			
Glucocorticoid use, n (%)	60 (73.2)	11 (52.4)	0.066
PD equivalent, mg/day	2.5 (0-5)	2.5 (0-5)	0.333
c-DMARD users, n (%)	40 (48.8)	13 (61.9)	0.283
b-DMARD users, n (%)	41 (50.0)	8 (38.1)	0.330
Baseline VEGF	0.028 (0.006-0.064)	0.028 (0.001-0.042)	0.519
Baseline IL-6	0.007 (0.003-0.018)	0.004 (0.002-0.016)	0.202
△VEGF (Baseline-F/U)	0.010 (0, 0.031)	-0.001 (-0.015, 0.016)	**0.011**
△IL-6 (Baseline-F/U)	0.003 (-0.003, 0.011)	0 (-0.005, 0.009)	0.494
△ESR (Baseline-F/U)	17 (9, 36.5)	6 (-3.5, 22)	**0.005**
△CRP (Baseline-F/U)	0.62 (0.10, 1.77)	0.61 (0.02, 1.17)	0.307

Data are expressed as mean (standard deviation) or median (interquartile range) for continuous variables. BMI: body mass index; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; DAS28: disease activity score 28; RF: rheumatoid factor; ACPA: anti-citrullinated peptide antibodies; vdH: van der Heijde; PD: prednisone; c-DMARD: conventional disease-modifying anti-rheumatic drug; b-DMARD: biologic disease-modifying anti-rheumatic drug; VEGF: vascular endothelial growth factor; IL-6: interleukin-6; and F/U: follow-up.