Impact of human leukocyte antigen-DPB1 rs9277535 and vascular endothelial growth factor receptor 1 rs9943922 on Rheumatoid arthritis susceptibility and activity in an Egyptian case-control study

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Background/aim
Rheumatoid arthritis (RA) is one of the most common autoimmune systemic inflammatory diseases presenting with chronic articular and extra articular manifestations resulting in high incidences of deformities, disabilities, morbidities, and mortalities. Two polymorphisms, Rs9277535 of human leukocyte antigen (HLA)-DPB1 gene and rs9943922 of vascular endothelial growth factor receptor 1 (VEGFR1) gene, are suspected to have impact on the pathogenesis of RA. The current work aims to investigate the association of HLA-DPB1 gene rs9277535 and VEGFR1 gene rs9943922 with RA susceptibility in a sample of Egyptian patients.
Patients and methods
This case–control study involved 200 unrelated RA patients and 200 sex and age matched healthy volunteers acting as a control group. The patients were recruited from the Outpatient Clinics of the Rheumatology Department, National Research Centre, Centre of Medical Excellence, Cairo, Egypt. Patients were assessed clinically and laboratory for determination of disease activity. Genotyping of HLA-DPB1 (rs9277535) and VEGFR1 (rs9943922) was done using real-time PCR technique.
Results
The frequency of the G allele of HLA-DPB1 rs9277535 was found to be higher in RA group in comparison with the control group. Disease activity score28 and clinical disease activity index were significantly higher with GG genotype compared with AA after post hoc analysis (P=0.010, P=0.020, respectively). Moreover, this study found no significant differences regarding the distribution of VEGFR1 rs9943922 genotypes (P=0.120) or alleles frequencies (P=0.502) between healthy controls and studied patients.
Conclusion
Our study revealed an association between the G allele of HLA-DPB1 rs9277535 and RA susceptibility and disease activity while VEGFR1 rs9943922 single nucleotide polymorphism plays no role in predisposition to RA in a sample of the Egyptian population.

Keywords:
human leukocyte antigen-DPB1 gene, rheumatoid arthritis, rs9277535, rs9943922, vascular endothelial growth factor receptor 1 gene

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Introduction

Rheumatoid arthritis (RA) is one of the most common autoimmune systemic inflammatory diseases with up to 1% prevalence in some populations [1]. The hallmark of RA is chronic synovial joint tissue inflammation leading to joint erosion, destruction, deformity and disability [2]. Extra-articular manifestations of RA affect all body systems including cardiovascular, neurological, renal, and other vital systems with high rates of morbidity and mortality [3]. The etiology of the disease is multifactorial with interaction between genetic, epigenetic, environmental, and hormonal factors resulting in aberrant activation of the immune system and subsequently generalized and localized inflammatory responses [4]. Management of RA from conventional immunomodulation to targeted therapy towards interleukin-6, tumor necrosis factor, B cells, T cells or Janus kinase-signal transducers and activators of transcription (JAK–STAT) pathway did not establish complete remission in many patients [5]. The diversity of

clinical presentation, unclear pathogenesis, and treatment refractoriness calls for urgent and continuous research to unveil novel etiologic factors and future therapeutic targets for RA [6].

Genetic variations on the sixth chromosome in the region of human leukocyte antigen (HLA), known also as the major histocompatibility complex, including its three loci are highly associated with susceptibility to RA [7]. HLA-II antigen presenting molecules activate CD4+ T cells and trigger immune reactions. Belonging to HLA-II molecules, HLA-DP variants, are attracting more attention due to its critical role in autoantigens and viral antigens presentation to T cells [8]. Polymorphisms affecting HLA-DP were correlated to systemic lupus erythematosus (SLE) and multiple sclerosis in previous studies [9,10]. The HLA-DP molecule is a heterodimer that is formed of an alpha (DPA) and a beta (DPB) chain. DPA1 and DPB1 alleles encode the A- and B-chains separately with DPB1 exon's nucleotide sequence showing high significant variations and hence allele polymorphisms [11]. Rs9277535 is a single nucleotide polymorphism (SNPs) located in the 3′-UTR regions of the HLA-DPB1 which is suspected to impact the function and expression of HLA-DPB1 with potential contribution to the susceptibility of RA [12].

Vascular endothelial growth factor receptor 1 (VEGFR1) gene also known as fms-related tyrosine kinase 1 (Flt1) gene, is located in chromosome 13q12. VEGFR1 gene encodes VEGFR1 a member of the VEGFR family with high binding affinity to VEGF proteins [13], this gene uniquely expresses two mRNA types. The first mRNA encodes a full-length VEGFR1 which is expressed on various cells including synovial endothelial cells (ECs) and macrophages [14]. VEGFR1 might contribute to RA through either amplified angiogenesis that in turn enhances leukocyte infiltration into synovial tissue leading to disease progression [15] or through direct promotion of the inflammatory function of macrophages. The other RNA encodes a short soluble VEGFR1 protein (sVEGFR1) which acts as a natural competitor to VEGFR1 through capturing their ligands leading to an antiangiogenic effect. On the other hand, sVEGFR1 shows an angiogenic effect upon interacting with components of ECs [14]. Function discrepancy between VEGFR1 and sVEGFR1 in addition to fluctuation of sVEGFR serum levels in various autoimmune and inflammatory diseases highlight the role of VEGFR1 gene polymorphisms in the pathogenesis of those diseases. Rs9943922 polymorphism of VEGFR1 gene has been correlated to different diseases and to serum levels of sVEGFR1 which nominates it as a role player in RA pathogenesis [16].

The current work aims to investigate the association of HLA-DPB1 gene rs9277535 and VEGFR1 gene rs9943922 with RA susceptibility in a sample of Egyptian patients.

Patients and methods

Patients and study design

Our case–control study involved 200 unrelated RA patients and 200 sex and age matched healthy volunteers acting as a control group. The patients were recruited from the Outpatient Clinics of Rheumatology Department, Centre of Medical Excellence, National Research Centre, Cairo, Egypt Patients were diagnosed according to the classification criteria of the American College of Rheumatology/ European League Against Rheumatism (ACR/ EULAR) 2010 [17]. Exclusion criteria for the patients was the presence of any associated autoimmune disease and for healthy control was the presence of a history of autoimmune diseases in the family.

All patients underwent detailed medical history and full clinical examination focusing on assessment of the musculoskeletal system and swollen or tender joints in addition to laboratory checkups. The disease activity including clinical disease activity index (CDAI), disease activity score 28 (DAS28), and patient global assessment (PGA) were determined.

Ethical approval

The current work was approved by the local Ethical Committee of the National Research Centre (NRC), Cairo, Egypt with approval number 19047 following the Code of Ethics of the World Medical Association, in agreement with the principles expressed in the Declaration of Helsinki in 2015. All participants signed informed written consent before participation in the study.

Methods

RA disease activity

The Disease activity of RA was determined according to the CDAI by Aletaha *et al.* [18], DAS28 by Prevoo *et al.* [19], and PGA, by Nikiphorou *et al.* [20].

Sample collection

Six ml of venous blood were withdrawn from each patient, 2 ml of them were collected in serum separator

vacutainers, centrifuged and the serum was collected and stored at −20° C till analysis. The other 2 ml were withdrawn into Na-citrate tubes for erythrocyte sedimentation rate (ESR) measurement, while the final 2 ml were added to EDTA tubes for complete blood picture (CBC) and genotyping analysis.

Laboratory tests

ESR was measured with the Westergren method, CBC was done using Sysmex xs-500i Hematology Analyzer (Sysmex, Kobe, Japan), Mispa i2 semi-automated nephelometer and Aggape kits (AGAPPE Diagnostic, Switzerland GmbH) were used to assess serum concentrations of C-reactive protein (CRP) and rheumatoid factor (RF). Cobas 6000 analyzer series (Roche Diagnostics, Mannheim, Germany) was utilized to measure anticyclic citrullinated peptides (ACCP).

Genotyping analysis for HLA-DPB1 (rs9277535) and VEGFR1 (rs9943922) gene polymorphisms by PCR

Genomic DNA was extracted from whole-blood samples by QIAamp DNA Blood Kit (Qiagen, Hilden, Germany). Genotyping of HLA-DPB1 (rs9277535) and VEGFR1(rs9943922) polymorphisms on LightCycler 480 real-time PCR System (Roche Diagnostics, Basel, Switzerland) was done by TaqMan allelic discrimination method using probes and primers made by Applied Biosystems (TaqMan SNP Genotyping Assay, Cat no: 4351379, Foster City, California, USA). Samples were prepared by adding10 µl Master Mix, 0.5 µl of the assay primer, 20 ng of tested DNA, and nuclease free water to complete 20 µl volume. PCR thermal profile was 95°C initial activation step for 10 min then 40 repeated cycles of denaturation at 95°C for 15 s and annealing/extension at 60°C for 1 min. Collection of fluorescence data was done at the extension step and analysis of the final product was performed by LightCycler 480 real-time PCR System software program. All steps were done following manufacturers manual. Quality control was ensured by running positive and negative controls with each run in addition to assaying 10% of samples in duplicates with 100% concordance rate [8].

Statistical analysis

SPSS (IBM Corp, Armonk, New York, USA) version 22 software was used for Data analysis. Frequencies were used to describe qualitative data while mean±SD were used to describe quantitative data. Qualitative parameters were compared by the χ^2 test and quantitative data comparisons were done using Student's t test. Analysis of variance (ANOVA) test was used in analyzing the difference between the means of more than two groups. The Hardy–Weinberg equilibrium of the studied SNPs was detected among healthy controls using the χ^2 test. The logistic regression analysis was used to evaluate associations between the studied SNPs and RA with the calculation of the odds ratios and 95% confidence intervals and after adjustment of traditional risk factors.

Results

Our results showed no differences between the studied patients and the controls regarding age (45.3±12.4, 44.5±13, P=0.432) and sex (182 females/18 males, 188 females/12 males, respectively, P=0.302). Table 1 shows other characteristics of RA patients' group including presence of family history, disease duration, disease activity scores including CDAI, DAS28 and PGA, in addition to the laboratory results that includes, ESR, hemoglobin, total leukocytic count (TLC), platelets, CRP, RF and ACCP.

Deviation of the control group from Hardy–Weinberg equilibrium revealed no significant differences regarding rs9277535 and rs9943922 (P= 0.812 and 0.631, respectively), (In-tabulated data).

On studying genotypes of HLA-DPB1 gene (rs9277535), the current study revealed no differences in genotypes distribution between healthy controls and RA patients (P=0.200) (Table 2). However, the frequency of G allele was found to be higher in patients' group in comparison with the control group (Table 2). Further analysis using

Table 1 Characteristics of the studied rheumatoid arthritis (RA) patients

Variable	RA patient (n=200)
Family history of RA [+]	21 (10.5)
Disease duration (years) [++]	7.2±3.5
ESR (mm/1st h) [++]	31±12
CRP positivity [+]	109 (54.5)
RF positivity [+]	126 (63)
ACCP positivity [+]	83 (41.5)
Hemoglobin, g/dL [++]	12.2±1.1
TLC, $\times10^3$/cmm [++]	10.5±5.2
Platelets, $\times10^3$/cmm [++]	285±64
CDAI [++]	22.1±10.5
DAS28 [++]	4.5±1.7
PGA [++]	5.9±1.9

[+]: Data are represented as number (%). [++]: data are represented as mean±SD, ESR=erythrocyte sedimentation rate. ACCP, anticyclic citrullinated peptide; CDAI, clinical disease activity index; CRP, C-reactive protein; DAS, disease activity score, PGA=patient global assessment; RF, rheumatoid factor; TLC, total leucocytic count.

Table 2 Distribution of genotypes and alleles frequencies and different models in the two studied groups

SNP	Variable	Control group (n=200) N (%)	RA group (n=200) N (%)	*p-value*	OR (CI)
HLA-DPB1 (rs9277535)	**Genotypes**				
	AA	171 (85.5%)	158 (79%)	0.200	
	AG	8 (4%)	12 (6%)		
	GG	21(10.5%)	30(15%)		
	Alleles (2n)				
	A allele	350 (87.5%)	328 (82%)	0.030*	1.5 (1-2.2)
	G allele	50 (12.5%)	72 (18%)		
	Additive model: AA	171 (85.5%)	158 (79%)	reference	
	AG	8 (4%)	12 (6%)	0.815	1.1 (1.1-3.3)
	GG	21 (10.5%)	30(15%)	0.131	0.6 (0.3-1.2)
	Dominant model: (GG+AG / AA)	(29/171)	(42/158)	0.072	1.8 (0.9-2.7)
	Recessive model: (GG / AG+GG)	(21/179)	(30/170)	0.536	1.2 (0.9-2.3)
VEGFR1 (rs9943922)	**Genotypes**				
	CC	112 (56%)	94 (47%)	0.120	
	CT	41 (20.5%)	52 (26%)		
	TT	47 (23.5%)	54 (27%)		
	Alleles (2n)				
	C	271 (67.75)	264 (66%)	0.502	0.4 (0.3-0.6)
	T	129 (32.25%)	136 (34%)		
	Additive model: CC	112 (56%)	94 (47%)	reference	
	CT	41 (20.5%)	52(26%)	0.143	1.5 (0.9-2.4)
	TT	47 (23.5%)	54 (27%)	0.210	1.3 (0.8-2.2)
	Dominant model: (TT+CT/CC)	(88/112)	(106/94)	0.081	1.4 (0.9-2.1)
	Recessive model: (TT/CT+CC)	(47/153)	(54/146)	0.432	0.8(0.5-1.3)

CI, confidence interval; OR, odds ratio. Data are presented as number (%). *Significant difference at *P* value less than 0.05, using Chi-square test.

logistic regression analysis after adjustment of other cofounders including age, sex, family history and disease duration, rs9277535 was not associated with susceptibility of RA either under recessive or dominant models (Table 2).

Regarding HLA-DPB1 gene (rs9277535), ESR, CRP, RF, ACCP, hemoglobin, TLC, platelet count and PGA score, showed no statistical differences between different genotypes within patient group (Table 3), while DAS28 and CDAI scores were significantly higher with GG genotype compared with AA genotype after post hoc analysis (*P*=0.010, *P*=0.020, respectively)

Moreover, this study did not find any significant differences regarding the distribution of VEGFR1 rs9943922 genotypes (*P*=0.120) or alleles frequencies

Table 3 Different patients' parameters regarding HLA-DPB1 (rs9277535) genotypes

Parameter	AA (*n*=157)	AG (*n*=13)	GG (*n*=30)	*P* value
ESR (mm/1st h) $^{++}$	32.9±13	30.7±12	34±12	0.502
CRP Positivity $^{+}$	91 (58.0)	4 (30.7)	14 (46.7)	0.107
RF positivity $^{+}$	101 (64.3)	7 (53.8)	18 (60)	0.757
ACCP Positivity +	64 (40.8)	5 (38.5)	14 (46.7)	0.820
Hemoglobin, g/dl^{++}	12±1.2	12.5±1.3	11.5±1.2	0.083
TLC, $\times10^3$/cmm $^{++}$	9.2±3.9	10.8±2.7	10.5±2.6	0.804
Platelets, $\times10^3$/cmm $^{++}$	294±81	279±55.5	282±78	0.605
CDAI $^{++}$	21.3±9.8^{a}	20.8±9.7^{a}	22.1±10.2^{b}	0.042*
DAS28 $^{++}$	4.3±1.7^{a}	4.1±1.8^{a}	5.2±1.8^{b}	0.031*
PGA $^{++}$	4.7±2.1	4.8±2.3	5.8±2.4	0.206

$^{+}$: Data are represented as number (%) and insignificantly changed using Chi-Square at *P* value less than 0.05 within the same raw. $^{++}$: Data are represented as mean±SD. Data with different superscript number (a, b) within the same raw are significantly changed at **P* value less than 0.05, using analysis of variance test. ACCP, anti-cyclic citrullinated peptide; CDAI, clinical disease activity index; CRP, C-reactive protein; DAS, disease activity score; ESR, erythrocyte sedimentation rate; PGA, patient global assessment; RF, rheumatoid factor; TLC, total leucocytic count.

Table 4 Different patients' parameters regarding vascular endothelial growth factor receptor 1 rs9943922 genotypes

Parameter	CC (*n*=94)	CT (*n*=52)	TT (*n*=54)	*P* value
ESR (mm/1st h)$^{++}$	32±11	34±14	31±12	0.090
CRP positivity $^{+}$	50 (53)	30 (57.7)	29 (53.7)	0.863
RF positivity $^{+}$	61 (64.9)	35 (67.3)	30 (55.6)	0.398
ACCP positivity $^{+}$	40 (42.5)	22 (42.3)	21 (38.9)	0.569
Hemoglobin, g/dl^{++}	12±1.6	12.2±1.4	11.8±1.1	0.105
TLC, $\times10^3$/cmm $^{++}$	9±3.2	10.4±2.3	9±2.5	0.407
Platelets, $\times10^3$/cmm $^{++}$	275±66	296±91	286±79	0.082
CDAI $^{++}$	22±10.5	22.5±9.2	21±9.1	0.066
DAS28 $^{++}$	3.4±1.8	4.1±1.7	3.8±1.3	0.203
PGA $^{++}$	4.5±2.1	4.6± 2.2	3.9±1.3	0.404

$^{+}$: Data are represented as number (%) and insignificantly changed using Chi-Square at *P* value less than 0.05 within the same raw. $^{++}$: Data are represented as mean±SD and are insignificantly changed using analysis of variance tests at *P*value less than 0.05 within the same raw. ACCP, anti-cyclic citrullinated peptide; CDAI, composite index for quantifying disease activity; CRP, C-reactive protein; DAS, disease activity score; ESR, erythrocyte sedimentation rate; PGA, patient global assessment; RF, rheumatoid factor; TLC, total leucocytic count.

(*P*=0.502) between healthy controls and studied patients (Table 2). Regarding VEGFR1 rs9943922 polymorphism, ESR, CRP, RF, ACCP, hemoglobin TLC, platelet count DAS28, CDAI and PGA scores showed no statistical differences between different genotypes within patients' group (Table 4).

Discussion

RA is a widespread chronic autoimmune inflammatory disorder. Despite of the multifactorial nature of the disease, genetic elements are responsible for about 60% of its developing risk. Therefore, studying and pinpointing the exact genetic pathogenesis of RA has always been one of the most intriguing and challenging issues [21].

HLA-DPB1 is a highly polymorphic gene. It has an important immune regulatory role that deserves to be under the spot. Although HLA-DPB1 variants were reported to be associated with susceptibility to RA [22,23], the role of HLA-DPB1 rs9277535 in RA patients is not well clarified yet. This is the first study to investigate rs9277535 in RA patients in a sample of the Egyptian population.

In the current study, HLA-DPB1 rs9277535 genotypes showed no significant difference between RA cases and healthy controls, however, the frequency of G allele was found to be higher in patients' group compared with healthy control. Huang and colleagues described an association between HLA-DPB1 rs9277535 and increased risk of RA in the West China population. However, in contrast to our findings, they nominated the A allele of rs9277535 as the highest risk allele in RA [8]. Similarly, Yang et al. observed a lower susceptibility to RA with the G allele of HLA-DPB1 rs9277535 among Chinese individuals [12]. On the other hand, in agreement to our results, another study conducted by Zhang *el al.*, on a similar autoimmune disease found that allele G of rs9277535 predisposed to SLE in the Chinese Han population [10]. Previous gene expression studies found that allele G of rs9277535 was correlated with the down-regulated level of HLA-DPB1 mRNA [12,24]. Given into consideration that antigen presentation is crucial for establishing immune tolerance as well as normal immune response [25], down-regulation of HLA-DPB1 might lead to disrupted immune tolerance with subsequent autoimmunity.

Moreover, previous studies have reported an association between rs9277535 and other immune mediated diseases. Yang et al. demonstrated that rs9277535 is an associated genetic risk of IgA nephropathy in a Southwest Chinese population [26]. In addition, Genome-Wide Association studies have previously validated HLA-DPB1 rs9277535 variants as a genetic susceptibility factor for HBV chronicity in Asian community [27].

In the current study, we found that DAS28 and CDAI scores showed a statistically significant increase in patients with GG genotype compared with AA group signifying that GG genotype is related to high disease activity. Consistent with our findings, Zhang *et al.*, also revealed an association between GG genotype of the rs9277535 and markers of SLE disease activity indicated by consumed C3 and C4 [10].

It is well known that RA is characterized by inflammation and angiogenesis in the synovial tissues, which ultimately ends by hyperplastic growth of the synovial membrane and joint damage [28]. Angiogenesis essentially depends on the interaction

between fibroblasts, macrophages, vascular ECs, and the extracellular matrix. Activated immune cells generate numerous inflammatory cytokines that are pivotal in driving the onset and progression of RA. The angiogenic effects of these cytokines are exerted either through a direct effect on ECs or by indirect effect through the production of the pro-angiogenic VEGF [29]. High plasma and synovial VEGF levels have been previously reported in RA [30]. Moreover, a strong association between VEGF serum level and inflammatory markers of RA including ESR, CRP, and DAS28 have been described [31]. Accordingly, VEGF and its receptors are important subjects of research in RA, however, they remain insufficiently explored in this domain.

sVEGFR1 is a splice variant of the VEGFR that lacks both the transmembrane and cytoplasmic domains. As a decoy receptor, sVEGFR1 binds to VEGF and counteracts its pro-angiogenic actions. By inhibiting VEGF, sVEGFR1 consequently disrupts angiogenesis and impairs endothelial repair [32,33]. SNPs at the VEGFR1 locus may affect VEGF signaling pathway and increase predisposition to more angiogenic conditions.

This is the first study that investigated the VEGFR1 rs9943922 SNP in a sample of Egyptian patients with RA. Our findings did not identify any significant differences of sVEGFR1 rs9943922 genotypes or alleles frequencies between studied patients and controls. Our results were in agreement with Paradowska-Gorycka *et al.* [34], who also found no significant difference in genotypic distribution or allelic frequencies between RA and healthy groups for the VEGFR1 rs9943922 in 471 Polish patients. Conversely, Yuan *et al.* [35] described genotype TT of rs9943922 as high-risk factor for SLE susceptibility in the Chinese Han population. They explained their contradictory findings by noting that, although VEGFR1 rs9943922 SNP is situated in noncoding region of the gene (introns), it might influence the normal splicing and enhance the gene's transcriptional activity. They supported their perspective by citing previous studies that elucidated the effect of polymorphisms in noncoding region of VEGFR1 in silencing or enhancing sVEGFR1 transcriptional activity [36,37].

Discrepancies between results across different studies either in HLA-DPB1 rs9277535 or VEGFR1 rs9943922 SNP could be attributed to various factors including small sample size, heterogenic nature of the disease, diverse ethnic backgrounds of different population in addition to dietary and other regional and environmental factors. It is worthwhile to mention that limitations of the current study included a relatively small sample size and only Egyptian individuals in Cairo, Egypt were included. Further studies on the structure-function of those SNPs using large sample sizes across various ethnic groups are needed to confirm the results observed in this study.

Conclusion

In conclusion, our study revealed an association of the G allele of HLA-DPB1 rs9277535 with RA susceptibility and disease activity while VEGFR1 rs9943922 SNP plays no role in predisposition to RA in a sample of Egyptian population.

Acknowledgments

Authors' contributions: I.A. was responsible for conceptualization, study design, work administration, supervision, laboratory investigations, interpretation of the results, data validation, participated in writing the first draft of the manuscript in addition to the submission of the manuscript to the journal. M.H. was responsible for the laboratory investigations, and interpretation of the results, participated in writing the first draft and critical revision of the final version of the manuscript. A.R. was responsible for the laboratory investigations and interpretation of the results. M.A. was in charge of patients' selection, clinical history taking, medical data collection, and recording.

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Conflicts of interest

There are no conflicts of interest.

References

1 Alamanos Y, Voulgari PV, Drosos AA. Incidence and prevalence of rheumatoid arthritis, based on the 1987 American College of Rheumatology criteria: a systematic review. Semin Arthritis Rheum 2006; 36:182–188.

2 Gravallese EM, Firestein GS. Rheumatoid arthritis-common origins, divergent mechanisms. N Engl J Med 2023; 388:529–542.

3 Figus FA, Piga M, Azzolin I, McConnell R, Iagnocco A. Rheumatoid arthritis: Extra-articular manifestations and comorbidities. Autoimmun Rev 2021; 20:102776.

4 Bemis EA, Demoruelle MK, Seifert JA, Polinski KJ, Weisman MH, Buckner JH, *et al.* Factors associated with progression to inflammatory arthritis in first-degree relatives of individuals with RA following autoantibody positive screening in a non-clinical setting. Ann Rheum Dis 2021; 80:154–161.

5 Alivernini S, Laria A, Gremese E, Zoli A, Ferraccioli G. ACR70-disease activity score remission achievement from switches between all the available biological agents in rheumatoid arthritis: a systematic review of the literature. Arthritis Res Ther 2009; 11:R163.

6 Oliver J, Plant D, Webster AP, Barton A. Genetic and genomic markers of anti-TNF treatment response in rheumatoid arthritis. Biomark. Med 2015; 9:499–512.

7 Eyre S, Bowes J, Diogo D, Lee A, Barton A, Martin P, *et al.* High-density genetic mapping identifies new susceptibility loci for rheumatoid arthritis. Nat Genet 2012; 44:1336–1340.

8 Huang Z, Niu Q, Yang B, Zhang J, Yang M, Xu H, *et al.* Genetic polymorphism of rs9277535 in HLA-DP associated with rheumatoid arthritis and anti-CCP production in a Chinese population. Clin Rheumatol 2018; 37:1799–1805.

9 Field J, Browning SR, Johnson LJ, Danoy P, Varney MD, Tait BD, *et al.* A polymorphism in the HLA-DPB1 gene is associated with susceptibility to multiple sclerosis. PLoS 2010; 5:e13454.

10 Zhang J, Zhan W, Yang B, Tian A, Chen L, Liao Y, *et al.* Genetic polymorphisms of rs3077 and rs9277535 in HLA-DP associated with systemic lupus erythematosus in a Chinese population. Sci Rep 2017; 7:39757.

11 Watts RA, MacGregor AJ, Mackie SL. HLA allele variation as a potential explanation for the geographical distribution of granulomatosis with polyangiitis. Rheumatol 2015; 54:359–362.

12 Yang Z, Liu W, Yan T, Liu R. HLA-DPB1 rs9277535 polymorphism is associated with rheumatoid arthritis risk in a Chinese Han population. Aging (Albany NY) 2021; 13:11696–11704.

13 Wang H, Yang L, Deng J, Wang B, Yang X, Yang R, *et al.* Genetic variant in the 3'-untranslated region of VEGFR1 gene influences chronic obstructive pulmonary disease and lung cancer development in Chinese population. Mutagenesis 2014; 29:311–317.

14 Shibuya M. Vascular endothelial growth factor receptor-1 (VEGFR-1/Flt-1): a dual regulator for angiogenesis. Angiogenesis 2006; 9:225–230.

15 Maroutti N, Cantatore FP, Crivellto E, Vacca A, Ribatti D. Angiogenesis in rheumatoid arthritis. Histol Histopathol 2006; 21:557–566.

16 Owen LA, Morrison MA, Ahn J, Woo SJ, Sato H, Robinson R, *et al.* FLT1 genetic variation predisposes to neovascular AMD in ethnically diverse populations and alters systemic FLT1 expression. Invest Ophthalmol Vis Sci 2014; 55:3543–54.

17 Kay J, Upchurch K. ACR/EULAR 2010 rheumatoid arthritis classification criteria. Rheumatology 2012; 51(Suppl 6):vi5–vi9.

18 Aletaha D, Nell VPK, Stamm T, Uffmann M, Pflugbeil S, Machold K, Smolen JS. Acute phase reactants add little to composite disease activity indices for rheumatoid arthritis: validation of a clinical activity score. Arthr Res Therapy 2005; 7:R796–R806.

19 Prevoo ML, Van'T Hof MA, Kuper HH, Van Leeuwen MA, Van de Putte LB, Van Riel PL. Modified disease activity scores that include twenty-eight-joint counts development and validation in a prospective longitudinal study of patients with rheumatoid arthritis. Arthr Rheum 1995; 38:44–48.

20 Nikiphorou E, Radner H, Chatzidionysiou K, Desthieux C, Zabalan C, Van Eijk-Hustings Y, *et al.* Patient global assessment in measuring disease activity in rheumatoid arthritis: a review of the literature. Arthritis Res Ther 2016; 18:251.

21 Ibrahim A, Abdelhadi M, Ashmawy I, Ramadan A, Wahby A, Hassan M, *et al.* The association of TRAP1 gene and TNFSF4 gene polymorphisms with susceptibility of rheumatoid arthritis in a sample of Egyptian patients. J Arab Soc Med Res 2023; 18:76–81.

22 Yarwood A, Han B, Raychaudhuri S, Bowes J, Lunt M, Pappas DA, *et al.* A weighted genetic risk score using all known susceptibility variants to estimate rheumatoid arthritis risk. Ann Rheum Dis 2015; 74:170–176.

23 Furukawa H, Oka S, Shimada K, Hashimoto A, Komiya A, Tsunoda S, *et al.* Independent association of HLA-DPB1*02:01 with rheumatoid arthritis in Japanese populations. PLoS One 2018; 13:e0204459.

24 O'Brien TR, Kohaar I, Pfeiffer RM, Maeder D, Yeager M, Schadt EE, Prokunina-Olsson L. Risk alleles for chronic hepatitis B are associated with decreased mRNA expression of HLA-DPA1 and HLA-DPB1 in normal human liver. Genes Immun 2011; 12:428–433.

25 Matheoud D, Sugiura A, Bellemare-Pelletier A, Laplante A, Rondeau C, Chemali M, *et al.* Parkinson's Disease-Related Proteins PINK1 and Parkin Repress Mitochondrial Antigen Presentation. Cell 2016; 166:314–327.

26 Yang B, Zhang J, Liu X, Huang Z, Su Z, Liao Y, Wang L. Genetic polymorphisms in HLA-DP and STAT4 are associated with IgA nephropathy in a Southwest Chinese population. Oncotarget 2018; 9:7066–7074.

27 Kamatani Y, Wattanapokayakit S, Ochi H, Kawaguchi T, Takahashi A, Hosono N, *et al.* A genome-wide association study identifies variants in the HLA-DP locus associated with chronic hepatitis B in Asians. Nat Genet 2009; 41:591–595.

28 Leblond A, Allanore Y, Avouac J. Targeting synovial neoangiogenesis in rheumatoid arthritis. Autoimmun Rev 2017; 16:594–601.

29 Elshabrawy HA, Chen Z, Volin MV, Ravella S, Virupannavar S, Shahrara S. The pathogenic role of angiogenesis in rheumatoid arthritis. Angiogenesis 2015; 18:433–448.

30 Clavel G, Bessis N, Lemeiter D, Fardellone P, Mejjad O, Menard JF, *et al.* Angiogenesis markers (VEGF, soluble receptor of VEGF and angiopoietin-1) in very early arthritis and their association with inflammation and joint destruction. Clin Immunol 2007; 124:158–164.

31 Khodadust F, Ezdoglian A, Steinz MM, van Beijnum JR, Zwezerijnen GJC, Jansen G, *et al.* Systematic Review: Targeted Molecular Imaging of Angiogenesis and Its Mediators in Rheumatoid Arthritis. Int J Mol Sci 2022; 23:7071.

32 Rana S, Burke SD, Karumanchi SA. Imbalances in circulating angiogenic factors in the pathophysiology of preeclampsia and related disorders. Am J Obstet Gynecol 2022; 226(2S):S1019–S1034.

33 Torres-Torres J, Espino-Y-Sosa S, Martinez-Portilla R, Borboa-Olivares H, Estrada-Gutierrez G, Acevedo-Gallegos S, *et al.* A Narrative Review on the Pathophysiology of Preeclampsia. Int J Mol Sci 2024; 25:7569.

34 Paradowska-Gorycka A, Sowinska A, Pawlik A, Malinowski D, Stypinska B, Haladyj E, *et al.* FLT-1 gene polymorphisms and protein expression profile in rheumatoid arthritis. PLoS One 2017; 12:e0172018.

35 Yuan ZC, Xu WD, Wang JM, Wu Q, Zhou J, Huang AF. Gene polymorphisms and serum levels of sVEGFR-1 in patients with systemic lupus erythematosus. Sci Rep 2020; 10:15031.

36 Chirco KR, Lewis CJ, Scheetz TE, Johnston RM, Tucker BA, Stone EM, *et al.* Evaluation of sFLT1 protein levels in human eyes with the FLT1 rs9943922 polymorphism. Ophthalmic Genet 2018; 39:68–72.

37 Xiang W, Zhuang W, Chi H, Sheng X, Zhang W, Xue Z, *et al.* Evaluating VEGFR1 genetic polymorphisms as a predisposition to AMD in a cohort from northern China. Ophthalmic Genet 2016; 37:388–393.