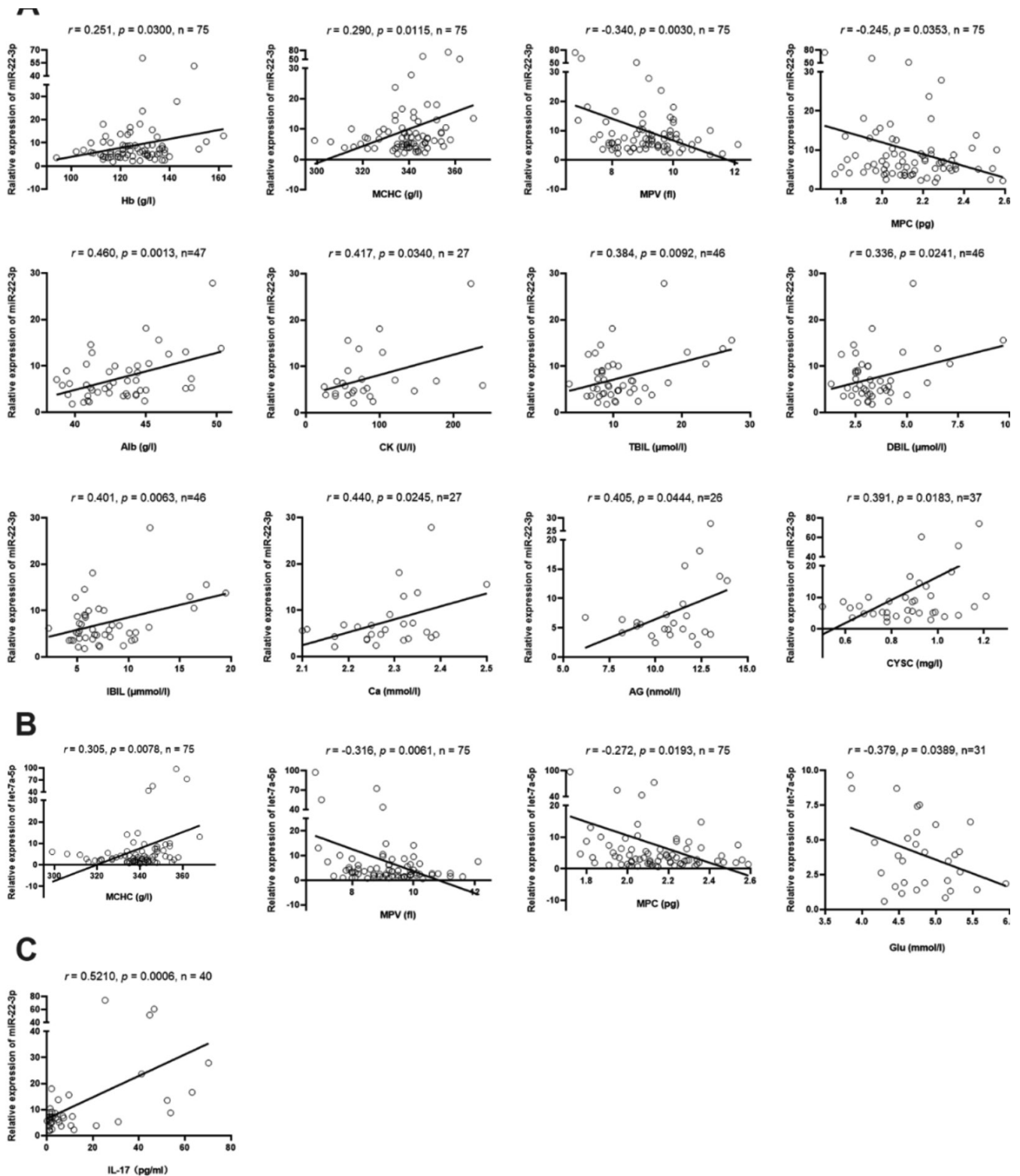


Supplementary Fig. S1. Circulating miR-22-3p and let-7a-5p in RA patients with normal/increased RF, anti-CCP, ESR and CRP. RA patients were divided into the four groups as follows: RA patients with increased RF and increased anti-CCP (RF+anti-CCP+), RA patients with increased RF and normal anti-CCP (RF+anti-CCP-), RA patients with normal RF and increased anti-CCP (RF-anti-CCP+), RA patients with normal RF and normal anti-CCP (RF-anti-CCP-). The levels of circulating miR-22-3p (A) and let-7a-5p (B) were further analysed in comparison among four groups and correlation analysis. For ESR and CRP, RA patients were also categorised into different groups based on different ESR and CRP levels: RA patients with increased ESR and increased CRP (ESR+CRP+), RA patients with increased ESR and normal CRP (ESR+CRP-), RA patients with normal ESR and increased CRP (ESR-CRP+), RA patients with normal ESR and normal CRP (ESR-CRP-). The levels of circulating miR-22-3p (C) and let-7a-5p (D) were analysed according to the method mentioned above. (E) Correlations of circulating miR-22-3p and let-7a-5p level with DAS28. Comparison among multiple groups was applied using Mann-Whitney U-test followed by Dunn's posttest and correlation coefficients (r) obtained from Pearson correlation test are shown. RF, rheumatoid factors; anti-CCP, anti-cyclic citrullinated peptide; ESR, erythrocyte sedimentation rate; CRP, C-reactive protein, DAS28, the disease activity score assessing 28 joints.



Supplementary Fig. S2. Correlation of circulating miR-22-3p and let-7a-5p with the clinical parameters of RA.

A: Correlations of circulating miR-22-3p levels with Hb, MCHC, MPV, MPC, Alb, CK, TBIL, DBIL, IBIL, Ca, AG and CYSC.

B: Correlations of circulating let-7a-5p levels with MCHC, MPV, MPC and Glu.

C: Correlations of circulating miR-22-3p level with serum cytokine IL-17. Correlation coefficients (r) obtained from Pearson correlation test are shown.

Hb: haemoglobin; MCHC: mean cell haemoglobin concentration; MPV: mean platelet volume; MPC: mean platelet contents; Alb: albumin; CK: creatine kinase; TBIL: total bilirubin; DBIL: direct bilirubin; IBIL: indirect bilirubin; Ca: Calcium; AG: anion gap; CYSC: cystatin C. Glu: Glucose; IL-17: interleukin-17.

Supplementary Table S1. Primers used for qRT-PCR.

Species	miRNA Name	Sequence (5'-3')
Homo sapiens	miR-22-3p	Fw: GCGAAGCTGCCAGTTGAAG Rv: AGTGCAGGGTCCGAGGTATT Rt: GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACACAGTT
	let-7a-5p	Fw: GCGCGTGAGGTAGTAGGTTGT Rv: AGTGCAGGGTCCGAGGTATT Rt: GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACAACTAT

Fw: Forward primer; Rv: Reverse primer; Rt: Reverse transcription primer.

Supplementary Table S2. Diagnostic performance of various indicators in differentiating healthy donors or patients with systemic lupus erythematosus (SLE) and Sjögren's syndrome (SS) from patients with rheumatoid arthritis (RA) by receiver operating characteristic (ROC) curve analysis.

Indicators	Subgroups	Youden Index	Cut-off value	Sensitivity	Specificity	ROC AUC	95% Confidence Interval	p-value
miR-22-3p	RA vs. HD	0.5175	0.000494754037 [#]	68.42	83.33	0.812	0.727 - 0.880	<0.0001
	RA vs. SLE	0.5088	0.0003749537921 [#]	84.21	66.67	0.803	0.715 - 0.874	<0.0001
	RA vs. SS	0.5214	0.0003906053951 [#]	80.26	71.87	0.785	0.695 - 0.858	<0.0001
let-7a-5p	RA vs. HD	0.5365	0.0001399307356 [#]	84.21	69.44	0.832	0.750 - 0.896	<0.0001
	RA vs. SLE	0.3211	0.0003838953835 [#]	42.11	90.00	0.684	0.586 - 0.770	0.0009
	RA vs. SS	0.4868	0.0001785977708	73.68	75.00	0.775	0.684 - 0.850	<0.0001
combined miR-22-3p/let-7a-5p	RA vs. HD	0.5731	0.69327213 [†]	68.42	88.89	0.848	0.768 - 0.909	<0.0001
	RA vs. SLE	0.4632	0.623727728 [†]	76.32	70.00	0.794	0.704 - 0.866	<0.0001
	RA vs. SS	0.4967	0.66927115 [†]	68.42	81.25	0.814	0.728 - 0.883	<0.0001

[#] number represents $2^{-\Delta Ct}$; $\Delta Ct = (Ct_{miR-22-3p/let-7a-5p} - Ct_{cel-miR-39-3p})$; [†] number represents predicted probability.

The optimum cut-off values of miR-22-3p and let-7a-5p in RA vs. HD subgroup were used to dichotomised RA patients as miRNA-positive or miRNA-negative. AUC, area under curve.

Supplementary Table S3. Univariate and multivariate logistic regression analysis.

Variables	Univariate logistic regression analysis			Multivariate logistic regression analysis		
	OR	95% Confidence Interval	p-value	OR	95% Confidence Interval	p-value
miR-22-3p	8.9762	3.4484 - 23.3651	<0.0001****	3.7766	0.7395 - 19.2871	0.1102
let-7a-5p	12.1212	4.7362 - 31.0211	<0.0001****	6.0011	1.0807 - 33.3257	0.0405*
RF	44.0000	11.8676 - 163.1326	<0.0001****	10.9368	1.8381 - 65.0754	0.0086**
anti-CCP	117.3529	14.9530 - 921.0008	<0.0001****	35.1984	3.3841 - 366.1020	0.0029**

The asterisk indicates statistically significant results. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

Supplementary Table S4. Comparison of the diagnostic ability among various combination indicators in RA vs. HD subgroup by evaluating the area under curve (AUC) using z tests (numbers represent p-values).

Indicators	AUC	miR-22-3p/anti-CCP/RF	let-7a-5p/anti-CCP/RF	miR-22-3p/let-7a-5p/anti-CCP/RF
miR-22-3p/anti-CCP/RF	0.951	-	-	-
let-7a-5p/anti-CCP/RF	0.954	0.8058	-	-
miR-22-3p/let-7a-5p/anti-CCP/RF	0.961	0.1961	0.2070	-

Supplementary Table S5. Diagnostic performance of various combination indicators in differentiating healthy donors from patients with rheumatoid arthritis (RA) by receiver operating characteristic (ROC) curve analysis.

Subgroups	Combination Indicators	Youden Index	Cut-off value	Sensitivity	Specificity	ROC AUC	95% Confidence interval	p-value
RA vs. HD	anti-CCP	0.7425	0.7425 [#]	77.03	97.22	0.871	0.794 - 0.928	<0.0001
	RF	0.7167	0.7167 ^{##}	80.00	91.67	0.858	0.779 - 0.917	<0.0001
	anti-CCP/RF	0.7538	0.226638804 [‡]	86.49	88.89	0.917	0.849 - 0.961	<0.0001
	let-7a-5p/RF	0.7167	0.55272609 [‡]	80.00	91.67	0.922	0.855 - 0.964	<0.0001
	let-7a-5p/anti-CCP	0.7425	0.572963135 [‡]	77.03	97.22	0.937	0.874 - 0.975	<0.0001
	miR-22-3p/anti-CCP/RF	0.7830	0.78648804 [‡]	81.08	97.22	0.951	0.892 - 0.983	<0.0001
	let-7a-5p/anti-CCP/RF	0.8101	0.746799948 [‡]	83.78	97.22	0.954	0.896 - 0.985	<0.0001
	miR-22-3p/let-7a-5p/anti-CCP/RF	0.8236	0.669814101 [‡]	85.14	97.22	0.961	0.906 - 0.989	<0.0001

[#]AUC, area under curve.

The unit of number is IU/ml; ^{##}The unit of number is RU/ml; [‡]numbers represent predicted probability.