Editorial introductions

Current Opinion in Rheumatology was launched in 1989. It is one of a successful series of review journals whose unique format is designed to provide a systematic and critical assessment of the literature as presented in the many primary journals. The field of Rheumatology is divided into 15 sections that are reviewed once a year. Each section is assigned a Section Editor, a leading authority in the area, who identifies the most important topics at that time. Here we are pleased to introduce the Journal's Section Editors for this issue.

SECTION EDITORS

Mariana J. Kaplan

Dr Mariana J. Kaplan is Senior Investigator and Chief of the Systemic Autoimmunity Branch at the National Institute of Arthritis and Musculoskeletal and Skin Diseases, National Institutes of Health, USA.

Dr Kaplan's research has focused on identifying mechanisms of organ damage and premature atherosclerosis in systemic autoimmunity. Her laboratory focuses on identifying how innate immunity promotes loss of tolerance and end-organ damage in SLE, rheumatoid arthritis and other systemic autoimmune diseases. Dr Kaplan also has an interest in identifying novel therapeutic targets to prevent premature vascular damage in systemic autoimmune diseases, using a bench to bedside approach. She has been principal investigator in various investigator-initiated clinical trials assessing new treatment approaches in autoimmunity. Dr Kaplan has served in numerous advisory and committee roles with the American College of Rheumatology, Rheumatology Research Foundation, American Association of Immunologists and Lupus Foundation of America and has been a member of the editorial boards of many immunology and rheumatology journals. She is a member of the American Society for Clinical Investigation and the Association of American Physicians and a recipient of the Henry Kunkel Young Investigator Award and the Edmund L. Dubois Memorial Lectureship, both from the American College of Rheumatology. She has also been recipient of the Evelyn Hess Award from the Lupus Foundation of America for her significant contributions to the understanding of systemic lupus erythematosus, and the Charles L. Christian award for her research impact in understanding lupus.

Polly J. Ferguson

Polly J. Ferguson received her BS and MD from the University of Iowa, USA. She completed her pediatric residency and pediatric rheumatology and immunology fellowship at the University of Virginia, USA, where she remained to complete a post-doctoral research fellowship. She is currently a Professor of Pediatrics and is the Director of Pediatric Rheumatology at the University of Iowa. Her NIH funded research program is focused on the etiology of autoinflammatory bone disorders including chronic recurrent multifocal osteomyelitis (CRMO). She has published more than 60 peer reviewed publications in addition to multiple book chapters and reviews.

Cardiovascular disease in systemic lupus erythematosus: an update

Yudong Liu and Mariana J. Kaplan

Purpose of review
The mechanisms leading to the development of premature atherosclerosis and vascular injury in systemic lupus erythematosus (SLE) remain to be fully elucidated. This is a comprehensive review of recent research developments related to the understanding of cardiovascular disease (CVD) in lupus.

Recent findings
SLE patients with lupus nephritis display significantly increased risk of myocardial infarction and CVD mortality than SLE patients without lupus nephritis. SLE disease-related parameters could be taken into consideration when calculating CVD risks. The type I interferon pathway is detrimental to the vasculature and may contribute to the development of insulin resistance. The level of low-density granulocytes, a distinct subset of proinflammatory neutrophils present in SLE, was independently associated with coronary plaque burden and endothelial dysfunction. Invariant natural killer T cells may promote an atheroprotective effect in SLE patients with asymptomatic atherosclerotic plaques. Oxidized lupus high-density lipoprotein promotes proinflammatory responses in macrophages.

Summary
Recent discoveries have further strengthened the critical role of SLE-related immune dysregulation and metabolic disturbances in promoting accelerated CVD. Understanding how these pathogenic factors promote vascular injury may provide better molecular candidates for therapeutic targeting, and ultimately to improve CVD outcomes.

Keywords
atherosclerosis, cardiovascular disease, immune dysregulation, systemic lupus erythematosus

EPIDEMIOLOGY OF VASCULAR DISEASE IN SYSTEMIC LUPUS ERYTHEMATOSUS

Systemic lupus erythematosus (SLE) is associated with a significant risk of cardiovascular disease (CVD) [1,2]. A recent study suggests that patients with SLE have two-fold higher number of atherosclerotic plaques in the carotid and femoral arteries, comparable with what has been reported in rheumatoid arthritis and diabetes mellitus, two other conditions associated to enhanced CV risk [3▪]. Another recent prospective study reported that during a 5-year follow-up period, 32% of SLE patients developed evidence of carotid atherosclerosis compared with 4% of healthy controls [4▪]. In addition, individuals with SLE have a two-fold increased rate of ischemic stroke or myocardial infarction (MI) compared with the general population [1,5]. In some patients with lupus, MI may develop even before the diagnosis of SLE or shortly thereafter, suggesting a potential link between autoimmune inflammation and atherosclerosis [6]. SLE patients with lupus nephritis display significantly increased risk of MI and CVD mortality than SLE patients without lupus nephritis [7▪]. Furthermore, lupus nephritis is associated with twice as often evidence of carotid atherosclerotic plaques when compared with age-matched nonnephritis SLE patients [even those with positivity for antiphospholipid (aPL) antibodies] and population controls [8]. The prevalence of CV events in SLE also shows racial and ethnic variations. Specifically, the risk of MI was recently reported to be lower among Hispanics and Asians compared with Whites, whereas the risk

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KEY POINTS

- SLE is associated with a significant risk of CVD, which is likely related to the immune dysregulation characteristic of this disease.
- Both dysregulation of innate and adaptive immune responses likely contribute to accelerated CVD in SLE.
- The levels of LDGs and a LDG gene signature significantly associates with vascular disease in SLE.
- A number of biomarkers have been recently demonstrated as potential surrogates for CVD in SLE.
- The type I interferon pathway fuels detrimental effects on the vasculature and interrupting this pathway may improve CVD outcomes in SLE.

of stroke was elevated among blacks and Hispanics compared with Whites [9▪].

Traditional Framingham risk factors do not fully explain the increased CVD risk in SLE [10]. A recent study compared the Framingham score with the recently described SLE-specific CVD risk equation (SLE score) and identified that a large proportion of SLE patients could be reclassified as high CVD risk using a formula that incorporates SLE disease-related parameters [11▪]. The authors found that the sex preference in SLE and low BMI in women may lead the traditional Framingham score to underestimate the CV risk in female SLE patients. In contrast, the SLE score may capture those patients as having high risk for CVD [11▪].

Genetic variants can play important roles in both SLE and CVD. A recent study indicates that an interleukin 19 (IL19) risk allele, rs17581834(T) is associated with stroke/MI in SLE by affecting protein binding. SLE patients with that risk allele had increased levels of plasma-IL10 and aPL antibodies [12]. In addition, an SRP54 Antisense RNA 1-AS1 risk allele, rs799454(G) was associated with stroke/transient ischemic attack in SLE [12]. Another recent study shows that apolipoprotein L1 risk variants associate with atherosclerotic disease in African-American SLE patients [13].

RECENT ADVANCES IN PATHOGENESIS OF PREMATURE ATHEROSCLEROSIS IN SYSTEMIC LUPUS ERYTHEMATOSUS

Vascular damage and cardiovascular disease in systemic lupus erythematosus

Endothelial dysfunction is one of the first recognized steps leading to established CVD. A recent study reported a high rate of endothelial dysfunction in individuals with recent onset of SLE (<5 years), even those with mild disease activity and without traditional CVD risk factors [14]. Various soluble adhesion molecules, such as vascular cell adhesion molecule, which are released after endothelial cell damage and have been proposed as markers of endothelial dysfunction, are increased in SLE and correlate with higher coronary calcium scores [15]. Nhek *et al.* [16▪] recently showed that SLE sera can induce platelet activation leading to endothelial cell activation and synthesis of proinflammatory mediators in an IL-1-dependent manner (Fig. 1). A profound imbalance between endothelial cell damage and repair has been identified in SLE [17]. Thus, patients with SLE have impaired endothelial cell and compromised repair of the damaged endothelial cells, which may promote the development of vascular plaque.

Arterial stiffness, as a marker of subclinical atherosclerosis, is significantly elevated in patients with SLE. A low level of cardiorespiratory fitness (CRF) has been shown to associate with the risk of CVD in the general population [18]. A recent study examined the association of CRF with arterial stiffness in SLE. CRF was inversely associated with pulse wave velocity (PWV), a marker for arterial stiffness [19,20], suggesting that CRF may attenuate the age-related arterial stiffening in SLE and contribute to primary prevention of CVD in SLE [20]. In contrast, a recent meta-analysis failed to show significant effects of exercise on CVD risk factors and disease activity, but reported that exercise improves cardiorespiratory capacity and reduces fatigue in SLE [21]. By utilizing PWV as a marker of arterial stiffness, Castejon *et al.* [22] reported that SLE patients with metabolic syndrome display increased arterial stiffness, which is associated with a decreased percentage of circulating endothelial progenitor cells (EPCs). In contrast, a recent study failed to demonstrate an association between EPC colonies, percentages of circulating EPCs, or SLE disease activity index with PWV [23]. These results indicate that additional longitudinal studies in larger cohorts of SLE patients are needed to conclusively assess the role of various biomarkers in vascular dysfunction in this patient population.

Dysregulation of the innate immune response and systemic lupus erythematosus-related cardiovascular disease

Owing to the central role of type I interferons (IFNs) in SLE, these cytokines have been extensively investigated as a contributing factor to the development of

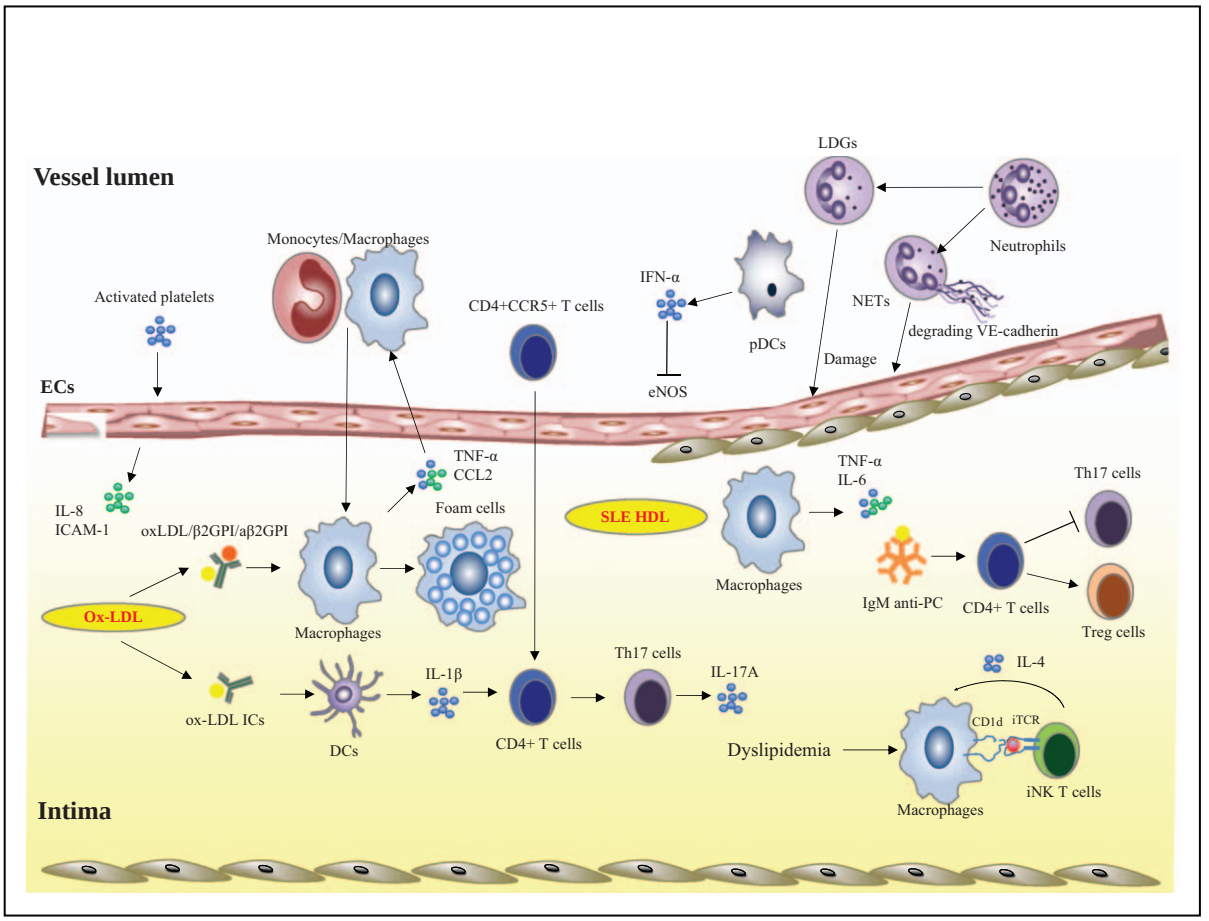

FIGURE 1. Recent developments in the understanding of the mechanisms of vascular risk in SLE. A number of pathogenic mechanisms contribute to the accelerated atherosclerosis and vascular injury in SLE and were recently highlighted. aβ2-GPI, antiβ2-glycoprotein I antibodies; CCL, C-C Motif Chemokine Ligand; EC, endothelial cell; eNOS, endothelial nitric oxide synthase; HDL, high-density lipoprotein; ICAM, intercellular adhesion molecule 1; IgM anti-PC, IgM antibodies against phosphorylcholine; ICs, immune complex; iTCR, invariant TCR; LDL, low-density lipoprotein; NETs, neutrophil extracellular traps; oxLDL, oxidized LDL; pDCs, plasmacytoid dendritic cells; SLE, systemic lupus erythematosus; VE-cadherin, vascular endothelial cadherin.

lupus-related CVD. Lupus patients with a high type I IFN signature have decreased endothelial function [24]. Enhanced serum IFN activity has been significantly associated with decreased endothelial function, whereas factors such as serum levels of high-sensitivity CRP, adhesion molecules, and lupus disease activity are not. This suggests that enhanced type I IFN signaling may be particularly important in driving increased CV risk in SLE [25]. Tyden *et al.* [26] recently reported that activation of the type I IFN system in SLE may impair endothelial function even in those lupus patients with low disease activity. Diminished activity of endothelial nitric oxide synthase (eNOS) and loss of nitric oxide production are critical in the development of endothelial dysfunction. One of the detrimental effects of IFN-α on endothelial dysfunction was recently reported by Buie *et al.* [27▪] as this cytokine was reported to inhibit eNOS expression at the mRNA and protein levels and to impair insulin-mediated nitric oxide production in endothelial cells (Fig. 1). In addition, a recent study demonstrated that diet-induced insulin resistance is initiated by a type I IFN response that triggers accumulation of cluster of differentiation (CD)8^+ T cells in the liver, resulting in glucose dysregulation and hepatic inflammation [28▪]. The pathogenic role of type I IFNs is also observed in the case of MI. King *et al.* [29▪▪] recently showed that ischemic cell death and uptake of cell debris by macrophages in the heart fuels a fatal response to MI by activating interferon regulatory factor 3 and type I IFN production through cyclic GMP-AMP synthase-stimulator of IFN genes. Further, treatment of mice with an type I Interferon receptor-neutralizing antibody after MI ablated the

IFN response and improved left ventricular dysfunction and survival [29▪▪].

A distinct subset of proinflammatory neutrophils present in lupus patients, called low-density granulocytes (LDGs), have been proposed to play pathogenic roles in lupus CVD by a variety of mechanisms, including their enhanced propensity to form neutrophil extracellular traps (NETs) [30]. A recent publication reported that NETs promote vascular leakage and endothelial-to-mesenchymal transition through the degradation of vascular endothelial cadherin and subsequent activation of β-catenin signaling, and this may promote endothelial dysfunction (Fig. 1) [31▪▪]. Carlucci *et al.* [32▪▪] recently reported that SLE patients with overall mild-moderate disease activity display a significant increase in aortic wall inflammation, as assessed by [^{18}F]-Fluorodeoxyglucose-PET/computed tomography scan, when compared with healthy controls. This same SLE cohort displayed a significant increase in noncalcified coronary plaque burden (NCB) and endothelial dysfunction. When analyzing the associations of lupus-related factors to these vascular abnormalities, the level of LDGs was independently associated with NCB [32▪▪]. In addition, an LDG gene signature obtained by RNA sequencing showed a significant association with the presence of high vascular inflammation and high NCB in SLE [32▪▪]. These results support the notion that aberrant neutrophil biology contributes to the development of premature vascular disease in SLE.

DYSREGULATION OF ADAPTIVE IMMUNE RESPONSES AND SYSTEMIC LUPUS ERYTHEMATOSUS-RELATED CARDIOVASCULAR DISEASE

T cells

As T cells play a critical role in both atherosclerosis and SLE, dysregulated T cells may contribute to SLE-associated CVD and this is also supported by animal models [33]. In patients with SLE, plasmacytoid dendritic cells induce the expansion of CXC chemokine receptor 3$^+$ CD4$^+$ T cells and their migration from the bloodstream into the arterial wall, where they may play proatherogenic roles [34]. The role of IL-17A in atherosclerosis development in SLE is unknown, but low-density lipoprotein receptor knockout mice that receive transfer of CD4$^+$ T cells from SLE-susceptible B6.Sle1.2.3 (B6.SLE) mice develop accelerated atherosclerosis because of an imbalance between IL-17 production and Treg function [35]. In a recent prospective 5-year study, increased levels of CD4$^+$CC chemokine receptor (CCR)5$^+$ T cells were independently associated with the development of carotid atherosclerosis in SLE patients [4▪]. In a recent murine atherosclerosis study, CCR5 was reported to be critical for the homing of CD4$^+$ T cells into the atherosclerotic plaque [36▪▪]. These findings support the possibility that increased levels of CD4$^+$CCR5$^+$ T cells in SLE may contribute to atherogenesis.

A recent study suggests that invariant natural killer T (iNKT) cells may promote an atheroprotective effect in SLE patients with asymptomatic atherosclerotic plaques [37▪▪]. The authors found that healthy iNKT cells differentiated in the presence of healthy monocytes and serum from SLE patients with asymptomatic plaque polarized macrophages toward an anti-inflammatory M2 phenotype, whereas SLE patients with clinical CVD had unresponsive iNKT cells and increased proinflammatory monocytes [37▪▪]. Furthermore, the authors demonstrated that the anti-inflammatory iNKT cell phenotype was associated with dyslipidemia and was driven by altered monocyte phospholipid expression and CD1d-mediated cross-talk between iNKT cells and monocytes (Fig. 1) [37▪▪].

Autoantibodies and immune complexes

Autoantibodies and immune complexes (ICs) may also contribute to vascular damage and atherosclerosis development in SLE. IgG antioxidized (ox) LDL (oxLDL) ICs are present in both rabbit and human atherosclerotic lesions [38]. In SLE, IgG anti oxLDL Abs are significantly elevated [39]. Stimulation of bone marrow-derived dendritic cells (BMDCs) with oxLDL ICs leads to enhanced secretion of IL-1β, a cytokine important in atherogenesis and inflammasome activation, whereas BMDCs stimulated with free oxLDL promote enhanced Th17 polarization (Fig. 1) [40▪▪].

aPL antibodies, including lupus anticoagulant, anticardiolipin antibodies and antiβ2-glycoprotein I antibodies (aβ2-GPI), are present in 20–30% of SLE patients and have been linked to an increased risk of venous and arterial thrombosis [41]. A significant percentage of SLE patients show a β2-GPI-specific T cell reactivity, which is associated with subclinical atherosclerosis [42]. β2-GPI binds to oxLDL to form oxLDL/β2-GPI complexes [43] that can increase foam cell formation and proinflammatory cytokine and chemokine expression [44]. Increases in oxLDL/β2-GPI and oxLDL/β2-GPI/aβ2-GPI complexes have been reported in SLE, and correlate with several CVD risk factors (Fig. 1) [45]. In contrast, immunoglobulin M (IgM) antibodies recognizing phosphorylcholine, may exert protective roles in CVD in SLE, at least in part by promoting Treg

polarization and reducing the production of IL-17 and tumor necrosis factor (TNF)-α (Fig. 1) [46▪].

Dyslipidemia in systemic lupus erythematosus

Dyslipidemia is a hallmark of atherosclerosis and CVD. In SLE, dyslipidemia is characterized by elevations in total cholesterol, LDL, triglycerides, and apolipoprotein B, and a reduction in high-density lipoprotein (HDL) [47]. This pattern is often observed at the time of lupus diagnosis and correlates to SLE activity [48]. SLE patients display increased levels of oxidized and dysfunctional HDL with impaired cholesterol efflux capacity and in association with atherosclerosis [49–51]. Mechanistically, HDL exerts vasculoprotective activities by promoting activating transcription factor 3 (ATF3), leading to downregulation of Toll-like receptor (TLR)-induced inflammatory responses [52]. In contrast, a recent study reported that oxidized lupus HDL promotes proinflammatory responses in macrophages [53▪▪]. Indeed, SLE HDL activates nuclear factor (NF)κB, promotes inflammatory cytokine production, and fails to block TLR-induced inflammation. This failure of lupus HDL to block inflammatory responses is because of an impaired ability to promote ATF3 synthesis and its nuclear translocation and this was driven by signaling through the oxidized LDL receptor (Fig. 1) [53▪▪]. Indeed, an HDL mimetic given to lupus-prone mice systemically promoted significant ATF3 induction and decreases in proinflammatory cytokine levels, supporting a putative therapeutic potential [53▪▪]. NETs were previously reported to have the ability to oxidized HDL in a region-specific proatherogenic manner that impairs the cholesterol efflux capacity of HDL [49]. In a recent study, impairments in cholesterol efflux capacity were significantly associated with vascular inflammation and NCB in multivariate analysis, suggesting that therapeutic strategies that improve HDL function may have significant cardioprotective effects in SLE [32▪▪].

Insulin resistance

Insulin resistance has been shown to contribute to CVD in SLE. A recent study assessed insulin sensitivity in SLE patients in response to a meal tolerance test. SLE patients displayed a bi-hormone metabolic abnormality characterized by increased insulin resistance and hyperglucagonemia despite normal glucose tolerance and preserved ß-cell function and skeletal muscle glucose transporter 4 translocation. The authors thus propose that strategies capable of ameliorating insulin sensitivity may require more than targeting insulin resistance alone [54].

Screening and assessing cardiovascular disease in systemic lupus erythematosus

Mavrogeni *et al.* [55] recently demonstrated that CV magnetic resonance can detect silent heart disease missed by echocardiography. Indeed, CV magnetic resonance detected abnormalities in 27.5% of SLE patients who presented normal echocardiography but had silent/past myocarditis, MI, or vasculitis [55]. Visceral adipose tissue correlates with CV risk factors. A recent study reveals that SLE is associated with increased visceral adipose tissue and altered adiposity distribution. [56]. Furthermore, aortic perivascular adipose tissue density associated with aortic calcification in SLE women, indicating that adipose tissue dysfunction may contribute to CVD in SLE [57].

Osteoprotegerin and osteopontin (OPN) are involved in vascular calcification and are upregulated in symptomatic human carotid atherosclerosis [58]. A recent study reported that serum OPN levels are significantly increased in SLE compared with healthy controls, particularly in those patients with lupus nephritis. [59]. OPN levels were significantly associated with CV events, indicating that this molecule may contribute to SLE CVD and could potentially serve as a biomarker of CV risk in this patient population [59]. In another study, although SLE patients with higher osteoprotegerin levels had higher measures of coronary artery calcium, carotid intima media thickness, and more carotid plaque, no statistically significant associations were noted after adjustment for age [60]. In addition, a recent study shows that biomarkers reflecting receptor-activated apoptosis and tissue degradation, including Fas, TNF receptor 1, TNF-related apoptosis inducing ligand receptor 2, matrix metalloproteinase-1, and matrix metalloproteinase-7, are significantly elevated in SLE patients with CVD than those without CVD [61].

Impairment of total antioxidant capacity is associated with subclinical coronary microvascular dysfunction in SLE patients without traditional CV risk factors [62]. Paraoxonase1 (PON1), an enzyme with antioxidant activity that attaches to HDL and can prevent oxidative modifications of LDL [63], is decreased in SLE and is associated with vascular damage [64]. A recent study evaluated the role of anti-PON1 and anti-HDL antibodies as biomarkers of lupus CVD. They found that anti-HDL antibodies were significantly associated with higher risk of CVD, and anti-PON1 antibodies were significantly associated with carotid intima media thickness in SLE [65]. Thus, those antibodies could be potential early biomarkers of premature atherosclerosis in SLE.

Cardiac troponin T (cTnT) has been proposed as a marker of myocyte necrosis and injury in the early phases of acute MI [66]. High-sensitivity cTnT has shown promising value in predicting CVD in the general population with apparent low CVD risk [67]. In a recent cross-sectional controlled study, Divard *et al.* [68] reported that levels of high-sensitivity cTnT were independently associated with subclinical atherosclerosis in asymptomatic SLE patients considered at low risk for CVD based on traditional risk factors.

As mentioned previously, LDGs play pathogenic roles in lupus CVD. A recent study demonstrated that LDG levels were significantly associated with NCB severity and lower cholesterol efflux capacity in SLE in an unadjusted linear regression analysis [32▪▪]. Furthermore, the authors found that a neutrophil gene signature was significantly associated with vascular disease in SLE. Indeed, some of the most upregulated genes in the high-NCB SLE groups were the genes previously found to be upregulated in LDGs when compared with normal density neutrophils [32▪▪]. Those findings suggest that the levels of LDGs may serve as a marker for CVD risk in SLE.

THERAPIES

Statins

A number of studies have indicated that statins may promote autoimmune responses [69,70]. In a recent population-based cohort study assessing the association between statin use and the risk of developing SLE, the authors failed to identify any association between current statin use with the risk of developing SLE among patients 40 years and older [71▪▪]. Instead, they observed a decreased SLE risk among current statin users who continued their therapy for more than 1 year [71▪▪]. Studies on whether statins can prevent CVD in SLE have given inconsistent results. Atorvastatin improved endothelial cell-dependent vasodilation in a short-term (8-week) trial [72], but failed to exert vasculoprotective effects in a longer (2-year) trial [73]. A recent study indicated that statin therapy might reduce the risk of mortality and CVD in SLE patients with hyperlipidemia [74]. Short-term atorvastatin therapy improved arterial stiffness in middle-aged SLE patients with abnormal PWV. Although these studies suggest that statin may benefit a subset of SLE patients, larger well-controlled, long-term trials are needed to conclude whether current statin regimens are sufficient in decreasing CV risk and what the guidelines for statin use should be in SLE.

Anti-IFN therapies and Janus kinase inhibitors

Targeting the IFN pathway has emerged as a promising therapeutic strategy in SLE [75,76]. Given these promising results and the putative role of IFN in atherogenesis, it will be important to determine whether disrupting this pathway can yield a beneficial therapeutic response in premature atherosclerosis in SLE. A recent study reported that interfering with downstream signaling of this pathway by utilizing the Janus kinase (JAK) inhibitor tofacitinib ameliorates murine lupus and its associated vascular dysfunction [77▪▪]. The role of JAK inhibitors and antitype I IFN therapies in vascular prevention in SLE remains to be determined.

Antimalarials

Antimalarials may have cardioprotective effects [78,79]. In a recent study, Fasano *et al.* [80] reported that long-term hydroxychloroquine (HCQ) use in conjunction with low-dose aspirin may provide added efficacy in primary CVD prevention in SLE. However, another recent database prospective cohort study failed to demonstrate the protective effect of long-term HCQ in reducing vascular events in SLE [81]. Ruiz-Arruza *et al.* [82] recently showed that, those SLE patients that received glucocorticoids later during the course of the disease and at lower doses while receiving more HCQ, displayed significantly decreased incidence of glucocorticoid-related CVD but similar SLE-related damage compared with SLE patients received glucocorticoids earlier and at higher doses. Additional studies are therefore needed to further define the role of antimalarials in CV prevention and the best strategies for treatment (single use versus combination therapy).

CONCLUSION

A number of SLE-specific mechanisms, such as dysfunctional immune regulation and defective endothelial cell function and vascular repair, contribute to the premature atherosclerosis in SLE. The biological insights in appreciating those complex interplays have progressed significantly, but further understanding of the clinical relevance of targeting those factors in reducing SLE-related CVD is required. In addition, continued efforts to investigate other mechanisms that lead to accelerated CVD in SLE are needed to provide better molecular candidates for therapeutic targeting, and ultimately to improve the CV outcomes.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Avina-Zubieta JA, To F, Vostretsova K, *et al.* Risk of myocardial infarction and stroke in newly diagnosed systemic lupus erythematosus: a general population-based study. Arthritis Care Res (Hoboken) 2017; 69:849–856.
2. Tselios K, Gladman DD, Su J, *et al.* Evolution of risk factors for atherosclerotic cardiovascular events in systemic lupus erythematosus: a longterm prospective study. J Rheumatol 2017; 44:1841–1849.
3. ■ Tektonidou MG, Kravvariti E, Konstantonis G, *et al.* Subclinical atherosclerosis in systemic lupus erythematosus: comparable risk with diabetes mellitus and rheumatoid arthritis. Autoimmun Rev 2017; 16:308–312.

This study suggests that SLE have comparable number of atherosclerotic plaques in the carotid and femoral arteries when compared with rheumatoid arthritis and diabetes mellitus.

4. ■ Baragetti A, Ramirez GA, Magnoni M, *et al.* Disease trends over time and CD4(+)CCR5(+) T-cells expansion predict carotid atherosclerosis development in patients with systemic lupus erythematosus. Nutr Metab Cardiovasc Dis 2018; 28:53–63.

This study shows that increased levels of $CD4^{+}CCR5^{+}$ T cells are independently associated with the development of carotid atherosclerosis in SLE.

5. Arkema EV, Svenungsson E, Von Euler M, *et al.* Stroke in systemic lupus erythematosus: a Swedish population-based cohort study. Ann Rheum Dis 2017; 76:1544–1549.
6. Urowitz MB, Gladman DD, Anderson NM, *et al.* Cardiovascular events prior to or early after diagnosis of systemic lupus erythematosus in the systemic lupus international collaborating clinics cohort. Lupus Sci Med 2016; 3:e000143.
7. ■ Hermansen ML, Lindhardsen J, Torp-Pedersen C, *et al.* The risk of cardiovascular morbidity and cardiovascular mortality in systemic lupus erythematosus and lupus nephritis: a Danish nationwide population-based cohort study. Rheumatology (Oxford) 2017; 56:709–715.

This study shows that SLE patients with lupus nephritis display significantly increased risk of cardiovascular mortality when compared with SLE patients without lupus nephritis.

8. Gustafsson JT, Herlitz Lindberg M, Gunnarsson I, *et al.* Excess atherosclerosis in systemic lupus erythematosus,-A matter of renal involvement: case control study of 281 SLE patients and 281 individually matched population controls. PLoS One 2017; 12:e0174572.
9. ■ Barbhaiya M, Feldman CH, Guan H, *et al.* Race/ethnicity and cardiovascular events among patients with systemic lupus erythematosus. Arthritis Rheumatol 2017; 69:1823–1831.

This study suggests that the prevalence of CVD in SLE shows racial/ethnic variations.

10. Esdaile JM, Abrahamowicz M, Grodzicky T, *et al.* Traditional Framingham risk factors fail to fully account for accelerated atherosclerosis in systemic lupus erythematosus. Arthritis Rheum 2001; 44:2331–2337.
11. ■ Boulos D, Koelmeyer RL, Morand EF, Hoi AY. Cardiovascular risk profiles in a lupus cohort: what do different calculators tell us? Lupus Sci Med 2017; 4:e000212.

This study demonstrates that the Framingham score underestimates CVD risk in female patients and suggests that the recently described SLE-specific CVD risk equation (SLE score) may capture patients whose CVD risk is underestimated.

12. Leonard D, Svenungsson E, Dahlqvist J, *et al.* Novel gene variants associated with cardiovascular disease in systemic lupus erythematosus and rheumatoid arthritis. Ann Rheum Dis 2018; pii: annrheumdis-2017-212614. doi: 10.1136/annrheumdis-2017-212614.
13. Blazer A, Wang B, Simpson D, *et al.* Apolipoprotein L1 risk variants associate with prevalent atherosclerotic disease in African American systemic lupus erythematosus patients. PLoS One 2017; 12:e0182483.
14. Taraborelli M, Sciatti E, Bonadei I, *et al.* Endothelial dysfunction in early systemic lupus erythematosus patients and controls without previous cardiovascular events. Arthritis Care Res (Hoboken) 2017; doi: 10.1002/acr.23495. [Epub ahead of print]
15. Rho YH, Chung CP, Oeser A, *et al.* Novel cardiovascular risk factors in premature coronary atherosclerosis associated with systemic lupus erythematosus. J Rheumatol 2008; 35:1789–1794.
16. ■ Nhek S, Clancy R, Lee KA, *et al.* Activated platelets induce endothelial cell activation via an interleukin-1? Pathway in systemic lupus erythematosus. Arterioscler Thromb Vasc Biol 2017; 37:707–716.

This study shows that SLE sera can induce platelet activation, and that these activated platelets can activate endothelial cells and promote proinflamamtory responses.

17. Rajagopalan S, Somers EC, Brook RD, *et al.* Endothelial cell apoptosis in systemic lupus erythematosus: a common pathway for abnormal vascular function and thrombosis propensity. Blood 2004; 103: 3677–3683.
18. Kodama S, Saito K, Tanaka S, *et al.* Cardiorespiratory fitness as a quantitative predictor of all-cause mortality and cardiovascular events in healthy men and women: a meta-analysis. JAMA 2009; 301:2024–2035.
19. Vlachopoulos C, Aznaouridis K, Stefanadis C. Prediction of cardiovascular events and all-cause mortality with arterial stiffness: a systematic review and meta-analysis. J Am Coll Cardiol 2010; 55:1318–1327.
20. Montalban-Mendez C, Soriano-Maldonado A, Vargas-Hitos JA, *et al.* Cardiorespiratory fitness and age-related arterial stiffness in women with systemic lupus erythematosus. Eur J Clin Invest 2018; 48: doi: 10.1111/eci.12885. [Epub 2018 Feb 2].
21. O'Dwyer T, Durcan L, Wilson F. Exercise and physical activity in systemic lupus erythematosus: A systematic review with meta-analyses. Semin Arthritis Rheum 2017; 47:204–215.
22. Castejon R, Jimenez-Ortiz C, Rosado S, *et al.* Metabolic syndrome is associated with decreased circulating endothelial progenitor cells and increased arterial stiffness in systemic lupus erythematosus. Lupus 2016; 25:129–136.
23. Korsten P, Patschan D, Henze E, *et al.* Dynamics of pulse wave velocity and vascular augmentation index in association with endothelial progenitor cells in SLE. Lupus Sci Med 2016; 3:e000185.
24. Lee PY, Li Y, Richards HB, *et al.* Type I interferon as a novel risk factor for endothelial progenitor cell depletion and endothelial dysfunction in systemic lupus erythematosus. Arthritis Rheum 2007; 56:3759–3769.
25. Somers EC, Zhao W, Lewis EE, *et al.* Type I interferons are associated with subclinical markers of cardiovascular disease in a cohort of systemic lupus erythematosus patients. PLoS One 2012; 7:e37000.
26. Tyden H, Lood C, Gullstrand B, *et al.* Endothelial dysfunction is associated with activation of the type I interferon system and platelets in patients with systemic lupus erythematosus. RMD Open 2017; 3:e000508.
27. ■ Buie JJ, Renaud LL, Muise-Helmericks R, Oates JC. IFN-alpha negatively regulates the expression of endothelial nitric oxide synthase and nitric oxide production: implications for systemic lupus erythematosus. J Immunol 2017; 199:1979–1988.

This study reports that IFN-α inhibits eNOS expression and impairs insulin-mediated nitric oxide production by endothelial cells.

28. ■ Ghazarian M, Revelo XS, Nohr MK, *et al.* Type I interferon responses drive intrahepatic T cells to promote metabolic syndrome. Sci Immunol 2017; 2: pii: eaai7616. doi: 10.1126/sciimmunol.aai7616.

This study indicates that IFN-I responses trigger CD8+ T cell accumulation in the liver, resulting in glucose dysregulation and hepatic inflammation.

29. ■■ King KR, Aguirre AD, Ye YX, *et al.* IRF3 and type I interferons fuel a fatal response to myocardial infarction. Nat Med 2017; 23:1481–1487.

This study reports that type I IFNs fuel a fatal response to MI through cyclic GMP-AMP synthase-stimulator of interferon genes pathway.

30. Knight JS, Kaplan MJ. Lupus neutrophils: 'NET' gain in understanding lupus pathogenesis. Curr Opin Rheumatol 2012; 24:441–450.
31. ■■ Pieterse E, Rother N, Garsen M, *et al.* Neutrophil extracellular traps drive endothelial-to-mesenchymal transition. Arterioscler Thromb Vasc Biol 2017; 37:1371–1379.

This study shows that NETs promote vascular leakage and endothelial-to-mesenchymal transition through the degradation of vascular endothelial cadherin and subsequent activation of β-catenin signaling.

32. ■■ Carlucci PM, Purmalek MM, Dey AK, *et al.* Neutrophil subsets and their gene signature associate with vascular inflammation and coronary atherosclerosis in lupus. JCI Insight 2018; 3:e99276.

This study shows that circulating LDG numbers and an LDG-driven gene signature significantly and independently associate with vascular inflammation and dysfunction and coronary NCB in SLE.

33. Crispin JC, Hedrich CM, Suarez-Fueyo A, *et al.* SLE-associated defects promote altered T cell function. Crit Rev Immunol 2017; 37:39–58.
34. Clement M, Charles N, Escoubet B, *et al.* CD4+CXCR3+ T cells and plasmacytoid dendritic cells drive accelerated atherosclerosis associated with systemic lupus erythematosus. J Autoimmun 2015; 63:59–67.
35. Wilhelm AJ, Rhoads JP, Wade NS, Major AS. Dysregulated CD4+ T cells from SLE-susceptible mice are sufficient to accelerate atherosclerosis in LDLr-/- mice. Ann Rheum Dis 2015; 74:778–785.
36. ■■ Li J, McArdle S, Gholami A, *et al.* CCR5+T-bet+FoxP3+ effector CD4 T cells drive atherosclerosis. Circ Res 2016; 118:1540–1552.

This study shows that CCR5 is required for CD4 T-cell homing to the atherosclerotic plaques. CCR5+T-bet+FoxP3+ effector CD4 T cells are not regulatory but pathogenic in atherosclerosis.

37. ■■ Smith E, Croca S, Waddington KE, *et al.* Cross-talk between iNKT cells and monocytes triggers an atheroprotective immune response in SLE patients with asymptomatic plaque. Sci Immunol 2016; 1: pii: eaah4081. doi: 10.1126/sciimmunol.aah4081.

This study suggests an atheroprotective role of iNKT cells in patients with SLE who have asymptomatic atherosclerotic plaques.

38. Yla-Herttuala S, Palinski W, Butler SW, *et al.* Rabbit and human atherosclerotic lesions contain IgG that recognizes epitopes of oxidized LDL. Arterioscler Thromb 1994; 14:32–40.
39. Soep JB, Mietus-Snyder M, Malloy MJ, *et al.* Assessment of atherosclerotic risk factors and endothelial function in children and young adults with pediatric-onset systemic lupus erythematosus. Arthritis Rheum 2004; 51:451–457.
40. ■■ Rhoads JP, Lukens JR, Wilhelm AJ, *et al.* Oxidized low-density lipoprotein immune complex priming of the Nlrp3 inflammasome involves TLR and FcgammaR cooperation and is dependent on CARD9. J Immunol 2017; 198:2105–2114.

This study shows that stimulation of BMDCs with oxLDL ICs leads to substantial IL-1β production.

41. Miyakis S, Lockshin MD, Atsumi T, *et al.* International consensus statement on an update of the classification criteria for definite antiphospholipid syndrome (APS). J Thromb Haemost 2006; 4:295–306.
42. Conti F, Spinelli FR, Alessandri C, *et al.* Subclinical atherosclerosis in systemic lupus erythematosus and antiphospholipid syndrome: focus on beta2GPI-specific T cell response. Arterioscler Thromb Vasc Biol 2014; 34:661–668.
43. Kobayashi K, Kishi M, Atsumi T, *et al.* Circulating oxidized LDL forms complexes with beta2-glycoprotein I: implication as an atherogenic autoantigen. J Lipid Res 2003; 44:716–726.
44. Zhang X, Xie Y, Zhou H, *et al.* Involvement of TLR4 in oxidized LDL/beta2GPI/antibeta2GPI-induced transformation of macrophages to foam cells. J Atheroscler Thromb 2014; 21:1140–1151.
45. Bassi N, Zampieri S, Ghirardello A, *et al.* oxLDL/beta2GPI complex and antioxLDL/beta2GPI in SLE: prevalence and correlates. Autoimmunity 2009; 42:289–291.
46. ■ Sun J, Lundstrom SL, Zhang B, *et al.* IgM antibodies against phosphorylcholine promote polarization of T regulatory cells from patients with atherosclerotic plaques, systemic lupus erythematosus and healthy donors. Atherosclerosis 2018; 268:36–48.

This study shows that IgM antibodies against phosphorylcholine may exert protective roles in CVD in patients of lupus by promoting polarization of Tregs, whereas reducing the production of IL-17 and TNF-α.

47. Szabo MZ, Szodoray P, Kiss E. Dyslipidemia in systemic lupus erythematosus. Immunol Res 2017; 65:543–550.
48. Borba EF, Bonfa E. Dyslipoproteinemias in systemic lupus erythematosus: influence of disease, activity, and anticardiolipin antibodies. Lupus 1997; 6:533–539.
49. Smith CK, Vivekanandan-Giri A, Tang C, *et al.* Neutrophil extracellular trap-derived enzymes oxidize high-density lipoprotein: an additional proatherogenic mechanism in systemic lupus erythematosus. Arthritis Rheumatol 2014; 66:2532–2544.
50. Ronda N, Favari E, Borghi MO, *et al.* Impaired serum cholesterol efflux capacity in rheumatoid arthritis and systemic lupus erythematosus. Ann Rheum Dis 2014; 73:609–615.
51. McMahon M, Grossman J, FitzGerald J, *et al.* Proinflammatory high-density lipoprotein as a biomarker for atherosclerosis in patients with systemic lupus erythematosus and rheumatoid arthritis. Arthritis Rheum 2006; 54:2541–2549.
52. De Nardo D, Labzin LI, Kono H, *et al.* High-density lipoprotein mediates anti-inflammatory reprogramming of macrophages via the transcriptional regulator ATF3. Nat Immunol 2014; 15:152–160.
53. ■■ Smith CK, Seto NL, Vivekanandan-Giri A, *et al.* Lupus high-density lipoprotein induces proinflammatory responses in macrophages by binding lectin-like oxidised low-density lipoprotein receptor 1 and failing to promote activating transcription factor 3 activity. Ann Rheum Dis 2017; 76:602–611.

The authors show that SLE HDL activates NFκB, promotes inflammatory cytokine production, and fails to block TLR-induced inflammation because of impaired ability of lupus HDL in promoting ATF3 synthesis and nuclear translocation.

54. Miyake CN, Gualano B, Dantas WS, *et al.* Increased insulin resistance and glucagon levels in mild/inactive systemic lupus erythematosus patients despite normal glucose tolerance. Arthritis Care Res (Hoboken) 2018; 70:114–124.
55. Mavrogeni S, Koutsogeorgopoulou L, Markousis-Mavrogenis G, *et al.* Cardiovascular magnetic resonance detects silent heart disease missed by echocardiography in systemic lupus erythematosus. Lupus 2018; 27:564–571.
56. Seguro LP, Paupitz JA, Caparbo VF, *et al.* Increased visceral adipose tissue and altered adiposity distribution in premenopausal lupus patients: correlation with cardiovascular risk factors. Lupus 2018; 27:1001–1006.
57. Shields KJ, El Khoudary SR, Ahearn JM, Manzi S. Association of aortic perivascular adipose tissue density with aortic calcification in women with systemic lupus erythematosus. Atherosclerosis 2017; 262:55–61.
58. Golledge J, McCann M, Mangan S, *et al.* Osteoprotegerin and osteopontin are expressed at high concentrations within symptomatic carotid atherosclerosis. Stroke 2004; 35:1636–1641.
59. Wirestam L, Frodlund M, Enocsson H, *et al.* Osteopontin is associated with disease severity and antiphospholipid syndrome in well characterised Swedish cases of SLE. Lupus Sci Med 2017; 4:e000225.
60. Kiani AN, Aukrust P, Ueland T, *et al.* Serum osteoprotegrin (OPG) in subclinical atherosclerosis in systemic lupus erythematosus. Lupus 2017; 26:865–870.
61. Wigren M, Svenungsson E, Mattisson IY, *et al.* Cardiovascular disease in systemic lupus erythematosus is associated with increased levels of biomarkers reflecting receptor-activated apoptosis. Atherosclerosis 2018; 270:1–7.
62. Yilmaz S, Caliskan M, Kulaksizoglu S, *et al.* Association between serum total antioxidant status and coronary microvascular functions in patients with SLE. Echocardiography 2012; 29:1218–1223.
63. Soran H, Schofield JD, Liu Y, Durrington PN. How HDL protects LDL against atherogenic modification: paraoxonase 1 and other dramatis personae. Curr Opin Lipidol 2015; 26:247–256.
64. Delgado Alves J, Ames PR, Donohue S, *et al.* Antibodies to high-density lipoprotein and beta2-glycoprotein I are inversely correlated with paraoxonase activity in systemic lupus erythematosus and primary antiphospholipid syndrome. Arthritis Rheum 2002; 46:2686–2694.
65. Lopez P, Rodriguez-Carrio J, Martinez-Zapico A, *et al.* Serum levels of anti-PON1 and anti-HDL antibodies as potential biomarkers of premature atherosclerosis in systemic lupus erythematosus. Thromb Haemost 2017; 117:2194–2206.
66. Katus HA, Remppis A, Neumann FJ, *et al.* Diagnostic efficiency of troponin T measurements in acute myocardial infarction. Circulation 1991; 83: 902–912.
67. Saunders JT, Nambi V, de Lemos JA, *et al.* Cardiac troponin T measured by a highly sensitive assay predicts coronary heart disease, heart failure, and mortality in the atherosclerosis risk in communities study. Circulation 2011; 123:1367–1376.
68. Divard G, Abbas R, Chenevier-Gobeaux C, *et al.* High-sensitivity cardiac troponin T is a biomarker for atherosclerosis in systemic lupus erythematous patients: a cross-sectional controlled study. Arthritis Res Ther 2017; 19:132.
69. de Jong HJ, Klungel OH, van Dijk L, *et al.* Use of statins is associated with an increased risk of rheumatoid arthritis. Ann Rheum Dis 2012; 71:648–654.
70. de Jong HJ, Tervaert JW, Saldi SR, *et al.* Association between statin use and lupus-like syndrome using spontaneous reports. Semin Arthritis Rheum 2011; 41:373–381.
71. ■■ De Jong HJ, van Staa TP, Lalmohamed A, *et al.* Pattern of risks of systemic lupus erythematosus among statin users: a population-based cohort study. Ann Rheum Dis 2017; 76:1723–1730.

This study shows that statin treatment does not increase the risk of developing SLE among patients older than 40 years. Instead, a decreased SLE risk among current statin users who continued their therapy for more than 1 year was reported.

72. Ferreira GA, Navarro TP, Telles RW, *et al.* Atorvastatin therapy improves endothelial-dependent vasodilation in patients with systemic lupus erythematosus: an 8 weeks controlled trial. Rheumatology (Oxford) 2007; 46:1560–1565.
73. Petri MA, Kiani AN, Post W, *et al.* Lupus Atherosclerosis Prevention Study (LAPS). Ann Rheum Dis 2011; 70:760–765.
74. Yu HH, Chen PC, Yang YH, *et al.* Statin reduces mortality and morbidity in systemic lupus erythematosus patients with hyperlipidemia: a nationwide population-based cohort study. Atherosclerosis 2015; 243:11–18.
75. Khamashta M, Merrill JT, Werth VP, *et al.* Sifalimumab, an antiinterferon-alpha monoclonal antibody, in moderate to severe systemic lupus erythematosus: a randomised, double-blind, placebo-controlled study. Ann Rheum Dis 2016; 75:1909–1916.
76. Furie R, Khamashta M, Merrill JT, *et al.* Anifrolumab, an anti-interferon-alpha receptor monoclonal antibody, in moderate-to-severe systemic lupus erythematosus. Arthritis Rheumatol 2017; 69:376–386.
77. ■■ Furumoto Y, Smith CK, Blanco L, *et al.* Tofacitinib ameliorates murine lupus and its associated vascular dysfunction. Arthritis Rheumatol 2017; 69: 148–160.

This study shows that the JAK inhibitor tofacitinib ameliorates murine lupus and its associated vascular dysfunction.

78. Ponticelli C, Moroni G. Hydroxychloroquine in systemic lupus erythematosus (SLE). Expert Opin Drug Saf 2017; 16:411–419.
79. Durcan L, Winegar DA, Connelly MA, *et al.* Longitudinal evaluation of lipoprotein variables in systemic lupus erythematosus reveals adverse changes with disease activity and prednisone and more favorable profiles with hydroxychloroquine therapy. J Rheumatol 2016; 43:745–750.
80. Fasano S, Pierro L, Pantano I, *et al.* Longterm hydroxychloroquine therapy and low-dose aspirin may have an additive effectiveness in the primary prevention of cardiovascular events in patients with systemic lupus erythematosus. J Rheumatol 2017; 44:1032–1038.
81. Hsu CY, Lin YS, Su YJ, *et al.* Effect of long-term hydroxychloroquine on vascular events in patients with systemic lupus erythematosus: a database prospective cohort study. Rheumatology (Oxford) 2017; 56:2212–2221.
82. Ruiz-Arruza I, Lozano J, Cabezas-Rodriguez I, *et al.* Restrictive use of oral glucocorticoids in systemic lupus erythematosus and prevention of damage without worsening long-term disease control: an observational study. Arthritis Care Res (Hoboken) 2018; 70:582–591.

Pulmonary manifestations of systemic lupus erythematosus and Sjögren's syndrome

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Purpose of review
Systemic lupus erythematosus (SLE) and Sjögren syndrome are chronic autoimmune inflammatory disorders that can present with multiorgan involvement including the lungs. This review will focus on recent literature pertaining to the epidemiology, pathogenesis, clinical presentation and diagnosis and management of SLE and Sjögren syndrome-associated pulmonary conditions.

Recent findings
Pulmonary manifestations of both disease entities have been well characterized and lung involvement can be observed during the course of the disease in most cases. Pulmonary manifestations of SLE and Sjögren syndrome can be classified based on anatomical site of involvement; and the large and small airways, lung parenchyma, lung vasculature, pleura and respiratory muscles can be involved. The pleura is most commonly involved in SLE, whereas the airways are most commonly involved in primary Sjögren's syndrome (pSS). Sleep disturbances have also been described in both entities.

Summary
Although further research into treatment strategies for the pulmonary complications seen in SLE and pSS is needed, the clinician should be aware of the risk factors and clinical presentation of the various pulmonary complications in SLE and pSS in order to identify patients who should be screened and/or have modifications in treatment strategies to mitigate the morbidity and mortality associated with these complications.

Keywords
bronchiolitis, interstitial lung disease, lymphocytic interstitial pneumonia, nonspecific interstitial pneumonia pulmonary hypertension, Sjögren's syndrome, systemic lupus erythematosus

INTRODUCTION

Pulmonary manifestations of systemic lupus erythematosus (SLE) and primary Sjögren's syndrome (pSS) are common and may result in significant morbidity and mortality. They may occur within every compartment of the respiratory system including the airways, lung parenchyma, pulmonary vasculature, respiratory muscles and pleura. We review the recent literature on the prevalence, risk factors, clinical, laboratory, physiologic, radiographic and pathologic findings as well as proposed treatment regimens for the myriad of pulmonary complications in both SLE and pSS.

SYSTEMIC LUPUS ERYTHEMATOSUS

SLE is a chronic inflammatory disease of autoimmune origin that can affect virtually every organ-system of the human body. It is characterized by the production of autoantibodies, particularly antinuclear antibodies (ANAs), and by formation and deposition of immune complexes resulting in end-organ damage [1]. SLE has a female predominance and tends to occur in women of childbearing age with a 10 : 1 female-to-male ratio [2]. SLE is chronic, relapsing and remitting in nature, and commonly presents with constitutional complaints along with skin, hematologic, renal, musculoskeletal and pulmonary manifestations. The American College of Rheumatology (ACR) proposed criteria for diagnosis in 1997 [3], and more recently a group of experts in SLE developed the Systemic Lupus International Collaborating Clinics classification criteria for SLE [4] (Table 1).

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KEY POINTS

- Any part of the pulmonary system may be involved in SLE and Sjögren's syndrome.
- SLE is the most common autoimmune disease to affect the respiratory system, and lupus pleuritis is the most common manifestation of SLE.
- The airways are the most frequently affected area of the respiratory system in pSS, with nonproductive cough the most common complaint.
- ALP may be a precursor of ILD in up to one-third of patients who survive.
- pSS–ILD is associated with significant morbidity and premature mortality. Nonspecific interstitial pneumonia is most commonly seen on surgical lung biopsy in pSS–ILD.
- Pulmonary hypertension due to SLE can fall under any of the five WHO categories of pulmonary hypertension. Immunosuppressive agents may be of added benefit in pulmonary arterial hypertension.
- Both patients with SLE and pSS have increased risk of malignancies. SLE patients have an overall increased risk of developing malignancies with non-Hodgkin lymphoma and lung cancer being common. Sjögren syndrome patients are at increased risk of lymphoma with mucosa-associated lymphoid tissue lymphomas as the most common sub-type.
- Sleep disorders are common in both SLE and pSS. Low vitamin D levels may be associated with poorer sleep quality in SLE.

Any part of the respiratory system can be affected including the airways, lung parenchyma, vasculature, pleura and respiratory musculature [5,6]. Pulmonary manifestations of SLE include airway disease, pleuritis (with or without effusion), inflammatory and fibrotic forms of interstitial lung disease (ILD), alveolar hemorrhage, acute lupus pneumonitis (ALP), pulmonary hypertension, shrinking lung syndrome (SLS) and thromboembolic disease, among others [7,8▪].

Epidemiology

SLE is the most common autoimmune disease to affect the respiratory system [9]. Pulmonary involvement is more common in men than in women and generally tends to manifest later in the course of the disease. Lung involvement is an important indicator of overall prognosis, being associated with a more than two-fold increase in mortality [6]. Pulmonary involvement in SLE also negatively impacts patient-reported outcomes and patient-performed outcomes [10▪▪].

The prevalence of pulmonary involvement varies depending on the criteria used to define lung abnormalities (e.g. clinical manifestations, imaging abnormalities or histopathologic evidence), with previous studies reporting a prevalence ranging between 20 and 90%. However, the higher end of the range may be overestimated due to imaging abnormalities as a consequence of infectious processes in this immunosuppressed population [11,12].

Nasal and airway involvement

Nasal involvement has been well documented in SLE [13,14]. In fact, the presence of nasal ulcers is one of the criteria for diagnosis of SLE [4]. The prevalence of nasal involvement is likely underestimated; however, a recent study found a prevalence of nasal involvement in 47.9% of patients [15▪▪]. The most common reported nasal symptoms included nasal congestion (31.5%), pruritus (26%), rhinorrhea (20.5%) and dryness (19.2%). Only 4/35 (11.45%) had nasal ulcers. Nasal endoscopy detected bleeding, mucositis (intranasal or nasopharynx), septal perforation, crusting, sinusitis, ulceration, dry mucosa and polypoidal mucosa.

Upper airway involvement in SLE is less common than in the other connective tissue diseases (CTDs) [7,16]. Manifestations can vary from mucosal inflammation to severe airway obstruction [17,18]. The reported incidence of laryngeal involvement in SLE patients varies from 0.3 to 30% [5]. Acute epiglottitis, laryngitis, tracheitis, vocal cord paralysis and cricoarytenoiditis have been reported. Presentation can be highly variable from asymptomatic to life-threatening respiratory compromise. Symptoms and physical exam findings include hoarseness, sore throat, dry cough, dyspnea and stridor. Most patients have an excellent response to mild-to-moderate doses of corticosteroids [16].

Lower airway involvement can also be seen in SLE, particularly in those patients with secondary Sjögren's syndrome (sSS). Most commonly, this is manifested by an obstructive ventilatory defect with decreased maximal expiratory flow rates (MEF25) less than 60% predicted [5]. Both follicular and obliterative bronchiolitis can be seen on pathology. Bronchiectasis and bronchial wall thickening have also been described [11,19].

Pleural involvement

Lupus pleuritis

Pleuritis (with or without pleural effusion) is the most common pulmonary manifestation of SLE affecting up to 60% of SLE patients during their lifetime and is

Table 1. Diagnostic criteria for systemic lupus erythematosus [4]

ACR criteria for classification of SLE		**SLICC criteria for classification of SLE**	
4 of 11 criteria needed for diagnosis		**4 of 17 criteria, including at least one clinical criterion and one immunologic criterion; OR biopsy-proven lupus nephritis**	
Criterion	**Definition**	**Criterion**	**Definition**
Malar rash	Fixed erythema (spares the nasolabial folds)	Acute cutaneous lupus	Lupus malar rash; bullous lupus; TEN variant of SLE; etc.; photosensitive lupus rash or subacute cutaneous lupus
Photosensitivity	Skin rash as a result of sunlight exposure	Chronic cutaneous lupus	Classic discoid rash; localized or generalized or discoid lupus/lichen planus overlap
Discoid rash	Raised patches with adherent keratotic scaling; atrophic scarring in older lesions	Nonscaring alopecia	Diffuse thinning or hair fragility with visible broken hairs
Oral ulcers	Oral or nasopharyngeal ulceration, painless	Oral/nasal ulcers	Palate, buccal, tongue or nasal ulcers. Painless
Arthritis	Nonerosive arthritis involving two or more peripheral joints	Joint disease	Synovitis involving two or more joints, or tenderness in two or more joints and at least 30 min of morning stiffness
Serositis	Pleuritis and/or pericarditis	Serositis	Pleuritis and/or pericarditis
Renal disorder	Persistent proteinuria >500 mg/24 h or cellular casts	Renal disorder	Urine protein-to-creatinine ratio (or 24-h urine protein) >500-mg protein/24 h, or red blood cell cast
Neurologic disorder	Seizures or psychosis – after excluding other causes (uremia, ketoacidosis, etc.)	Neurologic disoder	Seizures; psychosis; mononeuritis multiplex or acute confusional state (excluding other causes)
Hematologic disorder	Hemolytic anemia, leucopenia, lymphopenia, thrombocytopenia (in the absence of other offending drugs)	Hematologic disorder	Hemolytic anemia, leucopenia, lymphopenia, thrombocytopenia (in the absence of other offending drugs)
ANA	An abnormal titer of ANA by IFA or an equivalent assay. Exclude: 'drug-induced lupus' syndrome	ANA	ANA level above laboratory reference range Exclude: 'drug-induced lupus' syndrome
Immunologic disorders	Anti-DNA – antibody or anti-Sm antibody or positive finding of antiphospholipid antibody (positive anticardiolipin, lupus anticoagulant, beta 2 glycoprotein I or false-positive RPR)	Anti-Sm antibody, anti-dsDNA antibody, antiphospholipid antibodies, complement levels and Coombs test (each counts as one positive criterion).	Antiphospholipid antibodies are considered positive if there is evidence of anticardiolipin antibodies, LAC, anti-B2 glycoprotein I or false-positive RPR. Complement levels include low C3, C4 and/or CH50 Coombs test counts as positive even in the absence of hemolytic anemia

ACR, American College of Rheumatology; ANA, antinuclear antibodies; IFA, immunofixation; LAC, lupus anticoagulant; OR, odds ratio; RPR, rapid plasma regain; TEN, toxic epidermal necrolysis; SLE, systemic lupus erythematosus; SLICC, Systemic Lupus International Collaborating Clinics classification.

one of the diagnostic criteria for SLE [3,8▪]. Autopsy studies demonstrate pleural involvement in 93% of patients [5]. Symptoms include pleuritic chest pain, cough, dyspnea and fever. Pertinent physical exam findings include a transient pleural friction rub and dullness to percussion. Chest imaging may reveal effusions; however, absence of effusion does not rule out the diagnosis as inflammation of the pleura may cause chest pain in the absence of effusion. Conversely, some patients may have an asymptomatic pleural effusion [11].

Pleural effusions in SLE tend to be bilateral, small to moderate in size and rarely involve more than two-thirds of the thoracic cavity [20]. It is essential to determine the underlying cause of pleural effusions; heart failure, parapneumonic effusions, malignancy and pulmonary embolism among others need to be excluded. Nevertheless, a recent Thai study of 119 patients with 127 episodes of pleuritis/pleural effusion reported SLE-related pleural effusion as the most common cause of pleural effusion is SLE patients followed by transudative effusion related to heart failure and effusions related to infection [20].

Pleural fluid may vary from serous to bloody. Effusions are generally exudative with an elevated

lactate dehydrogenase. The total nucleated cell count is typically elevated and may reveal a neutrophilic or lymphocytic predominance depending upon the chronicity of the effusion. The pleural fluid glucose is generally low, but not as low as in patients with rheumatoid arthritis [6]. Other findings on pleural fluid analysis include a positive ANA, elevated anti-double-stranded DNA antibodies, low complement levels and lupus erythematosus cells noted on cytology. A pleural fluid ANA titer at least 1 : 160 has a sensitivity of 92% for SLE pleuritis when compared with all other causes for pleural effusions [19]. A pleural fluid to serum ANA ratio at least 1 strongly is highly suggestive as well. In the absence of SLE, an elevated ANA may be seen in paraneoplastic pleurisy [11]. Pleural biopsy might be needed in selected cases; however, it is not routinely performed [20].

Longer disease duration, late age of diagnosis of SLE (after age 50 years), greater cumulative damage and concomitant seropositivity for ribonucleoprotein (RNP) and Sm antibodies are factors that increase the risk of pleuritis by nearly two-fold [7]. Similarly, a recent review of 2390 SLE patients in the Hopkins Lupus Cohort identified fever, Raynaud phenomenon and anti-DNA antibodies as predictors for both pericarditis and pleurisy [21▪]. Data from the multicenter Latin American SLE cohort Grupo Latino Americano De Estudio del Lupus [22▪▪] suggests that nonischemic heart disease, Lupus severity of disease index more than 1 and anti-La antibody positivity at diagnosis or early in the disease course might be predictive of subsequent development of pleuropulmonary manifestations, whereas cutaneous manifestations were negatively associated. They however could not corroborate the association of anticardiolipin (aCL) antibodies, anti-RNP, anti-Ro, anti-Sm antibodies with pulmonary manifestations as described in previous investigations [5,19].

Treatment options are selected based on symptom severity and size of pleural effusion. Mild cases of SLE pleuritis typically respond to NSAIDs. Low-to-moderate doses of corticosteroids (e.g. 20–40 mg daily) are recommended for moderate-to-severe symptoms. Corticosteroid therapy may be discontinued in 3–4 weeks depending on clinical response. Pleurodesis can be considered for refractory cases, but is rarely required [6,11].

Parenchymal involvement

Acute lupus pneumonitis

ALP is a rare pulmonary manifestation with an estimated incidence of 1–4% [11,23]. It is characterized by diffuse alveolar inflammation with occasional hyaline membrane formation without evidence of vasculitis or hemorrhage. This complication is more likely to occur during an SLE flare with multisystem involvement including nephritis, serositis and arthritis [6]. Elevated anti-SSA/Ro antibodies have been associated with development of ALP [11].

ALP may be a presenting manifestation of SLE [11] or drug-induced SLE [24]. Its clinical presentation is generally nonspecific with fever, pleuritic chest pain, dyspnea, cough and rarely hemoptysis [12,23]. In patients without a history of SLE, the diagnosis of ALP can be challenging; clues to its diagnosis include lack of improvement with empiric antibiotics for pneumonia and extrapulmonary features of SLE (malar rash, alopecia, oral ulcers, serositis, nephritis, cytopenias, etc.) [23]. Patients are frequently hypoxemic with bibasilar crackles on examination. Radiologic features of ALP include bilateral alveolar infiltrates predominantly in the lower lung fields, although unilateral infiltrates have also been described. Associated pleural effusions may be present in up to 50%.

Bronchoalveolar lavage (BAL) is useful to evaluate for alternative causes such as alveolar hemorrhage and infection. Computed tomography (CT) angiography may be needed to exclude pulmonary embolism. Lung biopsy is typically reserved for patients that fail to respond to immunosuppressive therapy or in patients with a probable alternate diagnosis [5,6].

Treatment recommendations are based on clinical experience and case reports. Corticosteroids are the cornerstone of treatment for ALP with prednisone 1–2 mg/kg/day in divided doses as the initial agent of choice. Lack of improvement in 72 h and need for mechanical ventilation justify the need for intravenous pulse glucocorticoids (methylprednisolone 1 g/day for 3 consecutive days) [6]. Steroid sparing immunosuppressive agents should be started concomitantly (e.g. cyclophosphamide, azathioprine, mycophenolate mofetil) and intravenous immunoglobulin and/or plasmapheresis may be used in refractory cases [11,25]. ALP mortality rate has been reported as high as 50% [9,19,23] and may be a precursor of ILD in up to one-third of those who survive [26▪▪].

Chronic interstitial lung disease

SLE-associated ILD (SLE–ILD) is uncommon affecting approximately 1–15% of patients [27▪▪] and is usually less severe than ILD in other CTDs [19]. Risk factors for SLE–ILD include a history of ALP, longstanding disease (>10 years), Raynaud phenomenon, anti-U1 RNP antibodies, sclerodactyly and abnormal nailfold capillaroscopy [5].

Nonspecific interstitial pneumonia (NSIP) (both cellular and fibrotic) appears to be the most common pattern seen histopathologically and/or radiographically in SLE–ILD [11,27▪▪], but usual interstitial pneumonia (UIP), organizing pneumonia, lymphocytic interstitial pneumonia (LIP), follicular bronchiolitis and nodular lymphoid hyperplasia have also been reported [28].

Patients typically present with insidious onset of dyspnea resulting in decreased exercise tolerance and a nonproductive cough; however, some patients may be asymptomatic. Physical examination typically reveals basilar crackles [8▪]. Clubbing and peripheral cyanosis, which are more commonly found in idiopathic pulmonary fibrosis, are rarely seen in SLE–ILD.

Laboratory abnormalities seen in SLE–ILD include but are not limited to hypocomplementemia, elevated high-sensitivity C-reactive protein and cryoglobulins [19]. A weak association with anti-Ro/SSA antibodies has also been reported [11].

The most common pulmonary function test (PFT) finding in SLE–ILD is a diminished diffusion capacity for carbon monoxide (DLCO); other findings may include a restrictive pattern and/or hypoxia especially with exertion [27▪▪]. A radiographic pattern of NSIP is the most common finding on chest CT. However, the differential of ground glass opacities (GGO) includes infection, pulmonary edema, diffuse alveolar hemorrhage (DAH) and drug toxicity [6,29▪].

Treatment consists of systemic corticosteroids (prednisone 0.5–1 mg/kg/day). Steroid sparing agents commonly used in other forms of CTD–ILD (e.g. cyclophosphamide, azathioprine, mycophenolate, rituximab) have also been used for SLE–ILD, although data in SLE is limited and most recommendations come from experts [5,19,26▪▪] as there are no clinical trials in SS-ILD [27▪▪]. For ILD flares, intravenous methylprednisolone 0.5–1 g for 3 days may be needed. The prognosis of SLE–ILD is usually better than idiopathic ILDs and other CTD-associated ILDs. These patients tend to have a slow course and stabilize/improve over time.

Diffuse alveolar hemorrhage

DAH is a rare and potentially life-threatening complication of SLE affecting approximately 2% of patients. The hallmark of the disease is damage to the alveolar–capillary basement membrane in conjunction with alveolar inflammation. Risk factors for DAH include the presence of antiphospholipid antibodies (APLs) [30] and the presence of active lupus nephritis with hypocomplementemia [19].

Although DAH may be the initial manifestation of SLE, it tends to occur more frequently among patients who are already known to have lupus. Clinically, it is characterized by the triad of hemoptysis, acute anemia and diffuse alveolar infiltrates; however, hemoptysis may be absent in one-third of patients. Other symptoms and manifestations may include fever, cough, dyspnea and chest pain, hypoxemia and an increased diffusion capacity for carbon monoxide [31].

BAL is a key and preferred method for diagnosing DAH and is characterized by progressively hemorrhagic aliquots with the presence of hemosiderin laden macrophages. Lung biopsy is rarely needed for diagnosis, but if it is performed, it may show capillaritis with immune complex deposition or bland hemorrhage [31,32].

Due to the life-threatening nature of the disease, aggressive treatment with high-dose corticosteroids should be initiated promptly (methylprednisolone 0.5–1 g for 3 consecutive days or prednisone 1–2 mg/kg/day in more stable patients) followed by another immunosuppressive agent (cyclophosphamide, azathioprine, rituximab or mycophenolate) [19,31–33]. The choice of the second agent is based on disease severity, for example need for mechanical ventilation, partial or null response to intravenous steroids and so on [31]. There have also been case reports in which recombinant factor VII was successfully used for pulmonary hemorrhage refractory to standard treatment [33]. Plasmapheresis has been used, although it has not been proven to decrease mortality rates [25,32]. Finally, stem-cell transplantation has also been reported [32].

Twenty years ago, mortality rates for DAH were reported as high as 70% [34]. Recent data have shown improvement in mortality (~30–40%) perhaps related to increased use of immunosuppressive agents in addition to corticosteroids [32,35].

Respiratory muscle involvement

Shrinking lung syndrome

First described by Hoffbrand and Beck [36] in 1965 as 'unexplained dyspnea' and shrinking lungs in SLE, the SLS, also known as the 'vanishing lung' syndrome, is a rare pleuropulmonary manifestation of SLE characterized by diaphragmatic dysfunction and a restrictive pattern on pulmonary function testing with normal pulmonary parenchyma. The estimated prevalence ranges between 0.5 [37] and 10% [38,39] depending on the method and criteria used for diagnosis.

Although the cause of SLS remains controversial, multiple theories exist behind the mechanism for SLS including failure of surfactant production resulting in microatelectasis, diaphragmatic

weakness or dysfunction with decreased transdiaphragmatic pressure, diaphragmatic myopathy due to corticosteroid use, phrenic neuropathy and pleural inflammation leading to impaired inspiration with decreased lung volume [22▪▪,38,40].

Factors associated with SLS include greater disease duration, seropositivity for anti-RNP antibodies and a history of pleuritis [19]. An association with anti-Ro/SSA antibodies has been debated [41▪▪]. SLS can be the presenting manifestation of SLE or it can present up to 2–3 decades later [38].

Clinically, patients present with progressive dyspnea that characteristically is worse when supine. Sixty-five percent of patients with SLS also have pleuritic chest pain [41▪▪]. Physical examination may reveal decreased breath sounds at the bases sometimes with associated crackles. Abdominal paradox has been described in some cases [42].

Currently, there are no standardized criteria for the diagnosis of SLS. Pulmonary function testing reveals a progressive restrictive ventilatory defect. A decrease in forced vital capacity (FVC) from sitting to supine position may also aid in diagnosis [42]. The DLCO is also often reduced. Chest radiograph and high resolution computed tomography (HRCT) reveal elevation of the hemidiaphragms, low lung volumes, basilar atelectasis, and approximately 20% of patients have pleural thickening or small pleural effusions [37,41▪▪]. The main utility of HRCT in SLS is ruling out parenchymal, interstitial or vascular lung disease. Evaluation of diaphragmatic strength is particularly useful in confirming the diagnosis of SLS. A decrease in maximal inspiratory pressure and maximal expiratory pressure is often seen, indicating global respiratory muscle weakness [43]. Electromyography with phrenic nerve conduction studies can be normal or show a decreased or absent response to stimulation [38]. Ultrasonography and fluoroscopy have also been used for diaphragmatic evaluation. The Sniff test (have the patient 'sniff' or quickly breathe in through the nose) is used to evaluate diaphragmatic motion, which is often affected in SLS patients. A paradoxical response (diaphragm moving upward with inspiration and downward with expiration) has been observed in up to 46% of SLE–SLS patients; however, a normal ultrasound does not rule out the diagnosis [41▪▪]. Dynamic contrast-enhanced lung MRI has been reported as an alternative for diagnosis [40,41▪▪].

There is no evidence supporting the use of immunosuppressive therapy for SLS, although there are case series using corticosteroids, mycophenolate mofetil, azathioprine, methotrexate, cyclophosphamide and rituximab [5,6,19,41▪▪,44,45]. Noninvasive positive pressure ventilation at night may also be beneficial [42].

Overall, the long-term prognosis is usually good with modest improvement in some cases and minor worsening occurring rarely. In a case series [38] of 55 SLE–SLS patients with a median follow-up period of 35 ± 29 months (range from 6 months to 9 years), 44 patients (80%) had significant clinical improvement with stabilization or modest improvement on PFTs after treatment, and in 10 patients, (18%) there was no functional sequelae. One patient (1.8%) died due to pneumonia, concomitant active lupus nephritis and poor medication adherence. These findings correlate with the data reported by Langenskiöld *et al.* [46] in which 20% (7/35) of patients recuperated normal lung function. Duron and Cohen-Aubart also performed a review of the literature with focus on treatment and prognosis of 155 SLE–SLS patients. Outcomes were known in 52 cases (33.5%), clinical improvement was seen in 48 patients (94%), functional improvement in 77% of cases (36/47) and imaging resolution/improvement was seen in 57% of cases (8/14).

Vascular involvement

Pulmonary hypertension

SLE is the second most common cause of CTD-related pulmonary hypertension only after the scleroderma spectrum of disease [47]. Pulmonary hypertension is a progressive and symptomatic disease characterized by an elevation in pulmonary artery pressures, which leads to right ventricular (RV) failure and death. Pulmonary hypertension is defined by a mean pulmonary artery pressure of at least 25 mmHg [48] and requires a right heart catheterization (RHC) to make the diagnosis.

The WHO divides pulmonary hypertension into five different groups based on different disease mechanisms, clinical presentation and histopathology. Patients with SLE can have pulmonary hypertension from any group or a combination of groups. Group 1, pulmonary arterial hypertension (PAH), is characterized by a lesion confined to the pulmonary arteriole. PAH also requires a normal pulmonary capillary wedge pressure (PCWP) ($\leq$15 mmHg). In group 2, the increased pulmonary artery pressures are secondary to left heart disease, namely due to systolic and diastolic dysfunction and left-sided valvular disorders. The PCWP is more than 15 mmHg in this group. Disorders of the respiratory system and hypoxemia causes group 3 pulmonary hypertension. Group 4 is secondary to chronic thromboembolic pulmonary hypertension (CTEPH). Lastly, pulmonary hypertension secondary to vasculitis has been described in SLE patients and currently falls under group 5 [49].

Histological appearances are similar in idiopathic PAH (IPAH) and SLE–PAH [33]. Some of the pathological changes that can be seen include but are not limited to progressive arteriolar remodeling, vascular smooth muscle hypertrophy, adventitial and neointimal proliferation and in-situ thrombosis [50–53].

Endothelial damage is thought to be a key triggering factor in the pathogenesis of SLE–PAH. In one early study, antiendothelial antibodies were associated with the development of SLE–PAH [28], and evidence from animal models suggests that these antibodies may be pathogenic [54]. Others have found that endothelin levels are higher in SLE patients with PAH than in those without [29▪]. Immunoglobulin and complement deposits have been found in the pulmonary arteries of lupus patients along with the characteristic PAH vasculopathy suggesting a role for an immune complex deposition process in the pathogenesis of SLE–PAH that does not occur in PAH without CTD [4]. RhoA/Rho kinase has also been implicated in the pathophysiology of pulmonary hypertension, and Rho kinase has been shown to be upregulated in SLE patients compared with healthy controls [26▪▪]. A recent meta-analysis of 31 publications by Stéphane *et al.*, with a total of 4480 SLE patients, found that APL-positive SLE patients had a higher prevalence of PAH (12.3%) versus APL-negative SLE patients (7.3%). The overall pooled odds ratio (OR) for PAH was 2.28 [95% confidence interval (CI), 1.65–3.15] [30]. The risk of PAH was the highest amongst lupus anticoagulant-positive and IgG aCL antibody-positive patients. The presence of these antibodies creates a well known prothrombotic state [30] that can cause a four-to-five-fold increase in the risk of deep vein thrombosis and pulmonary emboli thus making SLE patients also at risk for CTEPH.

The exact prevalence of pulmonary hypertension has not been well established. Many of the case series of SLE patients with pulmonary hypertension have used echocardiography alone for diagnosis; this is a significant limitation as RHC is needed for diagnosis. For instance, in a recent Latin-American cohort of 424 SLE patients with pleuropulmonary manifestations; 2.2% (32 patients) had pulmonary hypertension [22▪▪]. Other studies have reported prevalences as low as 0.5% or as high as 14% depending on the diagnostic modality used [50,54,55▪▪,56]. Most studies using RHC for pulmonary hypertension diagnosis report an incidence less than 4% [47,55▪▪].

Clinical presentation depends on the type of pulmonary hypertension based on the WHO classification; the most common symptoms include progressive dyspnea, chest pain, peripheral edema and sometimes syncope. SLE patients with group 2 pulmonary hypertension can also have paroxysmal nocturnal dyspnea and orthopnea. A nonproductive cough associated with pleuritic chest pain can be seen in patients with pulmonary hypertension secondary to ILD (group 3 pulmonary hypertension).

Laboratory abnormalities or associations have been described in SLE-pulmonary hypertension patients. The study by Hachulla *et al.* [55▪▪] suggested that anti-U1-RNP antibodies might be a protective factor regarding survival in SLE–PAH patients, while suggesting that the presence of anti-SSA/SSB antibodies may be a risk factor for PAH. Conversely, a recent Chinese meta-analysis that included 12 studies identified anti-RNP antibody and anti-Sm antibody as risk factors for SLE-associated PAH with the pooled OR 3.68 (95% CI 2.04–6.63, $P < 0.0001$) and 1.71 (95% CI 1.06–2.76, P1/40.03), respectively. Another interesting association was reported by Castillo-Martıez *et al.* [57], who recently suggested that chronic hyperuricemia may predict the future development of pulmonary hypertension in SLE patients with normal pulmonary artery systolic pressure (PASP) at baseline.

According to the 2015 European Society of Cardiology (ESC)/European Respiratory Society (ERS) Guidelines for the diagnosis and treatment of pulmonary hypertension, pulmonary hypertension can be inferred if multiple different echocardiographic measurements are consistent with the diagnosis; however, when treatment modalities are being considered, echocardiography alone is not sufficient to support this treatment decision and RHC is needed [48]. Nevertheless, there have been some proposed utilities to echocardiographic evaluation. A post-hoc analysis by Hübbe-Tena *et al.* [58] from a cohort of patients with SLE followed over 6 years suggested that ESC/ERS/International Society for Heart and Lung Transplant echocardiography-based definitions of pulmonary hypertension can be useful to predict 6-year mortality in SLE patients. A Japanese cohort of SLE patients was followed over 5 years to investigate the usefulness of 6-minute walk (6-MW) stress echocardiography in detecting early pulmonary hypertension. Results suggested that 6-MW stress echocardiography might provide an incremental prognostic value of pulmonary hypertension development in CTD [59].

Once the diagnosis of PAH has been established by RHC, therapy should be started without delay. The treatment of SLE-pulmonary hypertension can be complex and most times requires a combination of immunosuppressive therapy along with vasodilators. The most commonly used vasodilators include the phosphodiesterase-5 inhibitors (PD5 inhibitors), soluble guanylate cyclase stimulators, endothelin receptor antagonists (ERAs), prostacyclin analogs and

prostacyclin receptor agonists. A meta-analysis of nine clinical trials of CTD–PAH reported improvement in 6-MW distance with these vasodilators. The improvement with PD5 inhibitors, ERAs and prostacyclin analogs was 37–47 min, 14.1–21.7 min and 21–108 min, respectively [47].

Increasing evidence supports the use of immunosuppressive therapy in SLE–PAH; however, there are no established guidelines to guide treatment. There have been a number of case series that reported improvement in hemodynamics and functional capacity [60,61]. Most patients were treated with intravenous cyclophosphamide for 3–6 months with or without corticosteroids, and vasodilators. In these studies, patients that responded to immunosuppressive therapy tended to have less severe disease manifested by better functional capacity, higher cardiac index and/or lower pulmonary vascular resistance index at presentation. Patients are likely to respond earlier in the course of their disease potentially because those with longstanding disease can have irreversible vascular damage [62].

It has been well described that patients with CTD–PAH have poorer prognosis when compared with IPAH patients. However, SLE–PAH patients have better survival curves that patients with scleroderma associated PAH [63▪▪]. Results from the French registry reported that patients with SLE–PAH have an overall 3-year and 5-year survival rate of 89.4 and 83.9%, respectively, and anti-U1-RNP antibodies were associated with a higher survival rate [55▪▪]. The REVEAL registry reported a 2-year survival rate of 85% and a Chinese cohort reported a 3-year survival rate of 88%.

Thromboembolic disease

It has been well established that SLE patients have an increased risk of venous thromboembolic (VTE) disease as a consequence of chronic inflammation and/or the presence of APLs. Yusuf *et al.* [64] found that amongst hospitalized SLE patients, the risk for VTE was estimated to be 1.23 (95% CI 1.15–1.32) when compared with hospitalized patients without autoimmune disease. A recent Danish nationwide cohort study [65] comparing the risk of VTE in patients with cutaneous lupus erythematosus (CLE) and SLE and the general population between 1997 and 2011 found that both CLE and SLE were significant risk factors for VTE with hazard ratio 1.39 (95% CI 1.10–1.78) for CLE and hazard ratio 3.32 (95% CI 2.73–4.03) for SLE.

Malignancy

SLE is associated with an overall increase in malignancies with female patients being 10% more susceptible than their male counterparts [66]. A recent meta-analysis by Wu *et al.* of 12 studies involving a total of 57 890 SLE patients showed that there is an increased risk of lung cancer in the SLE population (OR = 1.60; 95% CI: 1.44–1.77; $P < 0.00001$).

Abnormal B-cell activation and proliferation may lead to B-cell malignancies, which may explain why non-Hodgkin lymphoma (NHL) is one of the malignancies most commonly associated with SLE [6,19]. Immunosuppressive therapy-related toxicity is also implicated in the increased risk for malignancy due to resulting cellular dysregulation and mutation.

Sleep disorders

It has been estimated that the prevalence of sleep disorders in SLE patients is as high as 85%, which is higher than that of the general population [67]. Studies in SLE have estimated the prevalence of sleep apnea to be 26–50% and periodic limb movement to be 23–50% [68,69]. Daytime sleepiness was reported in up to 51% of patients and is associated with fatigue, somnolence and daytime dysfunction [70]. Data on other sleep disorders, such as insomnia, are not available [71].

Some of the studies that have evaluated sleep in SLE patients have found increased sleep latency and decreased sleep efficiency with high levels of fragmentation and increased number of awakenings during the night when compared with healthy controls [69,72]. SLE patients have increased alpha wave intrusions into nonrapid eye movement sleep, a decrease in stages 3 and 4 of slow-wave deep sleep and a compensatory increase in stage 1 sleep [68,70]. The evidence on sleep disturbances and SLE suggest that these disturbances may be associated with worse lupus activity [73].

Although sleep disturbances are a major health problem in SLE patients [71], their underlying mechanisms are still unclear [73]. Recent studies have suggested that pain, fatigue, disease activity and psychological health disturbances, particularly depression, may play a role [71,74]. A recent retrospective analysis of 63 women with SLE assessed the role of vitamin D on sleep quality and found that low vitamin D levels in SLE patients were associated with poorer sleep quality independent of demographics, disease severity, psychological-related factors and time of the year [71].

SJÖGREN'S SYNDROME

Sjögren syndrome is a chronic inflammatory disease characterized by mononuclear cell infiltration of exocrine glands, classically the salivary and lacrimal glands. Sjögren's syndrome was first described in

Table 2. Diagnostic criteria for Sjögren's syndrome [77]

	American-European Consensus Criteria for Sjögren's Syndrome (AECG criteria)	American College of Rheumatology Criteria for Sjögren's Syndrome (ACR criteria)
Symptoms	I. Ocular: symptoms of dry eyes for at least 3 months, foreign body sensation in the eyes and/or use of artificial tears 3 or more times per day (at least one) II. Oral: symptoms of dry mouth for at least 3 months, recurrent or persistently swollen salivary glands, need for liquids to swallow dry foods (at least one)	Does not include criteria based upon symptoms of glandular manifestations
Signs	III. Ocular: abnormal Schirmer's test (without anesthesia; $\leq$5 mm/5 min) or positive vital dye staining of the eye surface IV. Oral: unstimulated whole salivary flow ($\leq$1.5 ml in 15 min), abnormal parotid sialography or abnormal salivary scintigraphy	i. OSS $\geq$ 3 (assuming that individual is not currently using daily eye drops for glaucoma and has not had corneal surgery or cosmetic eyelid surgery in the last 5 years) *t*
Serology	V. Positive serum anti-SSA (Ro) or anti-SSB (La), or both	ii. Positive serum anti-SSA (Ro) and/or anti-SSB (La) antibody testing OR a positive rheumatoid factor and an antinuclear antibody titer $\geq$1 : 320
Biopsy	VI. Lip biopsy showing focal lymphocytic sialoadenitis (focus score $\geq$1 per 4 mm^2)	iii. Presence of focal lymphocytic sialadenitis with a focus score $\geq$1 focus/4 mm^2 in labial salivary gland biopsy samples

AECG criteria: for diagnosis of pSS: any four of the six criteria (including serology and biopsy) or any three of the four objective criteria (all exclusion criteria met). For diagnosis of sSS: previous CTD-related diagnosis, the presence of one symptom, and two of the three objective criteria (III, IV and VI). ACR criteria require two of three criteria for diagnosis of SS (no difference between pSS versus sSS). *t* OSS, ocular staining score. This score is the sum of a 0–6 score for fluorescein staining of the cornea and a 0–3 score for lissamine green staining of the conjunctiva. ACR, American College of Rheumatology; AECG, American-European Consensus Criteria for Sjögren's Syndrome; CTD, connective tissue disease; OR, odds ratio; pSS, primary Sjögren's syndrome; sSS, secondary Sjögren's syndrome.

1926 by Henri Gougerot a French dermatologist. The estimated worldwide incidence of Sjögren syndrome is seven cases per 100 000 with higher rates reported in Europe and Asia [75].

Sjögren syndrome is divided into primary and secondary forms. pSS occurs alone, whereas sSS occurs in association with another CTD. The focus on this review will be on the pulmonary manifestations of pSS.

The etiopathogenesis includes genetic, environmental and hormonal factors. It is more commonly diagnosed in the 4th or 5th decade of life with a female-to-male ratio of 9 : 1 [76]. The ACR in collaboration with the Sjögren's International Collaborative Clinical Alliance research group revised criteria for diagnosis of Sjögren syndrome in 2012 [77] (Table 2).

Lung involvement in Sjögren's syndrome has been well described, and negatively impacts quality of life and increases mortality for patients with pSS [6,78]. The reported prevalence of lung involvement varies widely (9–75%) depending on the methods of detection and patient selection [79]. However, studies that define pulmonary involvement by respiratory symptoms associated with abnormal PFTs and/or abnormal imaging findings on HRCT [80] suggest a prevalence rate in the range of 9–22%. Some of the well described risk factors for lung disease in pSS include hypergammaglobulinemia, lymphopenia, a positive rheumatoid factor, anti-Ro/SSA and anti-La/SSB antibodies, a decreased FVC and forced expiratory volume in 1 s, history of smoking, male sex and increased age [78].

Airway involvement

The airways are likely the most frequently involved area of the respiratory system in pSS. Upper and lower airway involvement have been described in pSS and can be related either to destruction of exocrine glands or due to cell infiltration. Histopathologically, infiltration of CD4-positive T lymphocytes have been demonstrated in bronchial and bronchiolar mucosa even in the absence of imaging abnormalities or symptoms, and BAL may reveal CD4-positive lymphocytic alveolitis in up to 55% of patients with pSS.

Airway disease may involve the nasal and oral mucosa, trachea (xerotrachea), bronchi (xerobronchitis) or bronchioles [6,81▪▪]. Most pSS patients complain of a nonproductive cough; those with lower airway disease may complain of dyspnea and wheezing.

Cough

Cough is one of the most commonly reported complaint by pSS patients with a prevalence reported between 41 and 61%, and it significantly impacts

the quality of life in over 50% of pSS. A nonproductive cough may precede the diagnosis of pSS by several years. The pathogenesis has been hypothesized to be related to airway dryness, inflammation, hyperresponsiveness and gastroesophageal reflux [81▪▪]. The severity of cough correlates with tracheal dryness. Treatment options include hypertonic (3% or 7%) nebulized saline and secretagogues such as pilocarpine and cevimeline.

Bronchiolitis

The term bronchiolitis refers to inflammation of the small airways, particularly the bronchioles, which are the airways that lead to the alveoli. Bronchiolitis is likely the most frequent airway complication in pSS. It may be isolated or associated with ILD, such as NSIP or LIP. Previous studies have shown bronchiolitis in up to 12% of pSS patients on surgical lung biopsy, and when new anatomical and radiographical criteria were used, this rate increased to 24% [82] (Fig. 1).

The most common symptoms seen with bronchiolitis are dry cough, dyspnea, wheezing and recurrent infections. On lung biopsy, follicular bronchiolitis appears to be the most common pattern seen in up to approximately 25–75% of cases [83,84]. Follicular bronchiolitis refers to the presence of hyperplastic lymphoid follicles with reactive germinal centers distributed along the bronchovascular bundles. Other types of bronchiolitis include chronic bronchiolitis with fibrosis, obliterative bronchiolitis, lymphocytic bronchiolitis and panbronchiolitis [85].

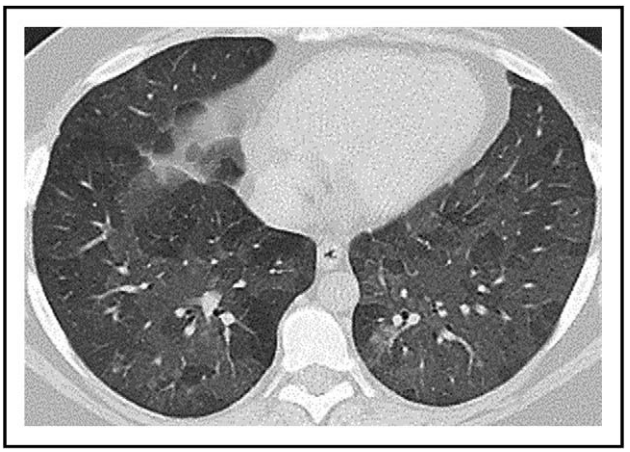

FIGURE 1. High-resolution chest computed tomography in patient with primary Sjögren's syndrome and bronchiolitis obliterans. Imaging reveals mosaic attenuation with patchwork areas of increased attenuation with lobular low attenuation suggestive of air trapping.

Treatment options include inhaled corticosteroids, rituximab and/or chronic macrolide therapy. However, these recommendations are based on low levels of scientific evidence. The prognosis of bronchiolitis is typically good with most patients improving by symptoms, imaging and PFTs; however, cases of severe bronchiolitis have been described [83,86].

Bronchiectasis

Bronchiectasis is characterized by permanent enlargement or dilatation of the airways. Patients most commonly present with chronic cough with increased mucus production. Other symptoms include dyspnea, wheezing, hemoptysis and chest pain. Nail clubbing may be seen in advanced disease. The frequency of bronchiectasis in pSS varies from 7 to 54% and is typically cylindrical as assessed by CT [19]. An older age at time of diagnosis, female sex, presence of a hiatal hernia and an elevated anti-smooth muscle antibody are associated with a higher incidence of bronchiectasis in pSS. Significantly, there is a lower frequency of anti-SSA antibody [8▪,87,88▪].

Parenchymal involvement

Interstitial lung disease

ILD is associated with significant morbidity and premature mortality in pSS [83]. Although older studies suggest a prevalence of ILD in 70% of patients with pSS [19], more recent studies suggest the prevalence is 3–11% [89]. In the largest series, 21 of 263 pSS-patients (8%) were diagnosed with ILD [90]. The time of onset of ILD in pSS is variable, and ILD may frequently precede the extrapulmonary manifestations of pSS or be diagnosed concomitantly [90]. Risk factors for pSS–ILD include the presence of high titer ANA, rheumatoid factor, anti-SSA and anti-SSB antibodies, hypergammaglobulinemia older age, the presence of Raynaud phenomenon, peripheral arthritis or esophageal involvement. Patients with these risk factors should be screened for subclinical ILD to identify patients that may benefit from earlier treatment [79,81▪▪,90]. Older age and esophageal involvement have also been associated with ILD deterioration [91]. A potential biomarker is endostatin. Higher mean levels of serum endostatin are seen in pSS compared with healthy controls, and patients with ILD have higher levels compared with those without respiratory involvement [92▪].

The most common clinical manifestations of pSS–ILD reported by European League Against Rheumatism-Sjögren Syndrome Task Force include

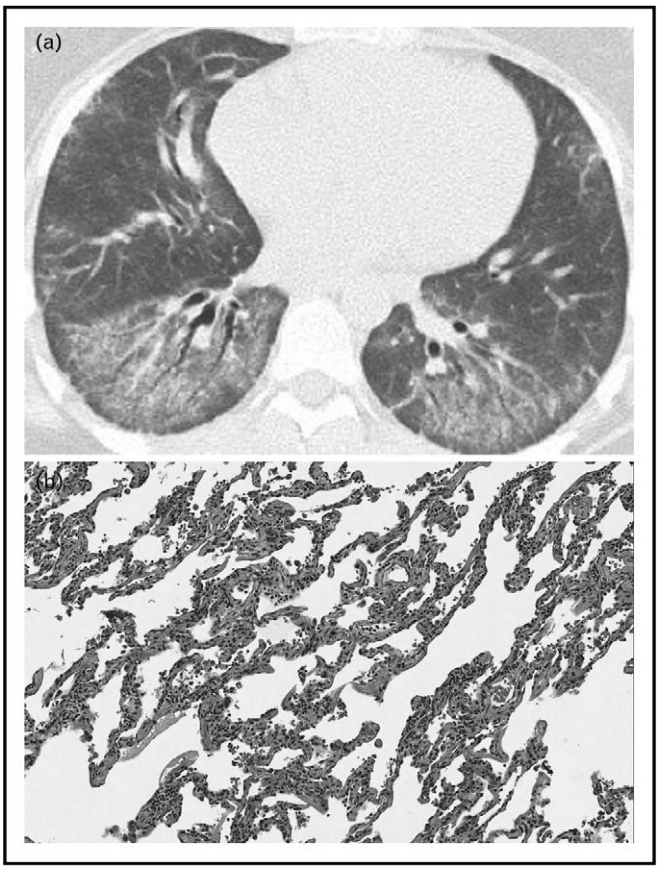

FIGURE 2. (a) High-resolution chest computed tomography of patient with primary Sjögren's syndrome and fibrotic nonspecific interstitial pneumonia. Imaging reveals lower lobe predominant ground glass opacity with traction bronchiectasis and subpleural sparing. (b) Surgical lung biopsy from patient with primary Sjögren's syndrome and nonspecific interstitial pneumonia, cellular type: alveolar walls with scattered lynmphocytes and evidence of fibrosis. (Hematoxylin and eosin, 100×).

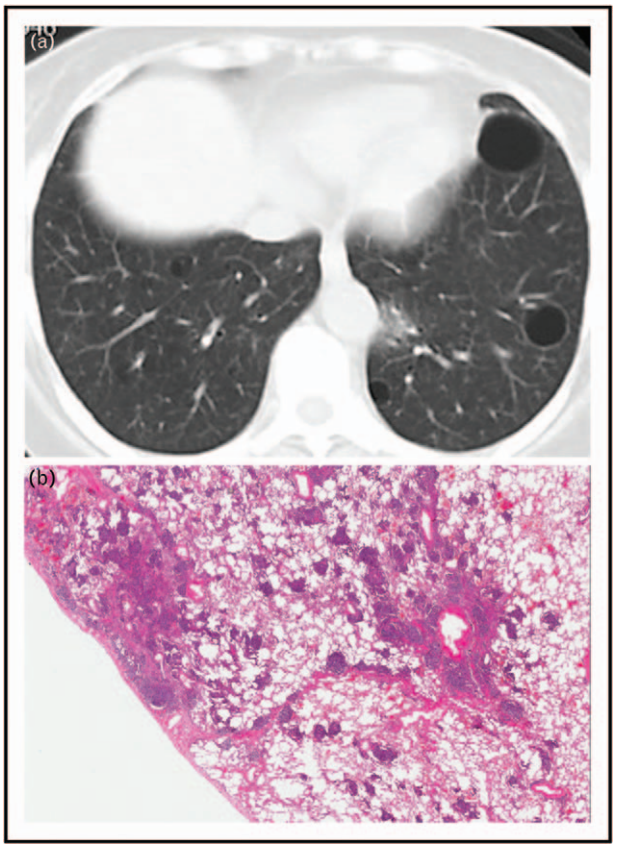

FIGURE 3. (a) High-resolution chest computed tomography of patient with primary Sjögren's syndrome and lymphocytic interstitial pneumonia. Imaging reveals lower lobe predominant perivascular subpleural cysts. (b) Surgical lung biopsy from patient with primary Sjögren's syndrome and lymphocytic interstitial pneumonia: multiple foci of benign chronic lymphocytic inflammation extending from the peribronchiolar area to the interstitium. (Hematoxylin and eosin, 15×).

dyspnea, a nonproductive cough, crackles, chest pain and fever. PFTs are usually abnormal when ILD is discovered and typically reveal a restrictive pattern with a decrease in DLCO (64% of pSS patients) an obstructive pattern (21% of patients) or a mixed restrictive and obstructive pattern (25% of patients) [79].

HRCT remains the most sensitive method for detection of parenchymal abnormalities but may also detect changes of uncertain clinical significance. Imaging abnormalities often do not correlate with PFTs or respiratory symptoms [83]. On HRCT scans, bronchial wall thickening (8–68%), nodules (6–29%), bronchiectasis (5–46%), air trapping (32%), ground-glass opacification and consolidation are the most common abnormalities [91]. Specific patterns on HRCT correlate with pulmonary histopathology. NSIP has primarily ground-glass opacities and irregular linear opacities with subpleural sparing in a peripheral and lower lobe distribution [81▪▪] (Fig. 2a). UIP is also lower lobe predominant and peripheral. It is characterized by intralobular septal thickening, honeycombing and traction bronchiectasis with the absence of significant ground glass. It is associated with increased mortality [81▪▪,93]. LIP is characterized by interlobular septal and bronchovascular bundle thickening, centrilobular and subpleural nodules, cysts and ground-glass opacities (Fig. 3a). Cysts tend to be thin-walled, vary in size and have geographic simplification of parenchymal architecture producing a 'dissolving lung appearance' [29▪,94]. Cysts can also be seen in lymphoma or amyloidosis; therefore, it is essential to make the appropriate diagnosis [83]. In organizing pneumonia, HRCT typically has patchy areas of consolidation, areas

of ground-glass attenuation and centrilobular nodules [6,78,95].

BAL shows lymphocytic alveolitis in up to 65% of patients, even amongst those that are asymptomatic and portends a poorer prognosis than when alveolitis is absent.

Despite the classic association of pSS and LIP, NSIP appears to be the most common pathological subtype revealed by lung biopsy [80,82,89,96]. Both the cellular and fibrotic forms of NSIP may occur [5,95]. Histological features of NSIP include interstitial inflammation and fibrosis in a uniform appearance [28] (Fig. 2b). UIP is uncommon in pSS. When present, histopathology reveals temporal and spatial heterogeneity with normal lung alternating with fibrotic lung. Fibroblastic foci and honeycombing are also seen [28,95]. Approximately 25% of LIP cases are associated with pSS, and it is estimated that 1% of pSS patients will develop LIP during their lifetime [7,29▪]. LIP is characterized by benign proliferation of lymphoid cells and varying numbers of plasma cells along the alveolar septae [81▪▪,95] (Fig. 3b). Lung biopsy and assessment of clonality are essential to exclude the diagnosis of lymphoma. LIP can coexist with follicular bronchiolitis and both have the potential to progress to lymphoma [78]. Histology reveals T and B lymphocyte-rich infiltrates along with plasma cells in the interstitial septa and in the alveolar space [6,28,95]. Organizing pneumonia is characterized by alveolar fibroblast infiltration on a background of chronic inflammation in the surrounding alveoli. Histology frequently reveals NSIP-organizing pneumonia overlap [28].

Corticosteroids tend to be the mainstay of therapy followed by immunosuppressive agents such as hydroxychloroquine, azathioprine, cyclosporine, mycophenolate, cyclophosphamide, rituximab tumor necrosis factor antagonists and chlorambucil, which have been used with variable results [29▪]. Recently cases of severe pSS–ILD have been reported that were successfully treated with tociluzumab, tacrolimus and abatacept [97].

Outcomes are variable and unpredictable and are contingent on histopathology. The 5-year survival rate of NSIP in pSS is estimated to be 39–83% depending upon the case series [83,89,98]. UIP has a high risk of progression and is typically not response to immunosuppressive therapy, although no clinical trials have been performed to adequately evaluate response [6,83]. LIP and organizing pneumonia are usually corticosteroid responsive and have a good prognosis. However, fibrotic changes with cyst formation and honeycombing can occur in LIP [29▪,81▪▪]. Acute exacerbations of underlying ILD have been reported in up to 16% of pSS patients, and new areas of GGO or consolidation should raise the concern for an acute exacerbation versus infection.

Vascular involvement

Pulmonary hypertension

Pulmonary hypertension due to PAH, pulmonary veno-occlussive disease, valvular heart disease or ILD has been described in pSS-patients [78]. pSS can precede the diagnosis or can also present concurrently. The exact prevalence is unknown as there are limited studies that have assessed cardiac filling pressures by RHC [56,99]. Several case series have shown a mild elevation in RV systolic pressure as measured by echocardiography in approximately 20–25% of patients with pSS [8▪,100▪]. The described cases in the literature are composed of predominantly Asian women in their 30s or 40s presenting with dyspnea, cough and chest pain. Patients with pSS-PAH are more likely to have Raynaud phenomenon, cutaneous vasculitis and ILD [6]. They are also more likely to have a positive ANA, anti-Ro/SSA antibody, anti-RNP antibody, positive rheumatoid factor and hypergammaglobulinemia. Hypocomplementemia and cryoglobulinemia have been shown to be predictors of PASP [95].

Most pSS patients with PAH receive standard therapy for PAH (ERAs, PD5 inhibitors, prostanoids) with or without immunosuppressants; however, there is no valid evidence upon which to recommend immunosuppressive therapy as the primary treatment for pSS-related PAH [56].

Thromboembolic disease

Like other patients with connective tissue disorders, patients with pSS are at increased risk for VTE. This is thought to occur in response to chronic inflammation or due to the increased prevalence (~30%) of positive APLs [78,101]. A recent meta-analysis on the risk of VTE in rheumatic disorders revealed a cumulative VTE incidence of 2.18% (95% CI: 0.79–3.57%) in pSS patients [65]. Similarly, a Scandinavian Group that examined the incidence of thromboembolic episodes in 90 pSS patients reported an incidence rate of 3.3% over a 4.6-year follow-up [65].

Malignancy

Patients with pSS are at increased risk of developing malignancies [78]. The most common lung malignancies associated with Sjögren's syndrome are NHLs; of these, the most common subtype is mucosa-associated lymphoid tissue (MALT). pSS

patients have a 16–44-fold increased risk of developing NHL, and 4–8% of pSS patients will develop lymphoma in the course of their disease [102]. The prevalence of primary pulmonary lymphoma is 1–2% in pSS patients; other common sites include lymph nodes, salivary glands and lacrimal glands [95].

Radiographic abnormalities include solitary or multiple nodules or masses with areas of consolidation or ground glass opacification. Lymphadenopathy and pleural effusions may also occur. A recent report of seven pSS patients with pulmonary MALT lymphoma reported an older age at diagnosis (mean 66), a female predominance, and nonspecific symptoms at presentation such as cough and dyspnea [81▪▪]. All patients were ANA-positive and anti-SSA/Ro-positive. Cryoglobulinemia, hypocomplementemia, parotid enlargement, palpable purpura and lymphopenia (particularly a decreased CD4+ and a low CD4+/CD8+ ratio) have been associated with an increased risk of NHL. MALT lymphoma tends to have a favorable course with an estimated 5-year survival with adequate hematological treatment of 65–90% [81▪▪,103].

NHL must be distinguished from pseudolymphoma, or pulmonary nodular lymphoid hyperplasia, which is a benign lesion characterized by plasma cell and mature polyclonal lymphocyte infiltration as determined by immunohistochemical and molecular studies [95]. Pseudolymphoma is more commonly seen in patients with lone sicca syndrome and usually responds to corticosteroid treatment. It rarely progresses to lymphoma.

Pulmonary amyloidosis

Pulmonary amyloidosis is a rare complication of Sjögren's syndrome that primarily occurs in female patients (95%). Patients may present with cough, dyspnea, hemoptysis and pleuritic chest pain. Thrombocytopenia, cryoglobulinemia, Raynaud phenomenon, lymphoma and APL syndrome are also reported. The most common imaging findings include nodules with or without calcification [81▪▪,95] and often on a background of LIP. The diagnosis usually requires surgical lung biopsy as lymphoma needs to be excluded. Nodular (AL) amyloidosis is the most common pathologic finding. No specific therapy has been reported to be successful; however, corticosteroids have been used in some patients [29▪].

Sarcoidosis

Sarcoidosis is systemic disease with multiorgan involvement characterized by the presence of noncaseating granulomas. The salivary and lacrimal gland may be involved and patients may experience sicca symptoms similar to pSS sometimes resulting in misdiagnosis. Nevertheless, the coexistence of sarcoidosis and pSS has been documented in many reports in the literature [104]. The estimated prevalence of sarcoidosis in patients with pSS is 1–2% [105]. In a case series of 59 patients with both diseases, 60% received their diagnosis of sarcoidosis and pSS simultaneously. The main features on presentation in this series included sicca symptoms, parotid enlargement, articular involvement, cutaneous involvement, respiratory symptoms and fatigue. Patients with sarcoidosis and coexisting pSS had a higher frequency of ANA, rheumatoid factor and anti-Ro/SSA antibodies [104].

Sleep disorders

Fatigue is one of the most common complaints of pSS patients; and sleep disturbances in pSS have been well documented [106]. A recent systematic review on the prevalence of sleep disturbances in pSS patients reported increased daytime somnolence, subjective sleep disturbances (including those related to sicca symptoms) and increased number of night awakenings. Further studies with polysomnography are needed to determine if there is a higher prevalence of sleep apnea [107▪▪].

CONCLUSION

In conclusion, any part of the respiratory system may be involved in SLE and pSS, the pleura is most commonly involved in SLE, whereas the airways are most commonly involved in pSS. The clinician should be aware of the risk factors and clinical presentation of the various pulmonary complications in SLE and pSS to identify patients who should be screened and/or have modifications in treatment strategies to mitigate the morbidity and mortality associated with these complications.

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REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Stojan G, Petri M. Epidemiology of systemic lupus erythematosus. Curr Opin Rheumatol 2018; 30:144–150.
2. Tsokos GC. Systemic lupus erythematosus. N Engl J Med 2016; 2:16040.
3. Scussel Lonzetti L, Joyal F, Raynauld JP, *et al.* Updating the American College of Rheumatology preliminary classification criteria for systemic sclerosis: addition of severe nailfold capillaroscopy abnormalities markedly increases the sensitivity for limited scleroderma. Arthritis Rheum 2001; 44:735–736.
4. Petri M, Orbai A-M, Alarcón GS, *et al.* Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. Arthritis Rheum 2012; 64:2677–2686.
5. Memet B, Ginzler EM. Pulmonary manifestations of systemic lupus erythematosus. Lung 2007; 1:441–450.
6. Mira-Avendano IC, Abril A. Pulmonary manifestations of Sjögren syndrome, systemic lupus erythematosus, and mixed connective tissue disease. Rheum Dis Clin North Am 2015; 41:263–277.
7. Mittoo S, Fell C. Pulmonary manifestations of systemic lupus erythematosus. Semin Respir Crit Care Med 2014; 35:249–254.
8. ■ Jawad H, McWilliams SR, Bhalla S. Cardiopulmonary manifestations of collagen vascular diseases. Curr Rheumatol Rep 2017; 19:71.

Nice review of the cardiopulmonary complications of collagen vascular diseases including ILD, pulmonary hypertension and malignancy.

9. Alamoudi OSB, Attar SM. Pulmonary manifestations in systemic lupus erythematosus: association with disease activity. Respirology 2015; 20:474–480.
10. ■■ Fidler L, Keen K, Touma Z, Mittoo S. Impact of pulmonary disease on patient-reported outcomes and patient-performed functional testing in systemic lupus erythematosus. Lupus 2016; 25:1004–1011.

Important review on the effect of pulmonary disease on patient-reported outcomes and patient-performed outcome in systemic lupus erythematosus (SLE) patients. Medical Outcome Short Form-36 Health Survey, Pittsburgh Sleep Quality Index, fatigue severity scale, Borg dyspnea scale, patient dyspnea and cough rates were reported. This article suggests that the six minute walk test may be a promising tool in the assessment of pulmonary disease in SLE patients.

11. Tselios K, Urowitz MB. Cardiovascular and pulmonary manifestations of systemic lupus erythematosus. Curr Rheumatol Rev 2017; 13:206–218.
12. Carmier D, Marchand-Adam S, Diot P, Diot E. Respiratory involvement in systemic lupus erythematosus. Rev Mal Respir 2010; 27:e66–e78.
13. Alobid I, Guilemany JM, Mullol J. Nasal manifestations of systemic illnesses. Curr Allergy Asthma Rep 2004; 4:208–216.
14. Bruno E, Russo S, Nucci R, *et al.* Nasal mucosal involvement in systemic lupus erythematosus: histopathologic and immunopathologic study. Int J Immunopathol Pharmacol 2000; 13:39–42.
15. ■■ Kusyairi KA, Gendeh BS, Sakthiswary R, *et al.* The spectrum of nasal involvement in systemic lupus erythematosus and its association with the disease activity. Lupus 2016; 25:520–524.

One of the few articles that discusses the spectrum of nasal involvement in SLE and its association with disease activity. A higher frequency of symptoms and findings was seen among patients with greater disease activity.

16. Karim A, Ahmed S, Siddiqui R, *et al.* Severe upper airway obstruction from cricoarytenoiditis as the sole presenting manifestation of a systemic lupus erythematosus flare. Chest 2002; 121:990–993.
17. Teitel AD, MacKenzie CR, Stern R, Paget SA. Laryngeal involvement in systemic lupus erythematosus. Semin Arthritis Rheum 1992; 22:203–214.
18. Langford CA, Van Waes C. Upper airway obstruction in the rheumatic diseases. Rheum Dis Clin North Am 1997; 23:345–363.
19. Mittoo S, Fell CD. Pulmonary manifestations of systemic lupus erythematosus. Semin Respir Crit Care Med 2014; 35:249–254.
20. Palavutitotai N, Buppajarntham T, Katchamart W. Etiologies and outcomes of pleural effusions in patients with systemic lupus erythematosus. J Clin Rheumatol 2014; 20:418–421.
21. ■ Ryu S, Fu W, Petri MA. Associates and predictors of pleurisy or pericarditis in SLE. Lupus Sci Med 2017; 4:e000221.

Very interesting work that provides further insight on associations and predictors of pleurisy versus pericarditis in whites and African-Americans with SLE. Long-term follow-up suggests that serositis is not as benign as once was thought.

22. ■■ Haye Salinas MJ, Caeiro F, Saurit V, *et al.* Pleuropulmonary involvement in patients with systemic lupus erythematosus from a Latin American inception cohort (GLADEL). Lupus 2017; 26:1368–1377.

The *Grupo Latino Americano De Estudio del Lupus* provides accurate and novel information on pleuropulmonary manifestations, associated and protective factors in Latin American patients with SLE.

23. Wan SA, Teh CL, Jobli AT. Lupus pneumonitis as the initial presentation of systemic lupus erythematosus: case series from a single institution. Lupus 2016; 25:1485–1490.
24. Holman SK, Parris D, Meyers S, Ramirez J. Acute low-dose hydralazine-induced lupus pneumonitis. Case Rep Pulmonol 2017; 2017:2650142.
25. Mulhearn B, Bruce IN. Indications for IVIG in rheumatic diseases. Rheumatology (Oxford) 2015; 54:383–391.
26. ■■ Atzeni F, Gerardi MC, Barilaro G, *et al.* Interstitial lung disease in systemic autoimmune rheumatic diseases: a comprehensive review. Expert Rev Clin Immunol 2018; 14:69–82.

A comprehensive review on the pathogenesis of connective tissue disease (CTD) associated-ILD and its distinguishing features in different rheumatic diseases. Clinical aspects and treatment options are also discussed with attention given to emergent treatments.

27. ■■ Mathai SC, Danoff SK. Management of interstitial lung disease associated with connective tissue disease. BMJ 2016; 352:h6819.

A thorough work that briefly discusses the differences in ILD between the different CTDs with a focus on management.

28. Vivero M, Padera RF. Histopathology of lung disease in the connective tissue diseases. Rheum Dis Clin North Am 2015; 41:197–211.
29. ■ Panchabhai TS, Farver C, Highland KB. Lymphocytic interstitial pneumonia. Clin Chest Med 2016; 37:463–474.

Nicely describes the clinical presentation and pathologic findings of lymphocytic interstitial pneumonia and characterizes some of the clues to distinguish this entity from lymphoma.

30. Zuily S, Domingues V, Suty-Selton C, *et al.* Antiphospholipid antibodies can identify lupus patients at risk of pulmonary hypertension: a systematic review and meta-analysis. Autoimmun Rev 2017; 16:576–586.
31. Martínez-Martínez MU, Abud-Mendoza C. Diffuse alveolar hemorrhage in patients with systemic lupus erythematosus. Clinical manifestations, treatment, and prognosis. Reumatol Clin 2014; 10:248–253.
32. Ednalino C, Yip J, Carsons SE. Systematic review of diffuse alveolar hemorrhage in systemic lupus erythematosus: focus on outcome and therapy. J Clin Rheumatol 2015; 21:305–310.
33. Alabed IB. Treatment of diffuse alveolar hemorrhage in systemic lupus erythematosus patient with local pulmonary administration of factor VIIa (rFVIIa): a case report. Medicine (Baltimore) 2014; 93:e72.
34. Liu MF, Lee JH, Weng TH, Lee YY. Clinical experience of 13 cases with severe pulmonary hemorrhage in systemic lupus erythematosus with active nephritis. Scand J Rheumatol 1998; 27:291–295.
35. Andrade C, Mendonça T, Farinha F, *et al.* Alveolar hemorrhage in systemic lupus erythematosus: a cohort review. Lupus 2016; 25:75–80.
36. Hoffbrand BI, Beck ER. 'Unexplained' dyspnoea and shrinking lungs in systemic lupus erythematosus. Br Med J 1965; 1:1273–1277.
37. Deeb M, Tselios K, Gladman D, *et al.* Shrinking lung syndrome in systemic lupus erythematosus: a single-centre experience. Lupus 2018; 27: 365–371.
38. Borrell H, Narváez J, Alegre JJ, *et al.* Shrinking lung syndrome in systemic lupus erythematosus: a case series and review of the literature. Medicine (Baltimore) 2016; 95:340–345.
39. Rolla G, Brussino L, Bertero MT, *et al.* Respiratory function in systemic lupus erythematosus: relation with activity and severity. Lupus 1996; 5:38–43.
40. Nemec M, Pradella M, Jahn K, *et al.* Magnetic resonance imaging-confirmed pleuritis in systemic lupus erythematosus-associated shrinking lung syndrome. Arthritis Rheumatol 2015; 67:1880.
41. ■■ Duron L, Cohen-Aubart F, Diot E, *et al.* Shrinking lung syndrome associated with systemic lupus erythematosus: a multicenter collaborative study of 15 new cases and a review of the 155 cases in the literature focusing on treatment response and long-term outcomes. Autoimmun Rev 2016; 15:944–1000.

Review that focuses of response to treatment, complications and long-term outcomes of SLE patients with shrinking lung syndrome. Also reports some of the serologies found in the reported patients. Compares new data from 15 new patients to prior reported findings.

42. Panchabhai TS, Bandyopadhyay D, Highland KB, *et al.* A 26-year-old woman with systemic lupus erythematosus presenting with orthopnea and restrictive lung impairment. Chest 2016; 149:e29–e33.
43. Munoz ML, Gelber AC, Houston BA. Into thin air: shrinking lung syndrome. Am J Med 2014; 127:711–713.
44. Goswami RP, Mondal S, Lahiri D, *et al.* Shrinking lung syndrome in systemic lupus erythematosus successfully treated with rituximab. QJM 2016; 109:617–618.
45. Blanco Pérez JJ, Pérez González A, Guerra Vales JL, *et al.* Shrinking lung in primary Sjogrën syndrome successfully treated with rituximab. Arch Bronconeumol 2015; 51:475–476.
46. Langenskiöld E, Bonetti A, Fitting JW, *et al.* Shrinking lung syndrome successfully treated with rituximab and cyclophosphamide. Respiration 2012; 84:144–149.
47. Schreiber BE, Connolly MJ, Coghlan JG. Pulmonary hypertension in systemic lupus erythematosus. Best Pract Res Clin Rheumatol 2013; 27:425–434.
48. Galiè N, Humbert M, Vachiery J-L, *et al.* 2015 ESC/ERS Guidelines for the diagnosis and treatment of pulmonary hypertension. Eur Heart J 2016; 37:67–119.
49. Simonneau G, Gatzoulis MA, Adatia I, *et al.* Updated clinical classification of pulmonary hypertension. J Am Coll Cardiol 2013; 62(25 Suppl):D34–D41.

50. Hassoun PM, Mouthon L, Barbera JA, *et al.* Inflammation, growth factors, and pulmonary vascular remodeling. J Am Coll Cardiol 2009; 54(1 Suppl): S10–S19.

51. Giaid A, Yanagisawa M, Langleben D, *et al.* Expression of endothelin-1 in the lungs of patients with pulmonary hypertension. N Engl J Med 1993; 328:1732–1739.

52. Rabinovitch M. Molecular pathogenesis of pulmonary arterial hypertension. J Clin Invest 2012; 122:4306–4313.

53. McLaughlin VV, McGoon MD. Pulmonary arterial hypertension. Circulation 2006; 114:1417–1431.

54. Elalouf O, Fireman E, Levartovsky D, *et al.* Decreased diffusion capacity on lung function testing in asymptomatic patients with systemic lupus erythematosus does not predict future lung disease. Lupus 2015; 24:973–979.

55. Hachulla E, Jais X, Cinquetti G, *et al.* Pulmonary arterial hypertension ■■ associated with systemic lupus erythematosus: results from the French Pulmonary Hypertension Registry. Chest 2018; 153:143–151.

The European Registry reports overall 5-year survival rate in SLE-associated pulmonary hypertension patients along with risk factors and protective factors.

56. Thakkar V, Lau EM. Connective tissue disease-related pulmonary arterial hypertension. Best Pract Res Clin Rheumatol 2016; 30:22–38.

57. Castillo-Martıez D, Marroquı-Fabia E, Lozada-Navarro A, *et al.* Levels of uric acid may predict the future development of pulmonary hypertension in systemic lupus erythematosus: a seven-year follow-up study. Lupus 2016; 25:61–66.

58. Hübbe-Tena C, Gallegos-Nava S, Márquez-Velasco R, *et al.* Pulmonary hypertension in systemic lupus erythematosus: echocardiography-based definitions predict 6-year survival. Rheumatology (Oxford) 2014; 53: 1256–1263.

59. Kusunose K, Yamada H, Hotchi J, *et al.* Prediction of future overt pulmonary hypertension by 6-min walk stress echocardiography in patients with connective tissue disease. J Am Coll Cardiol 2015; 66:376–384.

60. Kommireddy S, Bhyravavajhala S, Kurimeti K, *et al.* Pulmonary arterial hypertension in systemic lupus erythematosus may benefit by addition of immunosuppression to vasodilator therapy: an observational study. Rheumatology (Oxford) 2015; 54:1673–1679.

61. Qian J, Wang Y, Huang C, *et al.* Survival and prognostic factors of systemic lupus erythematosus-associated pulmonary arterial hypertension: a PRISMA-compliant systematic review and meta-analysis. Autoimmun Rev 2016; 15:250–257.

62. Sanchez O, Sitbon O, Jais X, *et al.* Immunosuppressive therapy in connective tissue diseases-associated pulmonary arterial hypertension. Chest 2006; 130:182–189.

63. Bazan IS, Mensah KA, Rudkovskaia AA, *et al.* Pulmonary arterial hypertension ■■ in the setting of scleroderma is different than in the setting of lupus: a review. Respir Med 2018; 134:42–46.

The review highlights the emerging evidence that suggests how SLE-associated pulmonary hypertension differs from scleroderma-associated pulmonary hypertension in pathogenesis, clinical course, prognosis and treatment response.

64. Yusuf HR, Hooper WC, Beckman MG, *et al.* Risk of venous thromboembolism among hospitalizations of adults with selected autoimmune diseases. J Thromb Thrombolysis 2014; 38:306–313.

65. Ahlehoff O, Wu JJ, Raunso J, *et al.* Cutaneous lupus erythematosus and the risk of deep venous thrombosis and pulmonary embolism: a Danish nationwide cohort study. Lupus 2017; 26:1435–1439.

66. Mao S, Shen H, Zhang J. Systemic lupus erythematosus and malignancies risk. J Cancer Res Clin Oncol 2016; 142:253–262.

67. Greenwood KM, Lederman L, Lindner HD. Self-reported sleep in systemic lupus erythematosus. Clin Rheumatol 2008; 27:1147–1151.

68. Valencia-Flores M, Resendiz M, Castano VA, *et al.* Objective and subjective sleep disturbances in patients with systemic lupus erythematosus. Arthritis Rheum 1999; 42:2189–2193.

69. Da Costa D, Bernatsky S, Dritsa M, *et al.* Determinants of sleep quality in women with systemic lupus erythematosus. Arthritis Rheum 2005; 53: 272–278.

70. Vina ER, Green SL, Trivedi T, *et al.* Correlates of sleep abnormalities in systemic lupus: a cross-sectional survey in an urban, academic center. J Clin Rheumatol 2013; 19:7–13.

71. Palagini L, Tani C, Mauri M, *et al.* Sleep disorders and systemic lupus erythematosus. Lupus 2014; 23:115–123.

72. McKinley PS, Ouellette SC, Winkel GH. The contributions of disease activity, sleep patterns, and depression to fatigue in systemic lupus erythematosus. A proposed model. Arthritis Rheum 1995; 38:826–834.

73. Sangle SR, Tench CM, D'Cruz DP. Autoimmune rheumatic disease and sleep: a review. Curr Opin Pulm Med 2015; 21:553–556.

74. McCarty DE, Chesson ALJ, Jain SK, Marino AA. The link between vitamin D metabolism and sleep medicine. Sleep Med Rev 2014; 18:311–319.

75. Qin B, Wang J, Yang Z, *et al.* Epidemiology of primary Sjogren's syndrome: a systematic review and meta-analysis. Ann Rheum Dis 2015; 74: 1983–1989.

76. Mavragani CP, Moutsopoulos HM. The geoepidemiology of Sjogren's syndrome. Autoimmun Rev 2010; 9:A305–A310.

77. Shiboski SC, Shiboski CH, Criswell LA, *et al.* American College of Rheumatology classification criteria for Sjogren's syndrome: a data-driven, expert consensus approach in the Sjogren's International Collaborative Clinical Alliance cohort. Arthritis Care Res 2012; 64:475–487.

78. Kokosi M, Riemer EC, Highland KB. Pulmonary involvement in Sjogren syndrome. Clin Chest Med 2014; 31:489–500.

79. Reina D, Roig Vilaseca D, Torrente-Segarra V, *et al.* Sjögren's syndrome-associated interstitial lung disease: a multicenter study. Reumatol Clin 2016; 12:201–205.

80. Ramos-Casals M, Brito-Zerón P, Seror R, *et al.* Characterization of systemic disease in primary Sjögren's syndrome: EULAR-SS Task Force recommendations for articular, cutaneous, pulmonary and renal involvements. Rheumatology (Oxford) 2015; 54:2230–2238.

81. Flament T, Bigot A, Chaigne B, *et al.* Pulmonary manifestations of Sjögren's ■■ syndrome. Eur Respir Rev 2016; 25:110–123.

An excellent review on the epidemiology, presentations, diagnosis and treatment of the multiple pulmonary manifestations of Sjögren syndrome patients. It divides each complication anatomically, provides histopathologic information including images and images of different lung modalities for the diagnosis of pulmonary complications. Serves to compare European data Twitch data from the United States.

82. Ito I, Nagai S, Kitaichi M, *et al.* Pulmonary manifestations of primary Sjogren's syndrome: a clinical, radiologic, and pathologic study. Am J Respir Crit Care Med 2005; 171:632–638.

83. Olsson KM, Palazzini M. Challenges in pulmonary hypertension: managing the unexpected. Eur Respir Rev 2015; 24:674–681.

84. Aerni MR, Vassallo R, Myers JL, *et al.* Follicular bronchiolitis in surgical lung biopsies: clinical implications in 12 patients. Respir Med 2008; 102: 307–312.

85. Nakanishi M, Fukuoka J, Tanaka T, *et al.* Small airway disease associated with Sjogren's syndrome: clinico-pathological correlations. Respir Med 2011; 105:1931–1938.

86. Borie R, Schneider S, Debray M-P, *et al.* Severe chronic bronchiolitis as the presenting feature of primary Sjogren's syndrome. Respir Med 2011; 105:130–136.

87. Soto-Cardenas M-J, Perez-De-Lis M, Bove A, *et al.* Bronchiectasis in primary Sjogren's syndrome: prevalence and clinical significance. Clin Exp Rheumatol 2010; 28:647–653.

88. Shen T-C, Wu B-R, Chen H-J, *et al.* Risk of chronic obstructive ■ pulmonary disease in female adults with primary Sjögren syndrome. Medicine (Baltimore) 2016; 95:e3066.

No large-scale population-based cohort study has ever investigated the risk of developing chronic obstructive pulmonary disease (COPD) in patients with Sjögren syndrome. This article evaluates the risk of COPD in women with primary Sjögren's syndrome (pSS) in a nationwide population.

89. Parambil JG, Myers JL, Lindell RM, *et al.* Interstitial lung disease in primary Sjogren syndrome. Chest 2006; 130:1489–1495.

90. Roca F, Dominique S, Schmidt J, *et al.* Interstitial lung disease in primary Sjögren's syndrome. Autoimmun Rev 2017; 16:48–54.

91. Sebastian A, Misterska-Skóra M, Silicki J, *et al.* Chest HRCT findings in patients with primary Sjögren's syndrome. Adv Clin Exp Med 2017; 26:1101–1106.

92. Palm O, Ueland T, Garen T, *et al.* Endostatin is higher and associated with ■ pulmonary involvement in primary Sjogren's syndrome. Clin Exp Rheumatol 2016; 34:690–693.

The very interesting study compares levels of endostatin in patients from the Norwegian systemic CTD and vasculitis registry versus healthy controls. Those with pSS had higher levels of endostatin when compared with controls and those with pulmonary involvement had higher levels when compared with those without. Endostatin may be a potential serum marker in the future.

93. Kadota J, Kusano S, Kawakami K, *et al.* Usual interstitial pneumonia associated with primary Sjogren's syndrome. Chest 1995; 108: 1756–1758.

94. Gupta N, Wikenheiser-Brokamp KA, Fischer A, McCormack FX. Diffuse cystic lung disease as the presenting manifestation of Sjögren syndrome. Ann Am Thorac Soc 2016; 13:371–375.

95. Kokosi M, Riemer EC, Highland KB. Pulmonary involvement in Sjögren syndrome. Clin Chest Med 2010; 31:489–500.

96. Honda O, Johkoh T, Ichikado K, *et al.* Differential diagnosis of lymphocytic interstitial pneumonia and malignant lymphoma on high-resolution CT. AJR Am J Roentgenol 1999; 173:71–74.

97. Thompson G, Mclean-Tooke A, Wrobel J, *et al.* Sjögren syndrome with associated lymphocytic interstitial pneumonia successfully treated with tacrolimus and abatacept as an alternative to rituximab. Chest 2018; 153:756–759.

98. Suda T, Kaida Y, Nakamura Y, *et al.* Acute exacerbation of interstitial pneumonia associated with collagen vascular diseases. Respir Med 2009; 103:846–853.

99. Sung YK, Chung L. Connective tissue disease-associated pulmonary arterial hypertension. Rheum Dis Clin North Am 2015; 41:295–313.

100. Zhao J, Wang Q, Liu Y, *et al.* Clinical characteristics and survival of pulmonary ■ arterial hypertension associated with three major connective tissue diseases: a cohort study in China. Int J Cardiol 2017; 236:432–437.

The study investigated the clinical characteristics and survival of CTD-pulmonary arterial hypertension in Chinese patients providing insight in some of the characteristics and differences in Asian patients when compared with other ethnic groups. The mortality rates reported and similar to those in the current literature.

101. Humbert M, Barst RJ, Robbins IM, *et al.* Combination of bosentan with epoprostenol in pulmonary arterial hypertension: BREATHE-2. Eur Respir J 2004; 24:353–359.
102. Voulgarelis M, Dafni UG, Isenberg DA, Moutsopoulos HM. Malignant lymphoma in primary Sjogren's syndrome: a multicenter, retrospective, clinical study by the European Concerted Action on Sjogren's syndrome. Arthritis Rheum 1999; 42:1765–1772.
103. Yachoui R, Leon C, Sitwala K, Kreidy M. Pulmonary MALT lymphoma in patients with Sjogren's syndrome. Clin Med Res 2017; 15:6–12.
104. Ramos-Casals M, Brito-Zeron P, Garcia-Carrasco M, Font J. Sarcoidosis or Sjogren syndrome? Clues to defining mimicry or coexistence in 59 cases. Medicine (Baltimore) 2004; 83:85–95.
105. Ramos-Casals M, Font J, Garcia-Carrasco M, *et al.* Primary Sjogren syndrome: hematologic patterns of disease expression. Medicine (Baltimore) 2002; 81:281–292.
106. Mansour MJ, He C, Al-Farra ST, *et al.* Sarcoidosis and Sjogren's syndrome: clinical and salivary evaluation. J Oral Pathol Med 2013; 42:594–599.
107. ■■ Hackett KL, Gotts ZM, Ellis J, *et al.* An investigation into the prevalence of sleep disturbances in primary Sjögren's syndrome: a systematic review of the literature. Rheumatology (Oxford) 2017; 56:570–580.
The sytematic review provides insight on some of the different subjective and objective differences in Sjogren patients when compared with the general population. This study also suggests that further research in sleep medicine and connective tissue disorders is needed.

Lupus and vaccinations

Alexis Mathian[a,b], *Micheline Pha*[a], *and Zahir Amoura*[a,b]

Purpose of review
To review the latest data in the field of vaccinations in systemic lupus erythematosus (SLE), focusing on pneumococcal, seasonal influenza, herpes zoster and human papillomavirus infections.

Recent findings
Less than 40% of patients responded adequately to the 23-valent pneumococcal polysaccharide (PPS23) vaccine. A randomized controlled trial showed that sequential administration of the pneumococcal conjugate vaccine followed by the PPS23 vaccine was well tolerated but was not superior to the PPS23 vaccine alone in terms of immunogenicity. A real-life observation confirmed that annual influenza vaccination has an impact on morbidity and mortality in SLE. Three meta-analyses did not show any particular adverse effect of influenza vaccines in SLE. These vaccines are less immunogenic in SLE. A study confirmed that the quadrivalent human papillomavirus vaccine was well tolerated and highly immunogenic in SLE.

Summary
New data confirm the safety and the lower immunogenicity of pneumococcal and influenza vaccine in SLE patients. Current efforts to improve immunization coverage in SLE should focus on spreading to patients and physicians information on the safety, efficacy and usefulness of vaccines in this population.

Keywords
herpes zoster, human papillomavirus, influenza, pneumococcal, systemic lupus erythematosus, vaccination

INTRODUCTION

Infections remain one of the leading causes of mortality, of excess mortality and of morbidity in systemic lupus erythematosus (SLE) [1,2]. The risk of infection in SLE is the consequence of the immunosuppressive effects of SLE treatments and of severe organ involvement such as lupus nephritis. Lupus itself may also be an independent factor of susceptibility to infection.

Vaccines are an important tool for preventing the risk of infections. However, and despite the lack of scientific evidence, their use in SLE have often been considered problematic because of the theoretical risk of triggering a lupus flare. Infectious risk in SLE has also often been underestimated by the physicians. In recent years, the attitude of the physicians has evolved and vaccines are now commonly proposed in the care of SLE patients. Recommendations and guidelines for vaccinations in adult patients with SLE have been published [3,4]. Experts recommend that lupus patient should receive vaccinations:

(1) accordingly to the recommendations and the schedules for the general population.
(2) and against pneumococcus and annually against seasonal influenza, regardless of treatment received and of visceral involvement.

Live attenuated vaccines are usually contraindicated for individuals with immunocompromising because of a risk that the vaccine could cause disease. In general, nonlive vaccines appear to be well tolerated and do not increase the activity of SLE but are less immunogenic than in the healthy population, especially in patients treated with immunosuppressants [3,4]. Yet, there is still very little data

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KEY POINTS

- Infections remain one of the leading causes of mortality, of excess mortality and of morbidity in SLE.
- SLE patients should receive vaccinations accordingly to the recommendations and schedules for the general population.
- SLE patients should be vaccinated against pneumococcus and annually against seasonal influenza (with the inactivated vaccine).
- The immunogenicity of vaccines in SLE decreases significantly for the pneumococcal and the flu vaccines.
- The factors decreasing immunogenicity of vaccines in SLE are an active SLE disease and/or treatment with an immunosuppressant and/or lymphopenia.
- The risk of a flare of SLE after vaccination has never been confirmed.

on vaccinations and the epidemiology of vaccine-preventable infections in SLE.

The aim of this work was to review the latest data in the field of vaccinations in SLE, focusing on pneumococcus, seasonal influenza, herpes zoster and human papillomavirus (HPV) infections and vaccines (Table 1).

PNEUMOCOCCAL VACCINE IN SYSTEMIC LUPUS ERYTHEMATOSUS

Pulmonary infections are an important cause of morbidity and mortality in lupus, with *Streptococcus pneumoniae* being the most frequent pathogen involved. Of the 3658 SLE patients recorded in the Spanish Rheumatology Society Lupus Registry, 705 (19.3%) suffered from at least one severe infection [2]. Respiratory infections were the most common (35.5%) infections [2].

The frequency and the risk factors favoring the occurrence and severity of pneumococcal infections in SLE are not precisely known. Schurder *et al.* [12] performed a retrospective study of all SLE patients admitted in their department from January 2005 to December 2014. Among a total of 2112.8 patient-years, the incidence of invasive pneumococcal infections was of 236/100 000 patient-years. Risk factors associated with pneumococcal infections in SLE were a serum gammaglobulin level below 5 g/l and a past history of lupus nephritis. Uses of steroids and of immunosuppressive drugs were associated with infection severity [12]. Pneumococcal infections in SLE were more severe, more often invasive and required a higher need for ICU admission [12]. Other risk factors of pneumococcal infections in SLE are a low complement activity, alterations of phagocytic cells and autosplenectomy. Autosplenectomy seems not frequent in

Table 1. List of the latest vaccination studies in systemic lupus erythematosus

Reference	Type of vaccine	Number of SLE patients	Control group	Safety concern	Immunogenicity[a]	Factors decreasing immunogenicity
Rezende *et al.* [5]	PPS23	54 adults	None	None	29.6–38.8%	None
Alyasin *et al.* [6]	PPS23	30 children	30 asthmatic children	None	77.8% for SLE children 86.2% for asthmatic children	Age and age of the disease onset SLEDAI > 8
Nagel *et al.* [7]	PnCj13	47 adults (11 treated with BMB)	21 healthy volunteers	None	SLE patients have a lower immunogenicity compared with healthy volunteers	Higher age Azathioprine use
Chatham *et al.* [8]	PPS23	79 adults treated with BMB	None	None	97.6% in the belimumab-concurrent cohort 97.0% in the prebelimumab cohort According to 'clinically relevant' immunogenicity: 63.4% in the belimumab-concurrent cohort 75.8% in the prebelimumab cohort	None
Grabar *et al.* [9▪]	PnCj7 PPS23	17 adults PnCj7/PPS23	25 adults Placebo/PPS23	None	76% in the PnCj7–PPS23 cohort 72% in the placebo-PPS23 cohort	None
Sacre *et al.* [10▪]	PnCj13 PPS23	21 adults PnCj13/PPS23	None	None	81.0% after 2-month survey 57.1% after 1-year survey	IS, lymphocyte <1000/μl B cell lymphopenia gammaglobulin level 5 g/l
Dhar *et al.* [11]	4vHPV	34 adults	None	None	100%	None

4vHPV, quadrivalent HPV vaccine; BMB, belimumab; HPV, human papillomavirus; IS, immunosuppressant; PnCj13, 13-valent pneumococcal conjugate vaccine; PnCj7, 7-valent pneumococcal conjugate vaccine; PPS23, 23-valent pneumococcal polysaccharide vaccine; SLE, systemic lupus erythematosus.
[a]According to the primary endpoint chosen by the authors.

SLE: in a cohort of 205 SLE patients, only two out of the 12 patients with persistent thrombocytosis had an autosplenectomy [13].

Rezende *et al.* [5] evaluated the immunogenicity of the 23-valent pneumococcal polysaccharide (PPS23) vaccine in a prospective open-label study including 54 adult SLE. IgG antibody levels against seven pneumococcal serotypes were measured by ELISA. The primary endpoint was the percentage of patients who responded appropriately to at least 70% of the tested serotypes. Less than 40% of the patients responded adequately to vaccination, being numerically lower among patients under immunosuppressive treatment [5]. The conclusions of the authors were that the PPS23 vaccine was well tolerated but insufficiently immunogenic in adult SLE.

Alyasin *et al.* [6] studied specific antibody in response to the PPS23 vaccine in SLE children. 78% of SLE children had an adequate immune response defined by at least a two-fold increase in antipneumococcal antibody titer following immunization [6]. The level of postimmunization antipneumococcal antibody significantly correlated with the age of children and their age of disease onset [6]. The mean fold of increase in level of antipneumococcal antibody in patients with a Systemic Lupus Erythematosus Disease Activity Index (SLEDAI) 8 or less was significantly higher than patients with SLEDAI more than 8 [6].

Nagel *et al.* [7] studied a cohort of 47 SLE patients and 21 healthy controls immunized with a single dose of the 13-valent pneumococcal conjugate (PnCj13) vaccine. Compared with controls, SLE patients had lower postvaccination antibody levels and lower fold increase of antibody levels after vaccination. Higher age was associated with lower postvaccination antibody in SLE patients but not in controls [7]. SLE patients on azathioprine had significantly lower fold increase in antibody levels after vaccination [7].

Belimumab given in addition to traditional disease-modifying antirheumatic drugs or prednisolone did not impair antibody response to the 23-valent nonconjugated vaccine [8] and the 13-valent conjugated pneumococcal vaccine [7].

Knowing the poor immunogenicity of the polysaccharide vaccine, new vaccination strategies have been proposed to patients with SLE, especially in case of drug immunosuppression and/or severe organ involvement. The primed-boost vaccination strategy, that is a one-dose regimen of the 7-valent or the 13-valent conjugate vaccine followed several weeks later by a dose of 23-valent nonconjugated vaccine improves vaccine immunogenicity in HIV-infected patients. This strategy was tested in SLE with stable disease in the VACCILUP multicenter randomized placebo-controlled double-blind trial [9▪]. The primary endpoint of this study was the proportion at week 28 of responders to at least five of the seven tested pneumococcal serotypes (4, 6B, 9V, 14, 18C, 19F and 23F) shared by both PPS and 7-valent pneumococcal conjugate (PnCj7). Sequential administration of the PnCj7 vaccine followed 6 months after by the PPS23 vaccine was well tolerated and showed short-term immunological efficacy in 76% of the patient but was not superior to the PPS23 vaccine alone. There were no differences between the rates of responders in patients (regardless of their vaccine schedule) treated with and without immunosuppressants and in those receiving 10 mg or less and more than 10 mg of daily prednisone. Compared with the PPS23 vaccine alone, the primed-boosted strategy did not confer a longer duration of immune protection: after 1-year survey, the rates of SLE patients still completing the primary endpoint decreased to 52% in the placebo-PPS23 group and 59% in the PnCj7 group [9▪]. The poor immunogenicity conferred by the primed-boost strategy was confirmed in an observational study on the durability of the efficiency of the pneumococcal 13-valent conjugate/23-valent polysaccharide vaccine [10▪]. The primary endpoint was the proportion of responders to at least five of seven pneumococcal serotypes (4, 6B, 9V, 14, 18C, 19F and 23F). After 1 year, nine of the 21 vaccinated patients (43%) had no immune protection following pneumococcal vaccination with seroconversion that never ($n = 4$) or only transiently ($n = 5$) occurred. Factors associated with failure to reach immune protection were exposure to immunosuppressant, a lymphocyte count less than 1000/μl, a B-cell lymphopenia notably in the naive and transitional subsets and a gammaglobulin level less than 5 g/l [10▪].

SEASONAL INFLUENZA VACCINE IN SYSTEMIC LUPUS ERYTHEMATOSUS

Influenza virus is often considered a causative agent preceding pulmonary bacterial superinfections. These data argue for recommending flu vaccination to all lupus patients [3] even if the frequency of influenza infection and the factors favoring this infection are not known in SLE.

A real-life cohort confirmed that annual influenza vaccination has an impact on morbidity and mortality in SLE [14▪]. Chang *et al.* used the National Health Insurance Research Database of Taiwan which covers more than 98% of the Taiwanese population and records inpatient and ambulatory care claims. Compared with the nonvaccine cohort, the vaccine cohort had a lower hospitalization rate [hazard ratio (HR) 0.82 (95% confidence interval

(CI) 0.73–0.92)], was less likely to be admitted to the ICU [HR 0.55 (95% CI 0.39–0.79)], to be hospitalized for septicemia, bacteremia or viremia [HR 0.48 (95% CI 0.32–0.73)] and were less predisposed to death [HR 0.41 (95% CI 0.27–0.61)] [14▪].

Vaccine strategies studied in SLE are very heterogeneous: mono or bivalent vaccine against pandemic H1N1 1976 and 2009, seasonal trivalent vaccine and few one with new adjuvants [4]. The studies comparing SLE with healthy controls have recently been reviewed in meta-analyses [15–17]. No study has shown a warning about an increase in SLE activity following influenza vaccines [4,15–17]. The results of the immunogenicity were discordant according to the meta-analysis and the viral strains, especially for H3N2 and B strains. Yet, influenza vaccines are significantly less immunogenic in lupus compared with healthy controls [15–17]. The factors identified for poor influenza vaccine response were corticosteroids and immunosuppressants [15,17]. There is no significant difference in immunogenicity rates between SLE patients without medications and healthy controls [15]. Hydroxychloroquine did not decrease significantly the flu vaccine immunogenicity [15]. As a result, new vaccination strategies should be proposed to patients with SLE. Two studies have shown that a vaccination protocol with two vaccine doses improves the immunogenicity of the vaccine [18,19]. However, since that time, no studies exploring new vaccine strategies have complemented the research on influenza vaccination in SLE.

HERPES ZOSTER IN SYSTEMIC LUPUS ERYTHEMATOSUS

Herpes zoster is reported as one of the most prevalent viral infections in SLE patients. In a meta-analysis, Kawai and Yawn [20] showed that SLE was associated with an elevated risk of herpes zoster with a pooled relative risk of 2.10; 95% CI, 1.40–3.15. Herpes zoster is a rising cause of hospitalizations in SLE [21]. Chen *et al.* [22] showed that the risk of varicella zoster virus (VZV) reactivation as shingles clusters within 3–6 months following SLE diagnosis and reduced thereafter. 67% of the patients experienced herpes zoster during this period [22]. Chen *et al.* [22] showed in a stepwise multivariate logistic regression analysis that lymphopenia below 1.0×10^9/l and corticosteroids at least 30-mg prednisone or equivalent per day were associated with increased herpes zoster risk.

Hu *et al.* showed in a nationwide population-based case–control study that the medications prescribed within the last 3-month period and associated with greater herpes zoster risk in patients with SLE included oral corticosteroids, intravenous methylprednisolone but also hydroxychloroquine, oral cyclophosphamide, intravenous cyclophosphamide, azathioprine, methotrexate and mycophenolate mofetil [23]. For oral corticosteroids and hydroxychloroquine, the risk of herpes zoster was strongly dependent on the medication dose [23]. Of note, an incidence rate of herpes zoster more than 8%/year has recently been reported in two different successful phase II trials studying anti-IFNα mAbs [24,25]. Therefore, preventing VZV reactivation and its complications may be a major challenge for the new lupus treatment.

Until recently, the only zoster vaccine available was the live attenuated virus vaccine. In 2011, European League Against Rheumatism (EULAR) experts recommended that herpes zoster vaccination may be considered in patients with autoimmune inflammatory rheumatic diseases [3]. EULAR expert also advised to administer herpes zoster vaccine only to patients less severely immunosuppressed and who are seropositive for VZV antibodies to prevent primary varicella infection with the vaccine strain [3]. As this recommendation has been made, only one pilot study on vaccination against shingles has been published in SLE [26]. A new vaccine composed of the recombinant VZV glycoprotein E antigen associated with the AS01B adjuvant will probably facilitate the use and efficacy of zoster vaccine in SLE especially in immunocompromized patients [27,28]. This vaccine was approved in the United States in 2017.

HUMAN PAPILLOMAVIRUS IN SYSTEMIC LUPUS ERYTHEMATOSUS

Due to the high incidence rate of HPV-induced malignancies in SLE, experts recommend anti-HPV vaccination in the management of lupus patients [3]. First prophylactic HPV vaccines included the bivalent (2vHPV; Cervarix, GSK, Rixensart, Belgium) and the quadrivalent (4vHPV; Gardasil/Silgard, Merck, Kenilworth, New Jersey, USA) vaccines. The global real-world impact of the 4vHPV vaccine has been confirmed in a systematic review over its first decade of use [29]. Maximal reductions of approximately 90% for HPV 6/11/16/18 infection prevalence, approximately 90% for genital warts, approximately 45% for low-grade cytological cervical abnormalities and approximately 85% for high-grade histologically proven cervical abnormalities have been reported [29].

Dhar *et al.* [11] conducted a phase I study on 4vHPV vaccine in 34 SLE women, ages 19–50 years. Patient with active SLE disease (SLEDAI > 2), history of severe SLE disease, deep venous thrombosis, on more than 400 mg/day of hydroxychloroquine, on

more than 15 mg/day of prednisone, or on immunosuppressant were excluded. No patient experienced a lupus flare or a serious adverse event related to vaccine. Seroconversion rate was 100% for those seronegative at baseline [11]. This study confirmed that HPV vaccine is safe, well tolerated and highly immunogenic in SLE as previously shown with the same 4vHPV [30,31] and the 2vHPV [32]. A nonavalent (9vHPV, Gardasil 9; Merck) vaccine has recently been approved in several countries. It could potentially provide broader coverage and prevent 90% of cervical cancer cases [33].

HOW TO IMPROVE VACCINE COVERAGE IN SYSTEMIC LUPUS ERYTHEMATOSUS?

Although patients with SLE are at increased risk for several vaccine preventable infections and despite better acceptance by patients and doctors, vaccination rates remain low in SLE, in particular for the strongly recommended vaccine against pneumococcus and influenza. Krasselt *et al.* [34] evaluated the vaccination rates among German SLE. A total of 68 patients were recruited from outpatient clinic during one of their routine visits. About 80% of the patients were treated with glucocorticoids and more 60% were on immunosuppressant. Based on their vaccination documents, the current immunization rates for pneumococcal and influenza were only 18 and 25%, respectively [34]. Lawson *et al.* [35] reported that among patients treated with immunosuppressant, the most common reason for not receiving an influenza vaccine was lack of doctor recommendation (55%), followed by efficacy or safety concerns (21%), and lack of time or motivation (19%). Reasons for not receiving pneumococcal vaccine were similar [35]. Thus, efforts need to be made to improve immunization coverage.

Harris *et al.* [36] showed that simple interventions can greatly improve the pneumococcal vaccine coverage in eligible pediatric SLE patients. Interventions included a presentation to rheumatology providers, creation of immunization algorithm, previsit planning, placing reminders on clinic forms, stocking the vaccine in clinic, sending reminder e-mails to providers and sending letters to patients. The PnCj13 vaccine rate increased from 5.3 to 63.2% and PPS23 vaccine rate increased from 15.8 to 57.9% [36]. Serre *et al.* [37▪] measured the impact of shifting pneumococcal vaccination responsibility to nurses. This study was run in a day-care hospital unit for clinical care and included 126 patients. Prior to the study, current recommendations for pneumococcal vaccination in patients with autoimmune inflammatory disorders were provided to the nurses. At admission, patients requiring pneumococcal vaccination were identified by nurses. The vaccine candidates were informed by nurses on benefice and risk of pneumococcal vaccination. Following patient's agreement the first injection (PnCj13) was administered and a second injection (PPS23 at 8 weeks) was planned. This program substantially raised the pneumococcal vaccination rate from 17 to 78% of the patients who were candidate for pneumococcal vaccination [37▪].

CONCLUSION

Available data on vaccination in SLE patients are gradually increasing. New data in this field confirm the lower immunogenicity of pneumococcal and influenza vaccine in SLE patients especially in case of drug immunosuppression, high disease activity and lymphopenia. Can be deduced from the existing data that although reduced, the immunogenicity of vaccines persists in patients receiving or not immunosuppressive therapy and the risk of a flare of SLE after vaccination has never been demonstrated in controlled studies. This risk is therefore theoretical, whereas the risk of infection is real. Yet one should keep in mind that the safety data are obtained most often on a small number of patients in studies underpowered with regard to adverse events detection. Therefore, the next studies on vaccination in SLE should include a maximum number of patients in a multicentric design to definitively convince patients and doctors of their safety.

It seems difficult to restore the immunogenicity of a vaccine in a patient with long-lasting lymphocytopenia. Therefore, vaccination during periods when patients are not treated with high dose corticosteroids and immunosuppressive drugs would improve the immunogenicity of the vaccine. The current efforts to improve vaccination in SLE should be simple interventions to spread information of the safety, efficacy and utility of vaccines in SLE amongst patients, physicians and nurses. Having vaccinations available for immediate administration in clinic or at local pharmacies will also decrease the complexity of care for patients.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Lee YH, Choi SJ, Ji JD, *et al.* Overall and cause-specific mortality in systemic lupus erythematosus: an updated meta-analysis. Lupus 2016; 25:727–734.
2. Rua-Figueroa I, Lopez-Longo J, Galindo-Izquierdo M, *et al.* Incidence, associated factors and clinical impact of severe infections in a large, multicentric cohort of patients with systemic lupus erythematosus. Semin Arthritis Rheum 2017; 47:38–45.
3. van Assen S, Agmon-Levin N, Elkayam O, *et al.* EULAR recommendations for vaccination in adult patients with autoimmune inflammatory rheumatic diseases. Ann Rheum Dis 2011; 70:414–422.
4. Mathian A, Arnaud L, Adoue D, *et al.* Prevention of infections in adults and adolescents with systemic lupus erythematosus: guidelines for the clinical practice based on the literature and expert opinion. Rev Med Interne 2016; 37:307–320.
5. Rezende RP, Ribeiro FM, Albuquerque EM, *et al.* Immunogenicity of pneumococcal polysaccharide vaccine in adult systemic lupus erythematosus patients undergoing immunosuppressive treatment. Lupus 2016; 25: 1254–1259.
6. Alyasin S, Adab M, Hosseinpour A, *et al.* Immunogenicity of 23-valent pneumococcal vaccine in children with systemic lupus erythematosus. Iran J Immunol 2016; 13:204–219.
7. Nagel J, Saxne T, Geborek P, *et al.* Treatment with belimumab in systemic lupus erythematosus does not impair antibody response to 13-valent pneumococcal conjugate vaccine. Lupus 2017; 26:1072–1081.
8. Chatham W, Chadha A, Fettiplace J, *et al.* A randomized, open-label study to investigate the effect of belimumab on pneumococcal vaccination in patients with active, autoantibody-positive systemic lupus erythematosus. Lupus 2017; 26:1483–1490.
9. ■ Grabar S, Groh M, Bahuaud M, *et al.* Pneumococcal vaccination in patients with systemic lupus erythematosus: a multicenter placebo-controlled randomized double-blind study. Vaccine 2017; 35:4877–4885.

A controlled randomized study showing that sequential administration of pneumococcal conjugate vaccine followed by polysaccharide vaccine is well tolerated but is not superior to the polysaccharide vaccine alone.

10. ■ Sacre K, Goulenok T, Bahuaud M, *et al.* Impaired long-term immune protection following pneumococcal 13-valent/23-valent polysaccharide vaccine in systemic lupus erythematosus (SLE). Ann Rheum Dis 2018. [Epub ahead of print]

An observational study demonstrating a poor immune protection conferred by the recommended sequential pneumococcal conjugate/polysaccharide vaccines.

11. Dhar JP, Essenmacher L, Dhar R, *et al.* The safety and immunogenicity of quadrivalent HPV (qHPV) vaccine in systemic lupus erythematosus. Vaccine 2017; 35:2642–2646.
12. Schurder J, Goulenok T, Jouenne R, *et al.* Pneumococcal infection in patients with systemic lupus erythematosus. Joint Bone Spine 2018; 85:333–336.
13. Uskudar Cansu D, Uskudar Teke H, Musmul A, *et al.* Is thrombocytosis always an indicator of autosplenectomy in patients with systemic lupus erythematosus? Rheumatol Int 2018; 38:239–247.
14. ■ Chang CC, Chang YS, Chen WS, *et al.* Effects of annual influenza vaccination on morbidity and mortality in patients with systemic lupus erythematosus: a nationwide cohort study. Sci Rep 2016; 6:37817.

Influenza vaccination is associated with a reduced risk of hospitalization, pneumonia, ICU admission and death in systemic lupus erythematosus patients.

15. Huang Y, Wang H, Wan L, *et al.* Is systemic lupus erythematosus associated with a declined immunogenicity and poor safety of influenza vaccination?: a systematic review and meta-analysis. Medicine (Baltimore) 2016; 95:e3637.
16. Puges M, Biscay P, Barnetche T, *et al.* Immunogenicity and impact on disease activity of influenza and pneumococcal vaccines in systemic lupus erythematosus: a systematic literature review and meta-analysis. Rheumatology (Oxford) 2016; 55:1664–1672.
17. Liao Z, Tang H, Xu X, *et al.* Immunogenicity and safety of influenza vaccination in systemic lupus erythematosus patients compared with healthy controls: a meta-analysis. PLoS One 2016; 11:e0147856.
18. Holvast A, van Assen S, de Haan A, *et al.* Effect of a second, booster, influenza vaccination on antibody responses in quiescent systemic lupus erythematosus: an open, prospective, controlled study. Rheumatology (Oxford) 2009; 48:1294–1299.
19. Mathian A, Devilliers H, Krivine A, *et al.* Factors influencing the efficacy of two injections of a pandemic 2009 influenza A (H1N1) nonadjuvanted vaccine in systemic lupus erythematosus. Arthritis Rheum 2011; 63:3502–3511.
20. Kawai K, Yawn BP. Risk factors for herpes zoster: a systematic review and meta-analysis. Mayo Clin Proc 2017; 92:1806–1821.
21. Murray SG, Schmajuk G, Trupin L, *et al.* National lupus hospitalization trends reveal rising rates of herpes zoster and declines in pneumocystis pneumonia. PLoS One 2016; 11:e0144918.
22. Chen D, Li H, Xie J, *et al.* Herpes zoster in patients with systemic lupus erythematosus: clinical features, complications and risk factors. Exp Ther Med 2017; 14:6222–6228.
23. Hu SC, Yen FL, Wang TN, *et al.* Immunosuppressive medication use and risk of herpes zoster (HZ) in patients with systemic lupus erythematosus (SLE): a nationwide case-control study. J Am Acad Dermatol 2016; 75:49–58.
24. Khamashta M, Merrill JT, Werth VP, *et al.* Sifalimumab, an antiinterferon-alpha monoclonal antibody, in moderate to severe systemic lupus erythematosus: a randomised, double-blind, placebo-controlled study. Ann Rheum Dis 2016; 75:1909–1916.
25. Furie R, Khamashta M, Merrill JT, *et al.* Anifrolumab, an antiinterferon-alpha receptor monoclonal antibody, in moderate-to-severe systemic lupus erythematosus. Arthritis Rheumatol 2017; 69:376–386.
26. Guthridge JM, Cogman A, Merrill JT, *et al.* Herpes zoster vaccination in SLE: a pilot study of immunogenicity. J Rheumatol 2013; 40:1875–1880.
27. Lal H, Cunningham AL, Godeaux O, *et al.* Efficacy of an adjuvanted herpes zoster subunit vaccine in older adults. N Engl J Med 2015; 372:2087–2096.
28. Cunningham AL, Lal H, Kovac M, *et al.* Efficacy of the herpes zoster subunit vaccine in adults 70 years of age or older. N Engl J Med 2016; 375:1019–1032.
29. Garland SM, Kjaer SK, Munoz N, *et al.* Impact and effectiveness of the quadrivalent human papillomavirus vaccine: a systematic review of 10 years of real-world experience. Clin Infect Dis 2016; 63:519–527.
30. Mok CC, Ho LY, Fong LS, *et al.* Immunogenicity and safety of a quadrivalent human papillomavirus vaccine in patients with systemic lupus erythematosus: a case–control study. Ann Rheum Dis 2013; 72:659–664.
31. Soybilgic A, Onel KB, Utset T, *et al.* Safety and immunogenicity of the quadrivalent HPV vaccine in female systemic lupus erythematosus patients aged 12 to 26 years. Pediatr Rheumatol Online J 2013; 11:29.
32. Heijstek MW, Scherpenisse M, Groot N, *et al.* Immunogenicity of the bivalent human papillomavirus vaccine in adolescents with juvenile systemic lupus erythematosus or juvenile dermatomyositis. J Rheumatol 2013; 40:1626–1627.
33. Huh WK, Joura EA, Giuliano AR, *et al.* Final efficacy, immunogenicity, and safety analyses of a nine-valent human papillomavirus vaccine in women aged 16–26 years: a randomised, double-blind trial. Lancet 2017; 390: 2143–2159.
34. Krasselt M, Baerwald C, Seifert O. Insufficient vaccination rates in patients with systemic lupus erythematosus in a German outpatient clinic. Z Rheumatol 2017. [Epub ahead of print]
35. Lawson EF, Trupin L, Yelin EH, *et al.* Reasons for failure to receive pneumococcal and influenza vaccinations among immunosuppressed patients with systemic lupus erythematosus. Semin Arthritis Rheum 2015; 44:666–671.
36. Harris JG, Maletta KI, Ren B, *et al.* Improving pneumococcal vaccination in pediatric rheumatology patients. Pediatrics 2015; 136:e681–e686.
37. ■ Serre J, Francois C, der Haegen MCV, *et al.* Nurse-led vaccination program dramatically improves pneumococcal vaccination coverage among patients with autoimmune inflammatory disorders. Eur J Intern Med 2017; 43:e43–e45.

Shifting pneumococcal vaccination responsibility to nurses dramatically increased the vaccine coverage.

An update on the role of type I interferons in systemic lupus erythematosus and Sjögren's syndrome

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Purpose of review
Systemic lupus erythematosus (SLE) and primary Sjögren's syndrome (pSS) share several clinical and laboratory features, including an overexpression of type I interferon (IFN) regulated genes. The genetic background to this IFN signature and the role of the type I IFN system in the disease process have been partly clarified. Here, we summarize the latest information concerning the type I IFN system in both diseases.

Recent findings
A number of gene variants in the type I IFN signalling pathways associate with an increased risk for both SLE and pSS in several ethnicities. The function of some risk gene variants has been elucidated, as well as the importance of epigenetic changes in type I IFN regulated genes. MicroRNA-451 and miR-302d have been shown to target IFN regulatory factor 8 and 9, suggesting that noncoding RNAs can control the IFN system. A prominent type I IFN activation is related to several disease manifestations, and in SLE to a more severe disease phenotype. Phase II studies in SLE suggest beneficial effects of blocking the type I IFN receptor.

Summary
The activated type I IFN system in SLE and pSS has a strong genetic component, is important in the disease etiopathogenesis and can be targeted.

Keywords
genes, Sjögren's syndrome, systemic lupus erythematosus, therapy, type I interferon

INTRODUCTION

The type I interferon (IFN) system is an important part of our defense against viral infections, but is also aberrantly activated in a number of systemic inflammatory autoimmune diseases. Both systemic lupus erythematosus (SLE) and Sjögren's syndrome belong to this group of disorders, and the patients have a prominent overexpression of type I IFN regulated genes, denoted as an IFN signature. Originally, nucleic acid containing immune complexes in the patients were demonstrated to induce IFNα production in plasmacytoid dendritic cells (pDCs) via interaction with Toll-like receptors (TLRs). Subsequently, several other triggers of type I IFN induction were described, such as neutrophil extracellular traps which contain HMGB1, LL37, histones and DNA that all can trigger pDC to produce IFNα, which after binding to the IFN receptor and following downstream signalling generates the IFN signature [1]. The understanding of the role and implications of type I IFNs in patients with SLE and primary Sjögren's syndrome (pSS) is rapidly growing, and in this review, we highlight recent findings, with special attention related to induction of type I IFN, the genetic and epigenetic regulation in the patients, as well as immunological effects of type I IFNs and their relation to organ disease. The emerging therapeutic possibilities in relation to the type I IFNs are also discussed.

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KEY POINTS

- The type I IFN system is activated in a large proportion of patients with SLE and pSS, leading to an IFN signature.
- There is a strong genetic background to the IFN signature, and several genes in the type I IFN signalling pathway have variants associated with a risk for SLE and pSS.
- A number of self-derived inducers of IFN production in pDC are now described in SLE and pSS.
- Produced type I IFN have prominent immunostimulatory effects, which contribute to the autoimmune reaction and clinical disease activity.
- The type I IFN system may be an effective therapeutic target in a proportion of patients with SLE and pSS.

INDUCTION OF TYPE I INTERFERON AND WAYS TO MEASURE IT

Recently, alternative routes of type I IFN induction and new sources of IFN-inducing nucleic acids have been suggested. Levels of circulating U1 and Y1 RNA, which bind the 70K, Sm and Ro60 autoantigens, have been found to be increased in patients with SLE compared with healthy individuals, and their levels correlate with SLE disease activity and *IFN* gene signature [2]. RNA derived from human genome-endogenous viral retroelements, of which increased levels were observed in target tissue biopsies of patients with both SLE and pSS, has been proposed to drive type I IFN expression in these diseases [3]. Mitochondrial antiviral signalling protein (MAVS) amplifies IFN production in response to viral RNA and has now been shown to aggregate in a subset of SLE patients, coinciding with higher levels of serum IFNβ and autoantibodies [4]. Also, serum exosomes are present in higher levels in serum from patients with SLE, their presence correlates with SLE disease activity and SLE exosomes induce more IFNα than those from healthy controls [5]. Thus, there exist a number of different mechanisms by which type I IFN production can be induced in both SLE and pSS.

Given that IFN-related biomarkers appear suitable for following clinical activity of SLE [6], it is essential that robust and easily managed methods for measurement of type I IFNs are developed. Indirect measurements of type I IFN activity through an IFN score, based on standardized expression levels of selected IFN stimulated genes, is currently the most used method for assessing systemic IFN activity in patients, but can be assumed to be less sensitive to change than direct measurements of the IFNs themselves. Although direct quantification of type I IFN has been challenging, some promising progress has been made with an assay relying on high-affinity antibodies isolated from APS1 patients and a digital ELISA technology [7▪]. With this method, it was possible to measure even very low concentrations of IFNα in several different body fluids. If it can be effectively translated to clinical laboratory routine formats, there is potential for use for example patient diagnostic and stratification purposes.

GENETIC VARIATION IN SYSTEMIC LUPUS ERYTHEMATOSUS AND PRIMARY SJÖGREN'S SYNDROME RELATED TO INTERFERON PATHWAYS IN DIFFERENT ETHNIC GROUPS

Systemic autoimmune diseases share several clinical, immunological and genetic features and the pathogenesis in both SLE and pSS have a strong genetic component (Supplementary Table 1, http://links.lww.com/COR/A39). As for most autoimmune diseases, associations to *HLA* explain most of the disease development risk hitherto attributed to genetics, but numerous non-*HLA* loci have also been associated with SLE or pSS, of which many overlap between the diseases (Fig. 1).

Further penetrating the genetic associations in SLE and pSS, several studies that highlight the importance of genetic variation within the type I IFN related pathways for these diseases have recently been published. Using the Immunochip, Langefeld *et al.* [8▪▪] reported a large transancestral association study of SLE, which included more than 27 000 individuals of European, African, Hispanic and Amerindian backgrounds. The results included confirming *IFR5* as the most significant non-MHC locus across the ethnic groups, and defining 24 novel genetic regions associated with SLE, mapping both ancestry-dependent and ancestry-independent contributions. An interesting observation from this study was also the role of genetic load in relation to risk of disease, wherein increasing allele count of risk variants accelerated the pattern of SLE risk, prompting the authors to propose a cumulative hit hypothesis for autoimmune disease. In a Han Chinese population, a large-scale exome-wide study of 5000 patients with SLE identified additional loci that so far appear specific for this population [9]. In a recent world-wide collaborative effort of multiethnic genome-wide association (GWA) analysis for pSS, Taylor *et al.* [10] replicated known genes and identified several novel genetic associations with pSS. Notably, they observed prominent differences in the patterns of association for individuals of different ancestry in their cohort, particularly in the *MHC* region. In a study focused on Han Chinese,

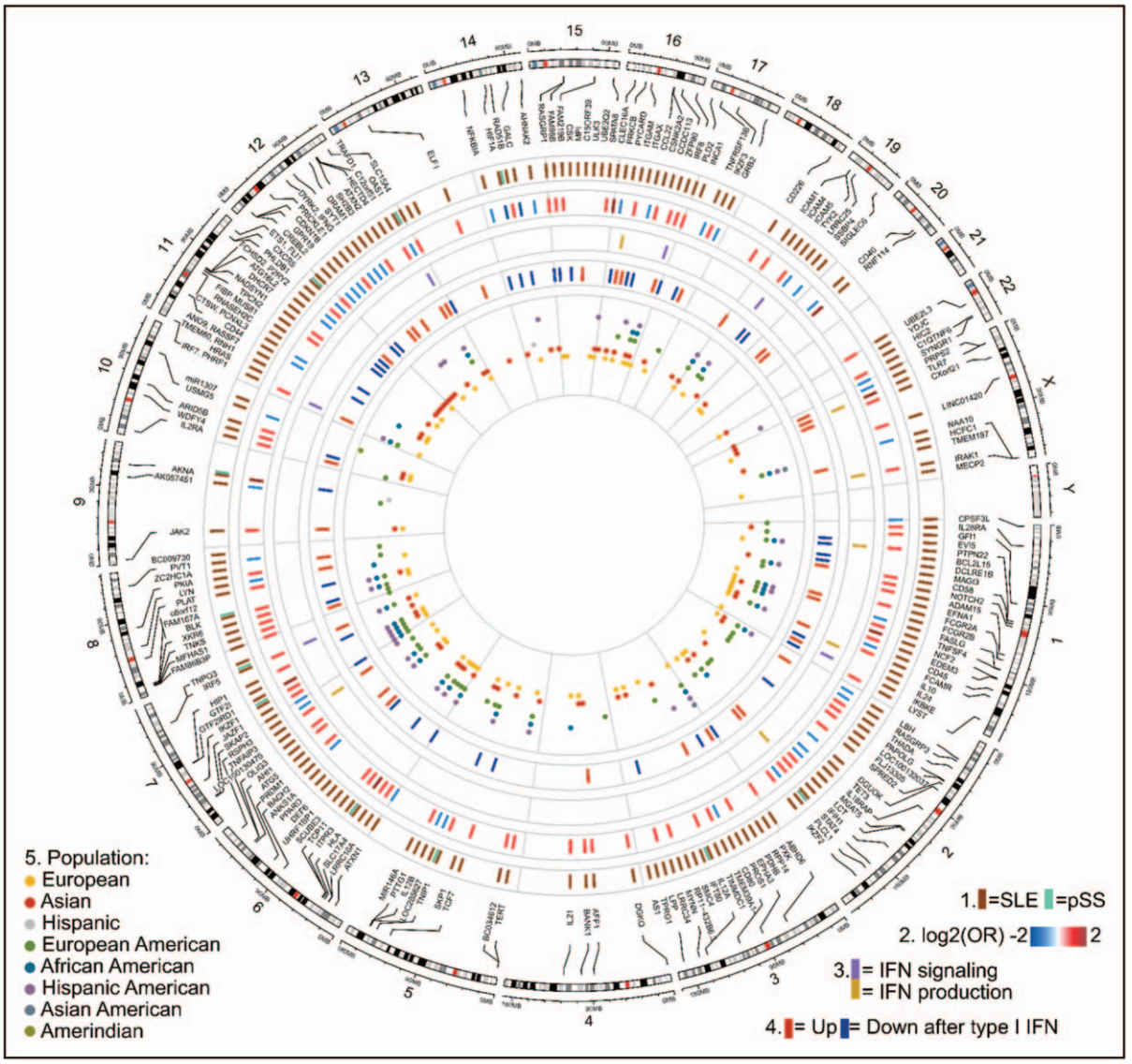

FIGURE 1. Circular representation of genetic loci associated with SLE or pSS. Outermost circle represents chromosomes, with genes near the associated loci labelled. First inner track represents the disease that has been associated (SLE in brown, pSS in green). Second track represents the best published odds ratio (log2 transformed) for allele carriers developing disease. Third track denotes annotation of the selected gene to a type I IFN pathway; yellow for type I IFN production (GO: 0032479), purple for downstream type I IFN signalling (GO: 0034340/0060338/0005132). Fourth track shows up or downregulation post type I IFN treatment (Interferome version 2.01, (http://www.interferome.org). Fifth track represents the associated population.

variants near the *signal transducer and activator of transcription* (*STAT4*) regulator *IKAROS family zinc finger 1* (*IKZF1*) previously associated with several autoimmune diseases including SLE were explored in pSS [11]. The study included a limited number of patients and controls (665 and 863, respectively), the analysis of which did not reach genome-wide significance. By combining the data with that from a previous GWA study, the *IKZF1* locus could be identified at this level of significance also for patients with pSS of Han Chinese origin [11].

Figure 1 details the identified genes and the relation to ethnic groups, their overlap between SLE and pSS, and their regulation by type I IFN. Together, these studies suggest that systemic autoimmune disease can be uniquely dictated by different genetic variance between ethnic groups, which may be an important factor to consider for treatment options.

GENETIC FEATURES OF TYPE I INTERFERON INDUCTION AND TYPE I INTERFERON SIGNALLING AND THEIR FUNCTIONAL IMPLICATIONS

In the pathways related to induction of type I IFN, loci containing toll-like receptors and components of

their downstream signalling have been associated with systemic autoimmunity (Fig. 2). Genetic variation of type I IFN receptors have so far not been associated with SLE or pSS, but molecules of their downstream signalling pathways have repeatedly been implicated in both diseases (Fig. 3). Several recent publications have further added to our understanding of these associations. Variants in *STAT4* associate with a more severe SLE phenotype and risk gene variants of *STAT4* have now been shown to affect the responses of $CD8^+$ and $CD4^+$ T cells to IFNα, with increased levels of phosphorylated, activated STAT4 following exposure to IFNα [12▪]. These observations provide a mechanism for how the increased levels of type I IFN in SLE and pSS patients may interplay with host genetics to drive a vicious

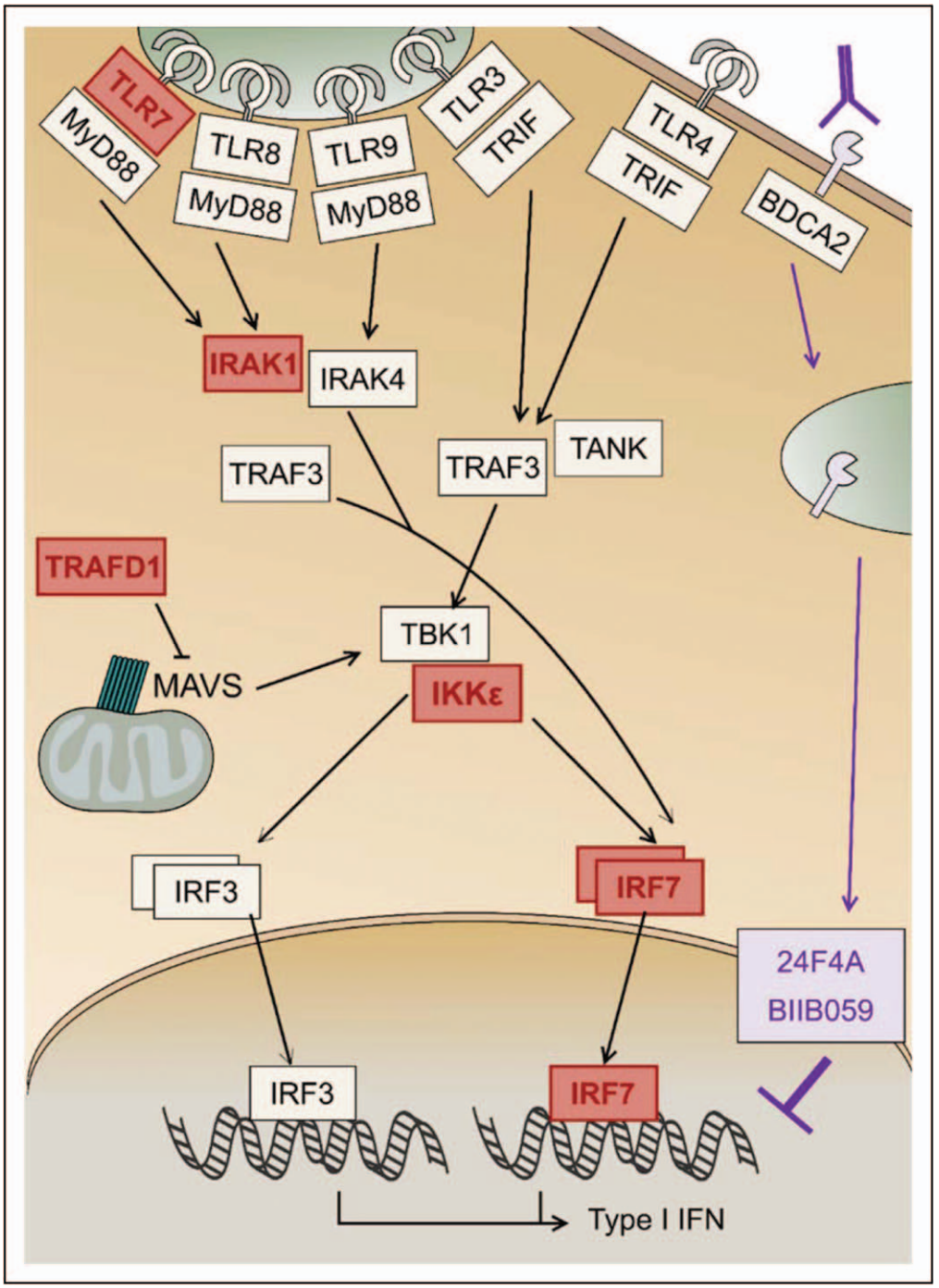

FIGURE 2. Simplified schematic view of genes in the TLR signalling pathway with polymorphisms associated with SLE or pSS. Red boxes represent genes with associated polymorphisms in pSS or SLE. Purple represents treatment effects.

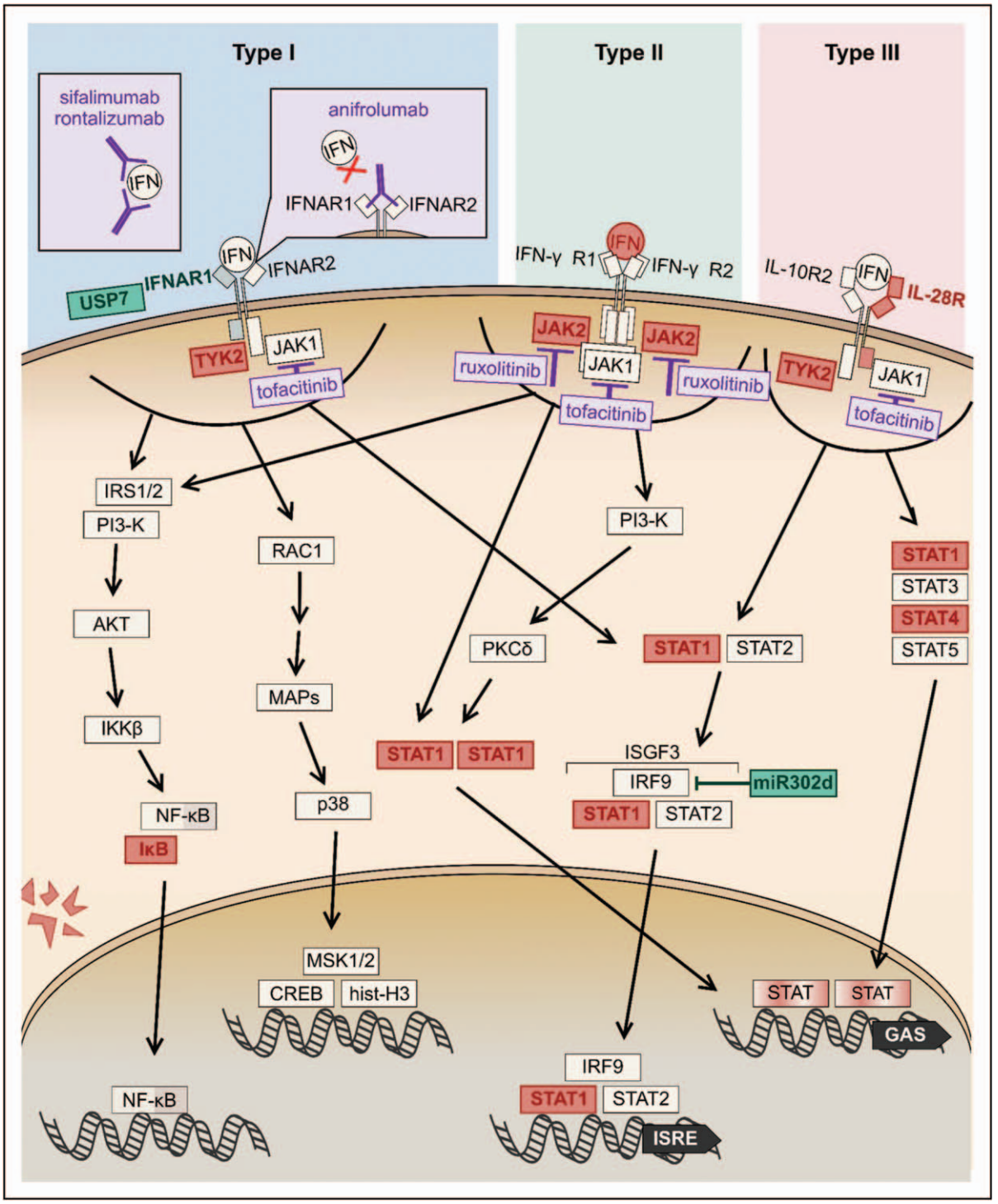

FIGURE 3. Simplified schematic view of genes in the IFN signalling pathways with polymorphisms associated with SLE or pSS. Red boxes represent genes with associated polymorphisms. Green represents newly described functional effects. Purple represents treatment effects.

circle of inflammatory activity. Apart from the STATs, IFN regulatory factors, IRFs, are central transcriptional regulators of IFNs and IFN responses. Interestingly, two microRNAs, miR-451a and miR-302d, have been shown to target *IRF8* and *IRF9*, respectively, in an opposite manner [13,14▪]. In particular, miR-302d was shown to be negatively correlated with IFN activity and to be underexpressed in SLE patients [14▪], demonstrating that also noncoding RNAs play a role in IFN pathway regulation.

Other implicated genes associated with changes in the type I IFN system homeostasis is a novel,

homozygous, loss-of-function mutation in the *complement 1R* gene found in a family afflicted by early-onset SLE, with reduced complement levels and strong type I IFN activation. More affected members of the family had an enrichment of variants commonly associated to SLE, albeit without a distinct *HLA* type involvement [15]. Polymorphisms in the *purine nucleoside phosphorylase* (*PNP*) gene have also been associated with type I IFN activation in SLE [16,17], and nonsynonymous *Integrin Subunit Alpha M* (*ITGAM*) variants known to associate with SLE, encoding defective forms of CD11b, have recently been shown to increase serum type I IFN activity, indicating a role for CD11b targeting as an SLE therapeutic [18▪▪].

GENETIC ASSOCIATIONS AND UNDERLYING MECHANISMS IN DOWNSTREAM EFFECTS OF TYPE I INTERFERONS IN SYSTEMIC LUPUS ERYTHEMATOSUS AND PRIMARY SJÖGREN'S SYNDROME

Several genetic polymorphisms in typical IFN-stimulated genes (ISGs) have recently been pinpointed as associated with SLE or pSS. A polymorphism in *2′-5′-oligoadenylate synthetase 1* (*OAS1*) that reduces its enzymatic activity has been described for pSS [19▪]. *OAS1* is a prototypic type I IFN response gene, and in their article, Li *et al.* [19▪] suggest a mechanism wherein altered IFN responses lead to chronic viral infections, resulting in initiated or exaggerated autoimmune responses. Considering the long-standing hypothesis of viral involvement in the pathogenesis of both pSS and SLE, this and other emerging mechanistic bases for gene-environment interaction to precipitate or drive autoimmunity is a good step forward for our understanding of how these diseases may develop. Further studies on ISGs have identified polymorphisms in the *interferon-induced helicase C domain* (*IFIH1*) as associated with SLE risk, most recently in a Brazilian cohort [20], and these polymorphisms appear to coincide with a reduction in IP-10/CXCL10 [21]. However, increased IP-10 levels have been observed in patients with both pSS and SLE [6], but variations in IP-10 levels appear to play a role, as a reduction in IP-10 is associated with pSS fatigue [22]. This suggests that overall, pathways involving both the production and responses to type I IFN appear to be aberrant in patients with SLE or pSS.

The production of reactive oxygen species (ROS) has historically been considered inflammatory and pathogenic in autoimmune settings. Yet, ROS production was later discovered to have a multitude of context-dependent immunological effects [23,24], including a role for reduced ROS production in the induction of autoimmunity with a type I IFN signature, reviewed by Holmdahl *et al.* [25]. Recent publications have confirmed that polymorphisms in the *neutrophil cytosolic factor 1* (*NCF1*) gene, the gene encoding the neutrophil NADPH oxidase, associate with reduced ROS levels, as well as the development of autoimmune diseases, including SLE and pSS [26▪▪,27▪▪]. In addition, aberrant nitric oxide levels may promote endothelial dysfunction [28], which in turn has been suggested to play a role for cardiovascular disease in SLE [29].

SIGLEC1 is a type I IFN induced protein and Rose *et al.* [30] recently demonstrated an association between expression of SIGLEC1 on peripheral monocytes, IFN activation and disease severity in pSS patients, corroborating earlier observations in SLE of a connection between SIGLEC1 expression and disease activity. An increased SIGLEC1 expression on maternal monocytes has also been associated with a risk of autoimmune congenital heart block in the foetus of mothers with anti Ro/La autoantibodies by way of type I IFN activation [31▪], indicating that it may be a useful biomarker of IFN activity and disease phenotype.

The interplay between pDCs and B cells has emerged as a key node in autoimmune pathogenesis. Interestingly, pDCs can trigger an increase in $CD27^{-}IgD^{-}$ B cells under SLE-like conditions *in vitro* [32]. pDCs are also important drivers of regulatory B cell (B_{reg}) differentiation and the controlled release of IFNα by pDCs pushes naive B cells towards either plasma cell or B_{reg} differentiation, and the disruption of this axis has been suggested to play a role in SLE pathogenesis and to normalize after rituximab treatment [33▪]. pDC density in pSS affected salivary glands correlates with the numbers of infiltrating $CXCL13^{+}$ macrophages, which are induced by type I IFN to upregulate CXCL13 and attract pathogenic $CXCR5^{+}$ B cells to the glands [34], and diminished levels of $CXCR5^{+}$ B cells have been observed in the periphery [35]. Notably, polymorphisms of *CXCR5* have been associated with pSS, and mediate an expression quantitative locus (eQTL) effect with lesser expression of *CXCR5* from disease-associated alleles [35]. This observation led to the proposal that the diminished *CXCR5* expression on peripheral B cells in patients with pSS may relate both the genotype and target organ homing.

The role of autoantigens in autoimmune disease pathogenesis, if any, has puzzled scientists and physicians for decades. Ro/SSA and La/SSB are autoantigens in both SLE and pSS, and while a distinct cellular role for the Ro52/TRIM21 in the ubiquitination of several IRFs has been described [36], the function of Ro60 and La has remained less clear.

However, La was recently shown to protect against viral infections and bind RIG-I directly to drive RIG-I antiviral responses [37]. Patients with pSS who have IFN activation also have upregulated RNA sensing receptors, including RIG-I, as compared with IFN low patients [38]. Furthermore, T_{reg} cells specific for La are expanded by antigen-presenting pDCs in a type I IFN dependent manner [39], showcasing the integral role of pDCs in driving pathogenic IFN responses. Although the role of anti-La autoantibodies in these mechanisms remains to be fully explained, La does play a role in viral and IFN responses and its function in SLE or pSS disease pathogenesis may very well be more significant than previously appreciated.

The recent progress in type I IFN-related immunopathological mechanisms in SLE and pSS is summarized in Fig. 4 [40]. The observations highlight that pathways both upstream and downstream of IFN activation are genetically polymorphic in individuals with both SLE and pSS, and impacts disease pathogenesis and organ dysfunction.

EPIGENETIC REGULATION OF THE TYPE I INTERFERON SYSTEM IN SYSTEMIC LUPUS ERYTHEMATOSUS AND PRIMARY SJÖGREN'S SYNDROME

Looking beyond the effects of single nucleotide variation, epigenetic factors have emerged as potent links to autoimmune pathogenesis. Both SLE and pSS patients have been shown to have prominent hypomethylation of IFN regulated genes and many of the hypomethylated sites are associated with increased expression of IFN-regulated genes. This appears true both for cells of peripheral blood and target organs such as salivary glands [41], and even in long time cultured salivary gland epithelial cells from pSS patients [42], indicating that the effect of type I IFN is penetrant throughout the body and stable for extended periods. Interestingly, in a study of twins discordant for disease, the epigenetic regulation in terms of DNA methylation was also associated with SLE flare [43▪], with a higher degree of hypomethylation of IFN-regulated genes observed in twins that had experienced flares within the past 2 years. These observations also highlight the relative stability of the DNA methylation pattern, and support previous reports of the association between IFN activity and disease activity.

At a mechanistic level, a study of human macrophages has demonstrated cross-talk of TNF and type I IFN at the epigenetic level [44▪▪]. The authors observed an important role for type I IFN in priming chromatin to enable robust transcriptional responses to rather weak upstream TNF signals, increasing the sensitivity of cells to stimuli that would otherwise be deemed harmless. The study reveals a mechanism by which the cytokine signalling crosstalk is integrated at the epigenomic level, and also potentially provide an explanation why disease flare is more easily provoked following periods of infections or low disease activity, compared with periods of complete remissions. In all, it appears that the chronic presence of type I IFN evident in both SLE and pSS can be both the signal and the echo of the genetic involvement seen in these systemic diseases.

RECENT DEVELOPMENT IN THERAPIES TARGETING DIFFERENT LEVELS AND MECHANISMS RELATED TO TYPE I INTERFERON

Following the discovery of increased IFNα levels and downstream activation of ISGs in patients with SLE and pSS, strategies to target type I IFNs were soon developed. The first approaches aimed to block IFNα in SLE using mAbs such as sifalimumab and rontalizumab, or to induce anti-IFNα antibodies by a vaccination strategy. Although these treatments have demonstrated some improvement in SLE patients, the effect is moderate and the high IFN signatures are not completely downregulated [45–47]. Part of the reason for this may be that all IFNα variants are not targeted, and that these therapies do not block other type I IFNs such as β, ω, κ or ε.

All type I IFNs signal through the same receptor, the IFN type I receptor (IFNAR) (Fig. 3), and with the hope to more efficiently block type I IFN activation, an alternative approach has been to develop mAbs with receptor antagonist activity. A randomized, double-blind, placebo-controlled phase IIb study of such an antibody (anifrolumab) notably reported substantially reduced disease activity compared with placebo (including standard therapy) across several clinical endpoints in patients with moderate to severe SLE [48▪]. It was particularly effective in patients with high IFN signature scores, but no differences from placebo were observed in patients with a low IFN gene signature at screening. A dosage-related increase in upper respiratory tract infections and reactivation of herpes zoster were noted in the anifrolumab-treated patients, which perhaps is not surprising given the important role of the type I IFNs in the antiviral defense system. A recent study by Bialas *et al.* [49▪] demonstrated a role for type I IFN in neuronal synapse loss and suggested that treatment with anifrolumab should be evaluated for potential effects on central nervous system involvement in SLE. IFNAR-targeted therapy has not yet been assessed in pSS.

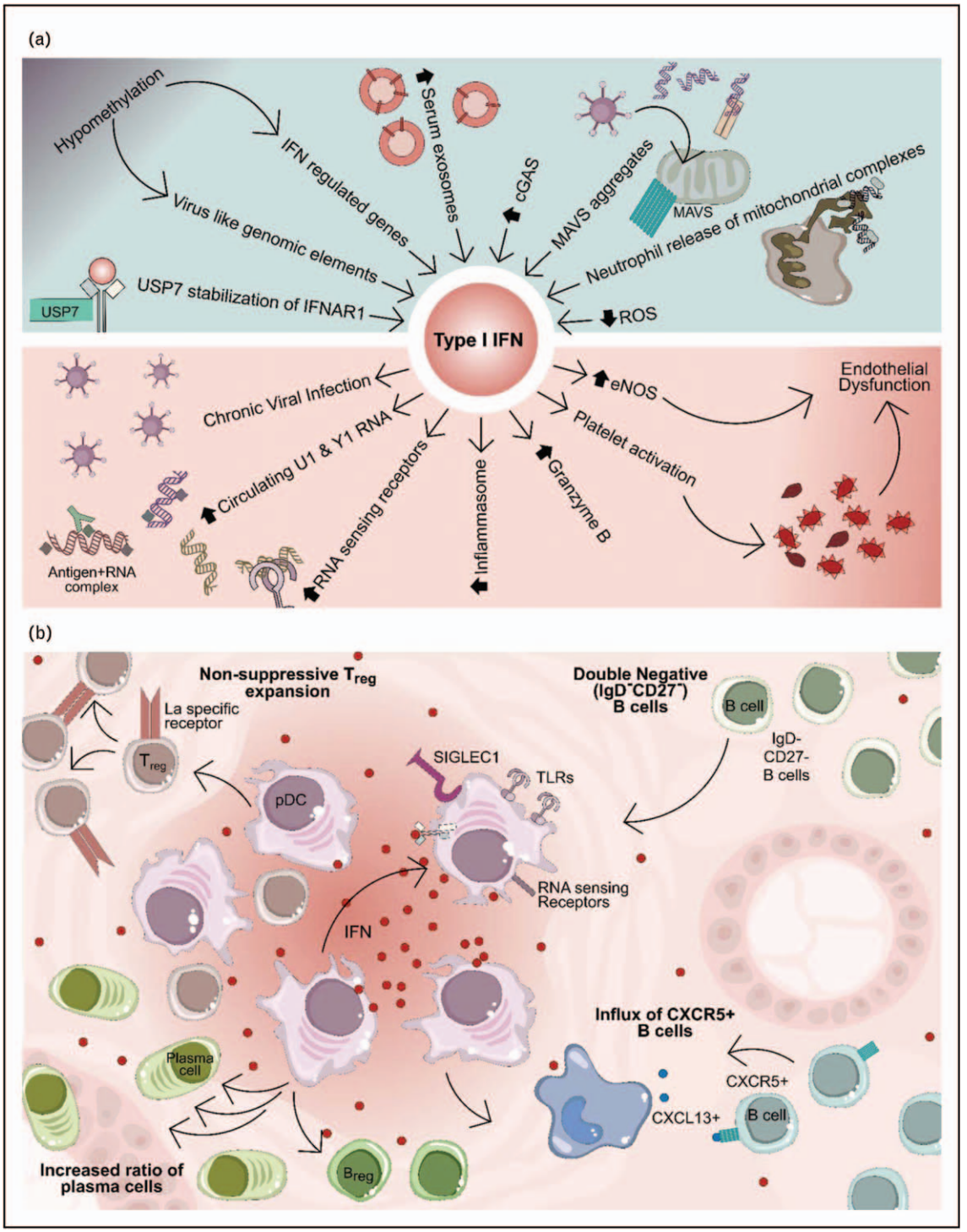

FIGURE 4. Summary of recent progress in type I IFN-related immunopathological mechanisms in SLE and pSS. (a) Type I IFN production and effects. (b) Summary of recently described functional effects of disease pathogenesis and IFN dysregulation in a disease setting, focusing on pDCs. Nonsuppressive T_{regs} are expanded in affected pSS tissues [39], and particularly cells with La specificities [40]. Double negative B cells are recruited to affected tissues by pDCs [32], as well as $CXCR5^{+}$ B cells via CXCL13 upregulation [34]. pDCs can dictate the ratio of B_{regs} vs. plasma cells, pushing the balance towards more antibody-producing plasma cells [33▪].

Therapies with a potential to inhibit signalling downstream of IFNAR are also in clinical trials. Janus kinases (JAKs) bind to and phosphorylate activated type I and II cytokine receptors including IFNAR, which in turn recruits signal transducer and activator of transcription (STATs), which are activated, translocate into the nucleus and regulate gene transcription (Fig. 3). Although not specific for inhibiting type I IFN signalling, several small molecule kinase inhibitors that target JAKs are in clinical trials for lupus and Sjögren's syndrome (www.clinicaltrials.gov), and two (tofacitinib, baracitinib) are approved by the European Medicines Agency (EMA) for treatment of rheumatoid arthritis. Tofacitinib, which targets JAK1 and JAK3, has also been demonstrated to ameliorate lupus in the murine NZB/NZW F1 and MRL/lpr models of the disease [50,51]. In a case report of a patient developing cutaneous lupus after withdrawal of ruxolitinib, which targets JAK1 and JAK2, the investigators observed that the skin lesions resolved completely after reinitiating the treatment, and that the drug decreased expression of ISGs in cells cultured *in vitro* [52].

The B cell activating factor (BAFF) is essential for B cell survival, differentiation and antibody production, including autoantibodies. It is induced by type I IFN via an IFN regulated factor (IRF)-controlled mechanism, the levels are enhanced in SLE and pSS, and the expression correlates positively with the IFN signature [53]. BAFF blockade is partly efficacious in SLE and SS, with indications that patients with high type I IFN, and accordingly higher BAFF levels, may be the best responders [54–56]. Notably, BAFF levels have been observed to increase upon treatment with rituximab [57], a phenomenon suggested to influence the efficacy of this CD20-directed B cell depleting therapy and prompting studies on combined treatment. Interestingly, although treatment by rituximab has generated variable results in studies of both SLE and pSS [58], several case reports indicate that sequential treatment by rituximab and belimumab may be beneficial [59–61]. Larger studies are needed and underway to draw any firm conclusion on the effects of sequential or combined B cells depleting and BAFF blocking therapy.

To prevent the production type I IFN, targeting of pDCs is also a current therapeutic strategy. pDCs have the highest capacity of all described cells to produce type I IFNs, and a C-type lectin, BDCA2, is a cell-specific receptor that upon ligation inhibits production of type I IFN and other inflammatory molecules (Fig. 2). Engagement of BDCA2 by a humanized mAb (24F4A) has been shown to lead to its internalization and following inhibition of TLR-induced type I IFN by pDCs [62], and a phase 1b study indicates that another BDCA2 binding monoclonal (BIIB059) reduces ISGs in peripheral blood and skin of SLE patients [63].

In all, several different approaches and strategies to accomplish IFN blockade are currently yielding promising results in both early and more advanced stages of trials en route to clinical practice, and confirm that targeting the IFN pathways may be a productive way forward for therapeutics in SLE.

CONCLUSION

Recent studies have demonstrated that not only there are many different reasons behind the ongoing production of type I IFNs in patients with SLE and pSS but also revealed that produced IFN have a major impact on the immune system. The effects on target cells are at least partially dependent on epigenetic modulation, generating cellular responses that contribute to both loss of tolerance and chronic inflammation. Several new therapies aiming to downregulate the type I IFN system are in clinical trials, and a challenge is to control this fundamental system without inducing increased susceptibility to infectious diseases in treated patients.

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Conflicts of interest

L.R. has received a research grant from AstraZeneca and received honoraria from AstraZeneca, Biogen and UCB. The remaining authors have no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Ronnblom L, Eloranta ML. The interferon signature in autoimmune diseases. Curr Opin Rheumatol 2013; 25:248–253.
2. Doedens JR, Jones WD, Hill K, *et al.* Blood-borne Rna correlates with disease activity and Ifn-stimulated gene expression in systemic lupus erythematosus. J Immunol 2016; 197:2854–2863.
3. Mavragani CP, Sagalovskiy I, Guo Q, *et al.* Expression of long interspersed nuclear element 1 retroelements and induction of type I interferon in patients with systemic autoimmune disease. Arthritis Rheumatol 2016; 68: 2686–2696.
4. Shao WH, Shu DH, Zhen Y, *et al.* Prion-like aggregation of mitochondrial antiviral signaling protein in lupus patients is associated with increased levels of type I interferon. Arthritis Rheumatol 2016; 68:2697–2707.

5. Lee JY, Park JK, Lee EY, *et al.* Circulating exosomes from patients with systemic lupus erythematosus induce an proinflammatory immune response. Arthritis Res Ther 2016; 18:264.
6. Rose T, Grutzkau A, Klotsche J, *et al.* Are interferon-related biomarkers advantageous for monitoring disease activity in systemic lupus erythematosus? A longitudinal benchmark study. Rheumatology (Oxford) 2017; 56: 1618–1626.
7. ▪ Rodero MP, Decalf J, Bondet V, *et al.* Detection of interferon alpha protein reveals differential levels and cellular sources in disease. J Exp Med 2017; 214:1547–1555.

Using a single-molecule array (Simoa) digital ELISA technology with mAbs from two patients with APS1/APECED mutations, the authors were able to record attomolar concentrations of IFNα. An ultrasensitive and reliable IFNα protein assay for clinical use is much needed and this is a step towards that goal.

8. ▪▪ Langefeld CD, Ainsworth HC, Cunninghame Graham DS, *et al.* Transancestral mapping and genetic load in systemic lupus erythematosus. Nat Commun 2017; 8:16021.

This is the largest genetic study in patients with SLE and the study establishes that more than 100 risk loci are associated with the disease. Several ethnic groups were investigated and both shared and ancestry-dependent loci were identified.

9. Wen L, Zhu C, Zhu Z, *et al.* Exome-wide association study identifies four novel loci for systemic lupus erythematosus in Han Chinese population. Ann Rheum Dis 2018; 77:417.
10. Taylor KE, Wong Q, Levine DM, *et al.* Genome-wide association analysis reveals genetic heterogeneity of Sjogren's syndrome according to ancestry. Arthritis Rheumatol 2017; 69:1294–1305.
11. Qu SS, Du Y, Chang SH, *et al.* Common variants near Ikzf1 are associated with primary Sjogren's syndrome in Han Chinese. PLos One 2017; 12: e0177320.
12. ▪ Hagberg N, Joelsson M, Leonard D, *et al.* The Stat4 Sle risk allele Rs7574865[T] is associated with increased Il-12-induced IFN-γ production in T cells from patients with SLE. Ann Rheum Dis 2018. [Epub ahead of print]

The functional consequences of the *STAT4* risk gene variant in intron 3 has been unknown for 10 years. This study establishes a molecular connection between the risk gene variant and an increased IL-12 and IFNα response in T cells from patients with SLE. JAK inhibitors could block the T cell response.

13. Cheng J, Wu R, Long L, *et al.* Mirna-451a targets Ifn regulatory factor 8 for the progression of systemic lupus erythematosus. Inflammation 2017; 40: 676–687.
14. ▪ Smith S, Fernando T, Wu PW, *et al.* Microrna-302d targets Irf9 to regulate the Ifn-induced gene expression in Sle. J Autoimmun 2017; 79:105–111.

MicroRNA can control the IFN response, and in this study, the authors show that microRNA-320d is low in monocytes from patients with SLE and correlates with an IFN signature. Treatment of pristane-induced lupus with miR-320d downregulated the IFN response, but effect on clinical disease activity remains to be established.

15. Demirkaya E, Zhou Q, Smith CK, *et al.* Brief report: deficiency of complement 1r subcomponent in early-onset systemic lupus erythematosus: the role of disease-modifying alleles in a monogenic disease. Arthritis Rheumatol 2017; 69:1832–1839.
16. Ghodke-Puranik Y, Dorschner JM, Vsetecka DM, *et al.* Lupus-associated functional polymorphism in Pnp causes cell cycle abnormalities and interferon pathway activation in human immune cells. Arthritis Rheumatol 2017; 69:2328–2337.
17. Kariuki SN, Ghodke-Puranik Y, Dorschner JM, *et al.* Genetic analysis of the pathogenic molecular sub-phenotype interferon-alpha identifies multiple novel loci involved in systemic lupus erythematosus. Genes Immun 2015; 16: 15–23.
18. ▪▪ Faridi MH, Khan SQ, Zhao W, *et al.* Cd11b activation suppresses TLR-dependent inflammation and autoimmunity in systemic lupus erythematosus. J Clin Invest 2017; 127:1271–1283.

ITGAM encodes CD11b and SLE-associated gene variants confer reduced inhibition of the type I IFN signalling pathway. In the present study, the authors show that activation of CD11b with a small molecule block the TLR4 activation and protects MLR/lpr mice from end organ damage. The effect on IFN production by TLR 7 and 9 activation was, however, not investigated.

19. ▪ Li H, Reksten TR, Ice JA, *et al.* Identification of a Sjogren's syndrome susceptibility locus at Oas1 that influences isoform switching, protein expression, and responsiveness to type I interferons. PLoS Genet 2017; 13:e1006820.

This study shows that a risk gene variant for pSS affects the transcription of a classical type I IFN-induced gene - 2'-5'-oligoadenylate synthetase 1 (OAS1) and causes reduced protein expression. This could contribute to an increased sensitivity for viral infections in pSS patients.

20. Silva JA, Lima SC, Addobbati C, *et al.* Association of interferon-induced helicase C domain (Ifih1) gene polymorphisms with systemic lupus erythematosus and a relevant updated meta-analysis. Genet Mol Res 2016; 15:1–9.
21. Munroe ME, Pezant N, Brown MA, *et al.* Association of Ifih1 and pro-inflammatory mediators: potential new clues in SLE-associated pathogenesis. PLoS One 2017; 12:e0171193.
22. Howard Tripp N, Tarn J, Natasari A, *et al.* Fatigue in primary Sjogren's syndrome is associated with lower levels of proinflammatory cytokines. RMD Open 2016; 2:e000282.
23. Padgett LE, Tse HM. Nadph oxidase-derived superoxide provides a third signal for Cd4 T cell effector responses. J Immunol 2016; 197:1733–1742.
24. Khmaladze I, Saxena A, Nandakumar KS, Holmdahl R. B-cell epitope spreading and inflammation in a mouse model of arthritis is associated with a deficiency in reactive oxygen species production. Eur J Immunol 2015; 45:2243–2251.
25. Holmdahl R, Sareila O, Olsson LM, *et al.* Ncf1 polymorphism reveals oxidative regulation of autoimmune chronic inflammation. Immunol Rev 2016; 269:228–247.
26. ▪▪ Zhao J, Ma J, Deng Y, *et al.* A missense variant in Ncf1 is associated with susceptibility to multiple autoimmune diseases. Nat Genet 2017; 49: 433–437.

These two publications [26▪▪,27▪▪] show that a genetically determined reduced ROS function is strongly associated with an increased risk for SLE. The articles add new information to the previous reported association between *SLE* and *NCF2* gene variants causing impaired ROS activity.

27. ▪▪ Olsson LM, Johansson AC, Gullstrand B, *et al.* A single nucleotide polymorphism in the Ncf1 gene leading to reduced oxidative burst is associated with systemic lupus erythematosus. Ann Rheum Dis 2017; 76:1607–1613.

These two publications [26▪▪,27▪▪] show that a genetically determined reduced ROS function is strongly associated with an increased risk for SLE. The articles add new information to the previous reported association between *SLE* and *NCF2* gene variants causing impaired ROS activity.

28. Buie JJ, Renaud LL, Muise-Helmericks R, Oates JC. Ifn-alpha negatively regulates the expression of endothelial nitric oxide synthase and nitric oxide production: implications for systemic lupus erythematosus. J Immunol 2017; 199:1979–1988.
29. Tyden H, Lood C, Gullstrand B, *et al.* Endothelial dysfunction is associated with activation of the type I interferon system and platelets in patients with systemic lupus erythematosus. RMD Open 2017; 3:e000508.
30. Rose T, Szelinski F, Lisney A, *et al.* Siglec1 is a biomarker of disease activity and indicates extraglandular manifestation in primary Sjogren's syndrome. RMD Open 2016; 2:e000292.
31. ▪ Lisney AR, Szelinski F, Reiter K, *et al.* High maternal expression of siglec1 on monocytes as a surrogate marker of a type i interferon signature is a risk factor for the development of autoimmune congenital heart block. Ann Rheum Dis 2017; 76:1476–1480.

This study shows that pregnant women with SSA antibodies and high expression of SIGLEC-1 on monocytes have an increased risk for giving birth to children with autoimmune congenital heart block. The study was small, but suggests that anti-SSA positive patients should be investigated for SIGLEC-1 expression and treated with hydroxychloroquine.

32. Berggren O, Hagberg N, Alexsson A, *et al.* Plasmacytoid dendritic cells and Rna-containing immune complexes drive expansion of peripheral B cell subsets with an Sle-like phenotype. PLoS One 2017; 12:e0183946.
33. ▪ Menon M, Blair PA, Isenberg DA, Mauri C. A regulatory feedback between plasmacytoid dendritic cells and regulatory B cells is aberrant in systemic lupus erythematosus. Immunity 2016; 44:683–697.

This study shows that pDCs from SLE patients promote plasmablast differentiation but fail to induce Breg cells. IFNα could induce the same aberration in cells from healthy individuals. Thus, type I IFN seems to promote a dysregulated pDC and B cell interaction in SLE.

34. Zhao J, Kubo S, Nakayamada S, *et al.* Association of plasmacytoid dendritic cells with b cell infiltration in minor salivary glands in patients with Sjogren's syndrome. Mod Rheumatol 2016; 26:716–724.
35. Aqrawi LA, Ivanchenko M, Bjork A, *et al.* Diminished Cxcr5 expression in peripheral blood of patients with Sjogren's syndrome may relate to both genotype and salivary gland homing. Clin Exp Immunol 2018. [Epub ahead of print]
36. Espinosa A, Dardalhon V, Brauner S, *et al.* Loss of the lupus autoantigen Ro52/Trim21 induces tissue inflammation and systemic autoimmunity by dysregulating the Il-23-Th17 pathway. J Exp Med 2009; 206:1661–1671.
37. Mahony R, Broadbent L, Maier-Moore JS, *et al.* The Rna binding protein La/Ss-B promotes Rig-I-mediated Type I and Type III Ifn responses following sendai viral infection. Sci Rep 2017; 7:14537.
38. Maria NI, Steenwijk EC, IJpma AS, *et al.* Contrasting expression pattern of Rna-sensing receptors Tlr7, Rig-I and Mda5 in interferon-positive and interferon-negative patients with primary Sjogren's syndrome. Ann Rheum Dis 2017; 76:721–730.
39. Pan ZJ, Horton CG, Lawrence C, Farris AD. Plasmacytoid dendritic cells and type 1 interferon promote peripheral expansion of forkhead box protein 3(+) regulatory T cells specific for the ubiquitous Rna-binding nuclear antigen La/Sjogren's syndrome (Ss)-B. Clin Exp Immunol 2016; 186:18–29.
40. Maria NI, van Helden-Meeuwsen CG, Brkic Z, *et al.* Association of increased treg cell levels with elevated indoleamine 2,3-dioxygenase activity and an imbalanced kynurenine pathway in interferon-positive primary Sjogren's syndrome. Arthritis Rheumatol 2016; 68:1688–1699.
41. Imgenberg-Kreuz J, Sandling JK, Almlof JC, *et al.* Genome-wide DNA methylation analysis in multiple tissues in primary Sjogren's syndrome reveals regulatory effects at interferon-induced genes. Ann Rheum Dis 2016; 75: 2029–2036.
42. Charras A, Konsta OD, Le Dantec C, *et al.* Cell-specific epigenome-wide DNA methylation profile in long-term cultured minor salivary gland epithelial cells from patients with Sjogren's syndrome. Ann Rheum Dis 2017; 76:625–628.

43. Ulff-Moller CJ, Asmar F, Liu Y, *et al.* Twin DNA methylation profiling
■ reveals flare-dependent interferon signature and B-cell promoter hypermethylation in systemic lupus erythematosus. Arthritis Rheumatol 2018; 70:878–890.
Several cell types from monozygotic and dizygotic twins discordant for SLE were investigated with the HumanMethylation450K BeadChip. The majority of differentially methylated genes were hypomethylated and nearly 40% were connected to the IFN pathway. Prominent hypomethylation in IFN-regulated genes were associated to risk for flare.
44. Park SH, Kang K, Giannopoulou E, *et al.* Type I interferons and the cytokine
■■ Tnf cooperatively reprogram the macrophage epigenome to promote inflammatory activation. Nat Immunol 2017; 18:1104–1116.
The tolerogenic effect of TNF on the TLR3 response by macrophages was abolished by IFNα exposure and this effect was mediated via epigenetic modifications.
45. Kalunian KC, Merrill JT, Maciuca R, *et al.* A phase Ii study of the efficacy and safety of rontalizumab (rhumab interferon-alpha) in patients with systemic lupus erythematosus (Rose). Ann Rheum Dis 2016; 75:196–202.
46. Khamashta M, Merrill JT, Werth VP, *et al.* Sifalimumab, an anti-interferon-alpha monoclonal antibody, in moderate to severe systemic lupus erythematosus: a randomised, double-blind, placebo-controlled study. Ann Rheum Dis 2016; 75:1909–1916.
47. Ducreux J, Houssiau FA, Vandepapeliere P, *et al.* Interferon alpha kinoid induces neutralizing anti-interferon alpha antibodies that decrease the expression of interferon-induced and B cell activation associated transcripts: analysis of extended follow-up data from the interferon alpha kinoid phase I/Ii study. Rheumatology (Oxford) 2016; 55:1901–1905.
48. Furie R, Khamashta M, Merrill JT, *et al.* Anifrolumab, an anti-interferon-alpha
■ receptor monoclonal antibody, in moderate-to-severe systemic lupus erythematosus. Arthritis Rheumatol 2017; 69:376–386.
This is the first clinical trial with a mAb against the type I IFN receptor in SLE and the study suggests that patients with a high IFN score should be selected for this treatment.
49. Bialas AR, Presumey J, Das A, *et al.* Microglia-dependent synapse loss in type
■ I interferon-mediated lupus. Nature 2017; 546:539–543.
Type I IFN can activate microglia to phagocytose synapses, which could be one mechanism behind neuropsychiatric lupus and the observed cognitive dysfunction in many patients.
50. Ikeda K, Hayakawa K, Fujishiro M, *et al.* Jak Inhibitor has the amelioration effect in lupus-prone mice: the involvement of Ifn signature gene downregulation. BMC Immunol 2017; 18:41.
51. Furumoto Y, Smith CK, Blanco L, *et al.* Tofacitinib ameliorates murine lupus and its associated vascular dysfunction. Arthritis Rheumatol 2017; 69: 148–160.
52. Klaeschen AS, Wolf D, Brossart P, *et al.* Jak inhibitor ruxolitinib inhibits the expression of cytokines characteristic of cutaneous lupus erythematosus. Exp Dermatol 2017; 26:728–730.
53. Sjostrand M, Johansson A, Aqrawi L, *et al.* The expression of Baff is controlled by Irf transcription factors. J Immunol 2016; 196:91–96.
54. Mariette X, Seror R, Quartuccio L, *et al.* Efficacy and safety of belimumab in primary Sjogren's syndrome: results of the Beliss open-label phase Ii study. Ann Rheum Dis 2015; 74:526–531.
55. De Vita S, Quartuccio L, Seror R, *et al.* Efficacy and safety of belimumab given for 12 months in primary Sjogren's syndrome: the Beliss open-label phase Ii study. Rheumatology (Oxford) 2015; 54:2249–2256.
56. Seror R, Nocturne G, Lazure T, *et al.* Low numbers of blood and salivary natural killer cells are associated with a better response to belimumab in primary Sjogren's syndrome: results of the Beliss study. Arthritis Res Ther 2015; 17:241.
57. Vallerskog T, Heimburger M, Gunnarsson I, *et al.* Differential effects on Baff and april levels in rituximab-treated patients with systemic lupus erythematosus and rheumatoid arthritis. Arthritis Res Ther 2006; 8:R167.
58. Bowman SJ, Everett CC, O'Dwyer JL, *et al.* Randomized controlled trial of rituximab and cost-effectiveness analysis in treating fatigue and oral dryness in primary Sjogren's syndrome. Arthritis Rheumatol 2017; 69: 1440–1450.
59. Kraaij T, Huizinga TW, Rabelink TJ, Teng YK. Belimumab after rituximab as maintenance therapy in lupus nephritis. Rheumatology (Oxford) 2014; 53:2122–2124.
60. De Vita S, Quartuccio L, Salvin S, *et al.* Sequential therapy with belimumab followed by rituximab in Sjogren's syndrome associated with B-cell lymphoproliferation and overexpression of Baff: evidence for long-term efficacy. Clin Exp Rheumatol 2014; 32:490–494.
61. Gualtierotti R, Borghi MO, Gerosa M, *et al.* Successful sequential therapy with rituximab and belimumab in patients with active systemic lupus erythematosus: a case series. Clin Exp Rheumatol 2018. [Epub ahead of print]
62. Pellerin A, Otero K, Czerkowicz JM, *et al.* Anti-Bdca2 monoclonal antibody inhibits plasmacytoid dendritic cell activation through Fc-dependent and Fc-independent mechanisms. EMBO Mol Med 2015; 7:464–476.
63. Furie R, Werth V, Merola J, *et al.* 83|Biib059, a monoclonal antibody targeting Bdca2, demonstrates evidence of proof of biological activity in subjects with cutaneous lupus. Lupus Sci Med 2017; 4:A35–A135.

Glucocorticoids and antimalarials in systemic lupus erythematosus: an update and future directions

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Purpose of review
The purpose of this review is highlighting the most recent evidence on the clinical efficacy and toxicity of glucocorticoids and antimalarials in systemic lupus erythematosus (SLE) and provide recommendations on their current use.

Recent findings
Glucocorticoid toxicity is well known. Recent data confirm the increased risk of infection and damage accrual. An observational study form Hong Kong has seen increased mortality among users of high-dose prednisone regimes. Several studies support the efficacy of medium-low doses and methyl-prednisolone pulses in lupus patients, both with and without nephritis.
New data confirm the effects of antimalarials in preventing SLE activity, damage and infections, and in decreasing mortality. New screening recommendations for hydroxychloroquine maculopathy have been recently published. Combining mepacrine and hydroxychloroquine in patients with refractory cutaneous and/or articular lupus activity has proved highly effective.

Summary
Universal therapy with hydroxychloroquine should be aimed to patients with SLE without contraindications. Doses greater than 4 mg/kg/day should be avoided and regular eye screening warranted to minimize the risk of macular toxicity. Every effort should be made to reduce the dose of oral glucocorticoids. In moderate-severe flares, pulse methyl-prednisolone are more effective and much less toxic than increasing the oral doses of prednisone.

Keywords
damage, efficacy, hydroxychloroquine, lupus activity, mepacrine, methyl-prednisolone, prednisone, survival, toxicity

Glucocorticoids and antimalarials constitute the basis of systemic lupus erythematosus (SLE) therapy. The recent British Society for Rheumatology guidelines for the management of lupus recommend both in all clinical scenarios [1▪]; however, the respective role of steroids and hydroxychloroquine has substantially changed over the time: we are increasingly more concerned about the toxicity of the former and more aware of the long-term beneficial effects of the latter.

The purpose of this review is highlighting the most recent evidence on the clinical efficacy and toxicity of glucocorticoids and antimalarials in SLE. Based on this new and previous data, recommendations on their use in lupus will be made.

GLUCOCORTICOIDS

Glucocorticoid-related toxicity

Table 1 Glucocorticoids provoke a myriad of safety concerns. Infections, one of the main causes of morbidity and mortality in SLE, are among them. A systematic review of 32 clinical trials involving 2611 lupus nephritis patients showed that the highest crude rate of serious infections was associated with high glucocorticoid doses [2▪]. A retrospective, new-user study from Kaiser Permanente including 3030 SLE patients showed an four-fold risk for serious infections in patients who received doses of prednisone 15 mg/day or less without antimalarials compared with

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KEY POINTS

- Glucocorticoids and antimalarials have constituted the cornerstone of SLE therapy. However, their respective roles in long-term management are rapidly changing.
- Recent data confirm the increased risk for infections, damage accrual, and death in patients treated with high-dose oral glucocorticoids.
- Long-term hydroxychloroquine reduces damage and increases survival of lupus patients; new data confirm these effects in patients from all ethnicities.
- Clinically apparent hydroxychloroquine maculopathy is rare even after prolonged use at doses less than 5 mg/kg/day and following the current recommendations for eye screening.
- Combination therapy, always including hydroxychloroquine, and, in moderate-severe cases, pulse methyl-prednisolone and early immunosuppressive drugs, spare oral glucocorticoids with a similar control of SLE activity and less side-effects.

patients on antimalarials only; this increased risk disappeared with the concomitant treatment with antimalarials. However, the incidence of serious infections in patients treated with prednisone doses greater than 15 mg/day was much higher than in any of the previous groups, whether treated with concomitant antimalarials or not [3]. Thus, these results suggest that the use of prednisone more than 15 mg/day overwhelms the protective effects of antimalarials against infections. Recent data from a retrospective registry from the Spanish Society of Rheumatology [4▪▪], including more than 3500 lupus patients, observed that infections of all types were more frequent among current users of glucocorticoids compared with nonusers. A history of glucocorticoid treatment at doses at least 10 mg/day was associated with a 27% higher risk of severe infections. Of note, 45% of patients who died from infection were on prednisone at least 10 mg/day, compared with 9% of survivors [4▪▪].

The effect of glucocorticoids on damage accrual in SLE has been consistently shown in several publications [5–7]. This effect is dose dependent, with doses more than 7.5 mg/day being associated with new damage [6,7]. In 2016, an Australian cohort study, including 132 patients with a mean follow-up of 4 years, observed damage accrual in 42% of glucocorticoid-exposed patients vs. 15% of nonexposed. Interestingly, permanent damage was also accrued in the range of doses between 4.5 and 7.5 mg/day, in Systemic Lupus International Collaborating Clinics (SLICC) Damage Index domains both traditionally and not traditionally related with glucocorticoids [8▪]. A Hungarian study of 357 SLE patients followed during a mean of 20 years showed a correlation between high and cumulative glucocorticoid doses and damage, with a strong association with cataracts and osteoporosis [9]. Recently, a cohort study including 803 patients from Hong Kong with a mean follow-up of 10 years found that prednisone more than 0.6 mg/kg/day during at least 4 weeks was associated with a propensity score-adjusted risk of all-cause mortality higher than 14-fold [10▪▪]. Of note, methyl-prednisolone pulses have not been associated with damage [5,6].

Despite the well-known toxicity of glucocorticoids, they are still extensively used at doses above the safety cut offs. The SLICC group reported an average dose of prednisone of 20 mg/day at enrolment, 10 mg/day at 1 year and 8 mg/day after 2 years of follow-up [11]. In the Spanish multicentric inception cohort Registro Español de Lupus Eritematoso Sistémico (RELES), 38% of patients were treated with average doses of prednisone >7.5 mg/day within the first year after the diagnosis [12]. Moreover, a study from the same group proved that doses given in the first month of treatment were predictive of doses used during the following 11 months, regardless the initial degree of disease activity [13▪].

Efficacy of reduced doses of glucocorticoids

Being well aware of glucocorticoid toxicity, the key question is whether high doses of prednisone are actually needed to suppress SLE activity. One randomised controlled trial (RCT) [14] and three observational studies [15,16,17▪] in patients with lupus nephritis suggest that lower doses can be equally effective, with similar [14,15] or even better [16,17▪] response rates. The Rituxilup regime, consisting of rituximab and methyl-prednisolone, followed by maintenance treatment with mycophenolate mofetil and no oral steroids, resulted in high remission rates in patients with class III, IV, and V lupus nephritis [18]. After more than 5 years of follow-up, the majority of patients are in remission with preserved renal function [19]. In a group of patients without or with mild forms of lupus nephritis presenting with a Systemic Lupus Erythematosus Disease Activity Index (SLEDAI) score at least 6, therapy with mean initial doses of prednisone of 11 mg/day resulted in a similar reduction of SLEDAI scores at 1 year and less damage at 5 years compared with a group treated with mean initial doses of 63.4 mg/day [20]. Hydroxychloroquine was used in 100 vs. 33% of patients and methyl-prednisolone pulses were given to 34 vs. 10%, respectively.

Pulse methyl-prednisolone is an essential element for effectively treating active lupus using lower doses of oral prednisone. The use of methyl-prednisolone

Table 1. Main studies focused on glucocorticoids in systemic lupus erythematosus

Reference	Study design and characteristics	Main results
Singh *et al.* [2▪]	Systematic review of 32 randomized controlled trials of lupus nephritis treatment	High doses of GC showed the highest crude rate of infections compared with other immunosuppressive agents (including CYC, AZA, MMF, and TAC)
	11 studies provided data about GC	
	2611 patients were included	
Herrinton *et al.* [3]	3030 patients from a Californian cohort	GC at doses ≤15 mg/day without antimalarials showed a four-fold increase in the risk of serious infections
	Mean follow-up per patient 4 years	
	Ethnicity diverse, predominantly Whites	
Rúa-Figueroa *et al.* [4▪▪]	3658 Spanish lupus patients from Rheumatology services	GC use at the time of infection was associated with viral, fungal, and bacterial isolation
	Mean follow-up 10 years	Death occurred more frequently in patients treated with GC ≥10 mg/day
	93% Whites	
Al Sawah *et al.* [7]	2199 patients from the Hopkins Lupus cohort	Patients who received prednisone ≥7.5 mg/day were more likely to develop any new organ damage over time
	Mean follow-up 6.2 years	Prednisone doses ≥7.5 mg/day increased the risk of developing cataracts, osteoporotic fractures, and cardiovascular damage
	Ethnicity diverse, 55% Whites, 38% African-American	An increase in average prednisone doses of 1 mg/day increases by 3% the risk of cataracts and osteoporotic fractures
Apostoloupoulos *et al.* [8▪]	162 patients	GC were associated with overall damage, both in SDI domains traditionally associated with corticoid damage and in nontraditionally associated
	Mean follow-up 3.6 years	At higher doses, grater damage
	Ethnicity diverse, 40% Asian descendent	Maintenance doses over 4.42 mg/day were probably associated with damage
Tarr *et al.* [9]	357 Hungarian SLE patients	Patients receiving higher-dose GC therapy had higher damage score value
	Mean follow-up 19.14 years	Strong positive correlation between damage and cumulative GC doses, independently from other variables
Mok *et al.* [10▪▪]	803 patients from Hong Kong	Prednisone >0.6 mg/kg/day during at least 4 weeks was associated with a propensity score-adjusted risk of all-cause mortality higher than 14-fold
	Mean follow-up 10.8 years	
	All Chinese	
Ruiz-Irastorza *et al.* [12]	223 Spanish lupus patients from an inception cohort	Prednisone doses in the first month predicted doses during the following 11 months. Those treated with higher initial doses had less chance to be with average doses ≤7.5 mg/day at month 12
	Follow-up 1 year	Methyl-prednisolone pulses reduced the likelihood to be treated with average medium-high doses of prednisone at month 12
Ruiz-Irastorza *et al.* [13▪]	223 Spanish lupus patients from an inception cohort	65% of patients received GC during the first year of follow-up
	Follow-up 1 year	38% of patients were treated with average doses >7.5 mg/day during the first year
Ruiz-Irastorza *et al.* [17▪]	29 lupus nephritis patients included in the CC compared with 44 lupus nephritis patients from the BC	Patients in the CC achieved more CR at 6 (69 vs. 30%, $P=0.001$) and 12 months (86 vs. 43%, $P<0.001$)
	Maximum doses of prednisone, number of weeks until 5 mg/day maintenance doses and mean doses at 6 months were lower in the CC	The reduction of proteinuria was higher in the CC
	The number of pulses of methyl-prednisolone was higher in the CC, but not the cumulative dose	The only treatment independently associated with a CR was the number of methyl-prednisolone pulses
		There was a significantly lower risk of developing GC-related toxicity in the CC
Ruiz-Arruza *et al.* [20]	30 patients included in the CC without or with mild forms of lupus nephritis, presenting with SLEDAI ≥6, treated with prednisone doses ≤30 mg/day (mean 11 mg/day) compared with 30 patients from the historic cohort treated with prednisone doses >30 mg/day (mean 63.4 mg/d)	Among patients in the low-dose prednisone group, hydroxychloroquine was used in 100 vs. 33% of patients and methyl-prednisolone pulses were given to 34 vs. 10%, respectively. Patients in the low-dose prednisone group had similar control of activity at 1 year and accrued less damage at 5 years
	Follow-up 5 years	
Danza *et al.* [21]	164 patients with flares of immune-mediated diseases treated with pulse methyl-prednisolone at different doses	Patients treated with cumulative doses ≤1.5 g had more chance to achieve a favourable response
Ruiz-Arruza *et al.* [23▪]	74 inception patients in the CC were compared with 213 historic controls	Less patients accrued damage in the first year and at the end of the follow-up in the inception cohort compared with the historic cohort
	The study compared a protocolized scheme with restrictive use of glucocorticoids vs. a traditional scheme of high doses	Cardiovascular and glucocorticoid-related damage were less likely to occur in the inception cohort
	Mean follow-up 8.2 vs. 9.6 years	Lupus-related damage was similar in both groups

AZA, azathioprine; BC, Bordeaux Hospital cohort; CC, Lupus-Cruces cohort; CR, complete remission; CYC, cyclophosphamide; GC, glucocorticoids; MMF, mycophenolate mofetil; SDI, SLICC damage index; SLEDAI, systemic lupus erythematosus disease activity index; SLE, systemic lupus erythematosus; TAC, tacrolimus.

pulses was a predictor of a more rapid reduction of prednisone doses in the RELES cohort, particularly in patients with a SLEDAI score at least 6 [13▪]. In a recent observational study comparing patients with lupus nephritis from the Lupus-Cruces and Lupus-Bordeaux cohorts, the number (rather than the total dose) of methyl-prednisolone pulses was an independent predictor of complete response and of reduced glucocorticoid-related toxicity. Lupus-Cruces patients were treated with three consecutive 250–500 mg pulses and then with additional 125 mg pulses every 2 weeks, previous to each intravenous cyclophosphamide dose [17▪]. Indeed, a recent study has shown similar clinical efficacy and lower rates of infections among patients treated with pulses less than 500 mg each compared with those receiving 1000 mg [21]. These results reinforce previous data [22].

The long-term effects of a therapeutic approach using reduced doses of oral prednisone and universal hydroxychloroquine, adding frequent pulse methyl-prednisolone and early immunosuppressive drugs in active patients, have been recently studied in an inception observational cohort. Compared with historic controls, patients received lower maximum prednisone doses (mean 15 vs. 36 mg/d), lower average doses during 5 years of follow-up (2.8 vs. 9.4 mg/day) and were treated more frequently with hydroxychloroquine (100 vs. 52%), methyl-prednisolone pulses (32 vs. 12%), and early immunosuppressive drugs (23 vs. 13% in the first year). Global damage accrual was reduced, with less glucocorticoid-related and cardiovascular damage and similar lupus-related damage [23▪]. This is the first study in showing that lowering oral doses of prednisone using combination therapy reduces glucocorticoid toxicity without compromising long-term SLE control.

ANTIMALARIALS

Clinical efficacy of antimalarials

Table 2 antimalarials, particularly hydroxychloroquine, have already shown a wide range of beneficial effects in SLE [24]. Some of them were addressed in a recent study from the longitudinal cohort of the University of Toronto in which 459 SLE patients were divided into three groups (antimalarials more than 60% and less than 60% of the time in the first 5 years and no antimalarial use). The authors found that disease activity, damage accrual, and the cumulative dose of steroids were significantly lower in the first group [25▪].

Beyond lupus activity, hydroxychloroquine has shown an independent protective effect over infections in several studies [26–28]. More recently, in a large multicentric cohort of 3658 SLE patients, an inverse association was found between the time of exposure to antimalarials and the risk of severe infection [4▪▪]. Also, results from an observational prospective cohort published in 2007 found a negative association between cancer risk and antimalarial treatment [29]. A 2017 population-based study did not reproduce these results – the risk of developing cancer did not differ between patients with connective tissue diseases exposed and nonexposed to hydroxychloroquine – however, the risk of metastases and death were significantly lower in the exposed group [30].

Between 2005 and 2010, several observational prospective cohort studies found an association between hydroxychloroquine use and improved survival of SLE patients [31–33]. Hydroxychloroquine was also an independent predictor of survival in a retrospective study including 495 SLE patients with biopsy-proven nephritis [34]. More recently, Mok *et al.* [10▪▪] confirmed the results of the preceding studies in a longitudinal cohort of 803 Chinese SLE patients, with a propensity-score adjusted hazard ratio for all-cause mortality of 0.59. Thus, the benefits of antimalarials on survival have been already shown in lupus patients of all ethnicities.

Effects on pregnancy

The European League Against Rheumatism 2016 Task Force and the British Society of Rheumatology/British Health Professionals in Rheumatology guidelines strongly recommend keeping hydroxychloroquine treatment during pregnancy and lactation [35,36]. Unfortunately, despite such data and recommendations, the use of hydroxychloroquine during pregnancy is still strikingly low, although it has remarkably increased within the last decade [37]. In recent studies in pregnant patients, Leroux *et al.* [38] analysed 118 pregnancies and found a significantly lower rate of preterm delivery and growth restriction among neonates from mothers under hydroxychloroquine treatment. In 110 pregnancies from a Dutch SLE cohort, the authors found that the use of hydroxychloroquine was associated with a longer duration of pregnancy among preterm births [39].

In anti-Ro positive patients, three studies pointed to a reduction in the risk of recurrent foetal heart block with the maternal use of hydroxychloroquine during pregnancy [40,41]. A more recent retrospective cohort study of 268 consecutive pregnancies in Ro-positive women found a decreased rate of cardiac neonatal lupus among babies born to antimalarial-exposed mothers (1.3 vs. 6.1% in nonexposed). Although the difference did not reach the statistical significance ($P = 0.07$), the Bayesian analysis showed a 98.7% probability that antimalarials given antenatally protect against cardiac neonatal lupus. On the

Table 2. Main studies focused on antimalarials in systemic lupus erythematosus

Reference	Study design	Main results
Pakchotanon *et al.* [25▪]	Longitudinal cohort of 459 SLE patients from the Toronto Lupus Clinic, divided into three groups (antimalarials >60% in the first 5 years; <60% of de time; no antimalarial use) Minimum follow-up 5 years	The occurrence of flares, changes in the SDI, and cumulative dose of steroids were lower in the first group
Rúa-Figueroa *et al.* [4▪▪]	3658 Spanish lupus patients from Rheumatology services Mean follow-up 10 years 93% Whites	Negative association between the time of exposure to antimalarials and the risk of severe infection
Fardet *et al.* [30]	Population-based cohort study from the United Kingdom 8999 individuals with connective tissue diseases exposed to HCQ vs. 24 118 unexposed individuals	Patients under long-term exposure to HCQ did not show a lower risk of cancer The risk of metastases was lower during the HCQ exposure period. The risk of death was lower in the HCQ-exposed group
Mok *et al.* [10▪▪]	803 patients from Hong Kong Mean follow-up 10.8 years All Chinese	The use of HCQ was associated with a probability of better survival. The propensity score-adjusted hazard ratio for all-cause mortality was 0.59 (95% CI 0.37–0.93)
Bermas *et al.* [37]	Data from public and private health insurance programs 5300 pregnancies of SLE patients from 2001 to 2015	Women taking HCQ during pregnancy rose from 2.7% in 2001 to 37.7% in 2015
Leroux *et al.* [38]	Retrospective cohort study 118 pregnancies from 2001 to 2011: 40 on HCQ, 77 unexposed	Lower rates of preterm delivery and IUGR in neonates from mothers under HCQ treatment
Kroese *et al.* [39]	Retrospective study 110 pregnancies within a Dutch SLE cohort from 2000 to 2015 30/110 under HCQ	HCQ group showed a trend towards a lower prednisone dose Among preterm births, the use of HCQ was associated with a longer duration of pregnancy
Barsalou *et al.* [42▪]	Retrospective cohort study of 268 consecutive anti-Ro positive pregnancies from 1984 to 2013 73/268 exposed to HCQ	98.7% probability that antimalarials given antenatally protect against cardiac (but not noncardiac) neonatal lupus
Tselios *et al.* [43]	Systematic review of the literature 42 cases of HCQ-induced cardiomyopathy	Median cumulative dose of HCQ 1542 g 18/42 patients died Evidence of left ventricular dysfunction reversibility upon discontinuation in 7/42 patients
Shulman *et al.* [48]	A questionnaire focused on assessing the implementation of the recommendations regarding HCQ-induced retinal toxicity, responded by 128 Israeli rheumatologists and ophthalmologists	A high proportion of the responders were not aware of the current recommendations, the appropriate diagnostic tools for screening, and the recognized risk factors
Costedoat-Chalumeau *et al.* [51▪]	Prospective international study evaluating nonadherence in 305 SLE patients with flares using HCQ blood levels and a self-administered questionnaire	137/302 patients were nonadherent by at least one criterion and 40/302 by both methods 43% of the nonadherent patients by blood levels would have been classified as adherent based on the questionnaire
Al-Rawi *et al.* [52]	Assessment of the inter and intra-patient variability of HCQ blood levels	The authors found a median 27% variation in the HCQ blood levels within an individual in a period of 12 h
Ugarte *et al.* [54]	Retrospective study assessing the clinical efficacy of HCQ and mepacrine combination in refractory skin and/or joint involvement in 46 SLE patients	91% patients showed complete or partial response. A significant reduction of SLEDAI and CLASI scores and prednisone dose was achieved. Prednisone could be discontinued in 20% of patients

CI, confidence interval; CLASI, cutaneous lupus erythematosus disease area and severity index; HCQ, hydroxychloroquine; IUGR, intrauterine growth retardation; SDI, SLICC damage index; SLEDAI, systemic lupus erythematosus disease activity index; SLE, systemic lupus erythematosus.

other hand, no protection was found for other manifestations of neonatal lupus [42▪].

Antimalarial toxicity

A systematic review by Tselios *et al.* [43] compiled 42 cases of antimalarial cardiotoxicity. The mean duration of therapy was 12.7 years with median cumulative doses of 1050 g of chloroquine and 1542 g of hydroxychloroquine. The most frequent clinical presentation was congestive heart failure, followed by syncope. The ECG findings were mainly conduction abnormalities, whereas left ventricular

Table 3. Current recommendations for the screening of HCQ retinopathy

A daily HCQ dose of 4–5 mg/kg (real weight) is associated with less than 1% risk in the first 5 years and <2% up to 10 years. The risk rises to 20% after 20 years of use
The recommended screening schedule includes a baseline visit, with annual screening starting 5 years later, provided there is an absence of associated risk factors
The recommended diagnostic techniques for the follow-up are the automated visual fields and SD-OCT
Associated risk factors for toxicity:
HCQ daily dose >5 mg/kg real weight
HCQ duration of use >5 years
Impaired renal function
Tamoxifen use
Underlying macular disease

HCQ, hydroxychloroquine; SD-OCT, spectral domain optical coherence tomography.
Modified from [46▪▪].

hypertrophy and impaired systolic function accounted for the most usual echocardiographic findings. Of the 42 patients, 18 died, two required heart transplantation and 13 a permanent pacemaker. Reversibility upon discontinuation was seen in seven patients. Given the high mortality rate, an early recognition of this complication is crucial.

Maculopathy is the most feared side-effect of antimalarials. In 2010, a study of nearly 4000 SLE and rheumatoid arthritis exposed patients associated the risk of retinal toxicity with the time of exposure, with a 4.5 to 5-fold increase among those treated for more than 7 years, and with the cumulative dose more than 1 g [44]. The 2011 recommendations on screening for antimalarial retinopathy of the American College of Ophthalmology recognized that the risk was higher over 5 years of use and with a cumulative dose of hydroxychloroquine more than 1000 g. The authors recommended the inclusion of new diagnostic techniques, such as automated visual fields and spectrum domain-optic coherence tomography, with higher sensitivity for subclinical toxicity [45]. The recommendations have been recently updated in 2016 [46▪▪], the main points are summarized in Table 3. Of note, doses of hydroxychloroquine more than 5 mg/kg/day have been identified as the main predictor of maculopathy [46▪▪].

Attention should be paid to the fact that retinal toxicity is nonreversible. Therefore, a prompt diagnosis is essential. However, the authors make it clear that, taking into account the wide beneficial effects of hydroxychloroquine in SLE, only patients with confirmed retinal toxicity should discontinue the drug. If needed, they recommend repeating the exploration within a few weeks and using additional techniques such as multifocal electroretinography or fundus autofluorescence [47]. Interestingly, a 2017 article revealed the striking unawareness of the current recommendations by rheumatologists and ophthalmologists, which could lead to unnecessary discontinuations of the drug in some cases and to a higher risk of harm when the physician is not aware of the risk factors for retinal toxicity [48].

Assessing adherence measuring blood hydroxychloroquine levels

Low blood levels of hydroxychloroquine were identified in the early 2000s as a predictor of SLE flares [49]. On the other hand, adapting the daily dose to measured blood levels did not result in a better control of SLE activity in a RCT [50]. Being a drug with a prolonged half-life, measuring hydroxychloroquine concentrations can be very useful to monitor adherence to medication, as a recent international prospective study has shown [51▪]. However, a study focusing on the inter and intra-patient variability of hydroxychloroquine whole blood levels found a median 27% fluctuation within an individual over 12 h, results that should be taken into account [52].

Mepacrine

Mepacrine was the first synthetic antimalarial drug. It was initially used in cutaneous lupus, being later replaced by chloroquine and hydroxychloroquine. In 2000, Toubi *et al.* [53] reported the efficacy and prednisone sparing effect of mepacrine addition to six patients with active SLE already treated with hydroxychloroquine. In a recent series from two Spanish longitudinal cohorts, mepacrine was added to baseline treatment in 46 patients with refractory skin and/or articular disease [54]. All patients were on hydroxychloroquine, along with different combinations of drugs such as prednisone, immunosuppressives, and retinoids, even belimumab in two patients. Within 1 year of follow-up, 91% of patients achieved clinical response, with significant reductions in SLEDAI and Cutaneous Lupus Erythematosus Disease Area and Severity Index scores. Mild side-effects were seen in only three patients. Of note, smoking patients were the most likely to achieve complete response. It must be remarked that mepacrine is free from ocular toxicity [55].

The main studies on antimalarials are summarized in Table 3.

CONCLUSION

Universal therapy with hydroxychloroquine should be aimed in patients with SLE without

contraindications. Consistent evidence supports the effect of hydroxychloroquine in improving survival in a dose-dependent manner in all ethnic groups of patients with lupus. Additional effects on thrombosis, damage, infections, and disease activity also contribute to improve not only the quantity but also the quality of life. In pregnant women with anti-Ro antibodies, whether or not diagnosed with a connective tissue disease, hydroxychloroquine therapy may contribute to decrease the risk of cardiac (but not other forms) of neonatal lupus. Doses more than 5 mg/kg/day should not be given, to minimize the risk of macular toxicity. Regular eye screening following the guidelines of the American College of Ophthalmology should be warranted for all patients.

One of the beneficial effects of antimalarials is the steroid-sparing effect. In fact, it is now agreed that every effort should be made to reduce the dose of glucocorticoids. Maintenance doses more than 5 mg/day should never be attained. Mild flares can be managed with transient increases of prednisone up to 10–15 mg/day with rapid reduction. In moderate-severe flares, pulse methyl-prednisolone 125, 250, or 500 mg/day for three consecutive days are more effective and much less toxic than increasing oral prednisone to 0.5–1 mg/kg/day. Rapid reduction from doses no higher than 30 mg/day to 5–2.5 mg/day should be accomplished within few weeks. Immunosuppressive therapy should be started early in severe forms of the disease, and whenever prednisone cannot be reduced to 5 mg/day or less. Adding-on mepacrine is a good and well tolerated option for patients with refractory cutaneous and/or articular activity and the alternative to hydroxychloroquine in the infrequent event of retinal toxicity.

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REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. ■ Gordon C, Amissah-Arthur M-B, Gayed M, *et al.*, British Society for Rheumatology Standards, Audit and Guidelines Working Group. The British Society for Rheumatology guideline for the management of systemic lupus erythematosus in adults. Rheumatology (Oxford) 2018; 57:e1–e45.

Recent exhaustive, evidence-based recommendations for the management of SLE.

2. ■ Singh JA, Hossain A, Kotb A, Wells G. Risk of serious infections with immunosuppressive drugs and glucocorticoids for lupus nephritis: a systematic review and network meta-analysis. BMC Med 2016; 14:137.

Systematic review showing that high-dose glucocorticoids increase the risk of infection more than any other immunosuppressive drug.

3. Herrinton LJ, Liu L, Goldfien R, *et al.* Risk of serious infection for patients with systemic lupus erythematosus starting glucocorticoids with or without antimalarials. J Rheumatol 2016; 43:1503–1509.

4. ■■ Rúa-Figueroa I, López-Longo J, Galindo-Izquierdo M, *et al.* Incidence, associated factors and clinical impact of severe infections in a large, multicentric cohort of patients with systemic lupus erythematosus. Semin Arthritis Rheum 2017; 47:38–45.

A large retrospective cohort study showing that prednisone greater than or equal to 10 mg/day increase the risk for severe and lethal infections, with a time-dependent protective effect of hydroxychloroquine.

5. Zonana-Nacach A, Barr SG, Magder LS, Petri M. Damage in systemic lupus erythematosus and its association with corticosteroids. Arthritis Rheum 2000; 43:1801–1808.

6. Ruiz-Arruza I, Ugarte A, Cabezas-Rodriguez I, *et al.* Glucocorticoids and irreversible damage in patients with systemic lupus erythematosus. Rheumatology (Oxford) 2014; 53:1470–1476.

7. Al Sawah S, Zhang X, Zhu B, *et al.* Effect of corticosteroid use by dose on the risk of developing organ damage over time in systemic lupus erythematosus-the Hopkins Lupus Cohort. Lupus Sci Med 2015; 2:e000066.

8. ■ Apostolopoulos D, Kandane-Rathnayake R, Raghunath S, *et al.* Independent association of glucocorticoids with damage accrual in SLE. Lupus Sci Med 2016; 3:e000157.

A cohort study showing that the risk of glucocorticoid-related damage could be increased even at low doses of prednisone.

9. Tarr T, Papp G, Nagy N, *et al.* Chronic high-dose glucocorticoid therapy triggers the development of chronic organ damage and worsens disease outcome in systemic lupus erythematosus. Clin Rheumatol 2017; 36:327–333.

10. ■■ Mok CC, Tse SM, Chan KL, Ho LY. Effect of immunosuppressive therapies on survival of systemic lupus erythematosus: a propensity score analysis of a longitudinal cohort. Lupus 2018; 27:722–727.

First study in showing an increased mortality risk of patients treated with high-dose prednisone. A reduced mortality with hydroxychloroquine was also confirmed in this study.

11. Parker B, Urowitz MB, Gladman DD, *et al.* Impact of early disease factors on metabolic syndrome in systemic lupus erythematosus: data from an international inception cohort. Ann Rheum Dis 2015; 74: 1530–1536.

12. Ruiz-Irastorza G, García M, Espinosa G, *et al.* Patterns of drug therapy in newly diagnosed Spanish patients with systemic lupus erythematosus. Clin Exp Rheumatol 2016; 34:466–472.

13. ■ Ruiz-Irastorza G, Garcia M, Espinosa G, *et al.* First month prednisone dose predicts prednisone burden during the following 11 months: an observational study from the RELES cohort. Lupus Sci Med 2016; 3:e000153.

In this multicentric prospective study, initial prednisone doses predicted cumulative doses over the following 11 months.

14. Zeher M, Doria A, Lan J, *et al.* Efficacy and safety of enteric-coated mycophenolate sodium in combination with two glucocorticoid regimens for the treatment of active lupus nephritis. Lupus 2011; 20: 1484–1493.

15. Fischer-Betz R, Chehab G, Sander O, *et al.* Renal outcome in patients with lupus nephritis using a steroid-free regimen of monthly intravenous cyclophosphamide: a prospective observational study. J Rheumatol 2012; 39: 2111–2117.

16. Ruiz-Irastorza G, Danza A, Perales I, *et al.* Prednisone in lupus nephritis: how much is enough? Autoimmun Rev 2014; 13:206–214.

17. ■ Ruiz-Irastorza G, Ugarte A, Saint-Pastou Terrier C, *et al.* Repeated pulses of methyl-prednisolone with reduced doses of prednisone improve the outcome of class III, IV and V lupus nephritis: an observational comparative study of the Lupus-Cruces and lupus-Bordeaux cohorts. Autoimmun Rev 2017; 16:826–832.

Efficacy and safety of repeated methyl-prednisolone pulses in lupus nephritis response rates and in prednisone sparing.

18. Condon MB, Ashby D, Pepper RJ, *et al.* Prospective observational single-centre cohort study to evaluate the effectiveness of treating lupus nephritis with rituximab and mycophenolate mofetil but no oral steroids. Ann Rheum Dis 2013; 72:1280–1286.

19. Lightstone L, Doria A, Wilson H, *et al.* Can we manage lupus nephritis without chronic corticosteroids administration? Autoimmun Rev 2018; 17:4–10.

20. Ruiz-Arruza I, Barbosa C, Ugarte A, Ruiz-Irastorza G. Comparison of high versus low-medium prednisone doses for the treatment of systemic lupus erythematosus patients with high activity at diagnosis. Autoimmun Rev 2015; 14:875–879.

21. Danza A, Borgia I, Narvaez J, *et al.* Intravenous pulses of methylprednisolone to treat flares of immune-mediated diseases. How much, how long? Lupus 2018; 27:1177–1184.

22. Parker BJ, Bruce IN. High dose methylprednisolone therapy for the treatment of severe systemic lupus erythematosus. Lupus 2007; 16:387–393.

23. Ruiz-Arruza I, Lozano J, Cabezas-Rodriguez I, *et al.* Restrictive use of oral
■ glucocorticoids in systemic lupus erythematosus and prevention of damage without worsening long-term disease control: an observational study. Arthritis Care Res 2018; 70:582–591.

First study in showing the long-term reduction of side-effects with similar SLE control in patients treated with a reduced-glucocorticoid schedule.

24. Ruiz-Irastorza G, Ramos-Casals M, Brito-Zeron P, Khamashta MA. Clinical efficacy and side effects of antimalarials in systemic lupus erythematosus: a systematic review. Ann Rheum Dis 2010; 69:20–28.

25. Pakchotanon R, Gladman DD, Su J, Urowitz MB. More consistent antimalarial
■ intake in first 5 years of disease is associated with better prognosis in patients with systemic lupus erythematosus. J Rheumatol 2018; 45:90–94.

Long-term hydroxychloroquine use reduces activity and damage accrual.

26. Sisó A, Ramos-Casals M, Bové A, *et al.* Previous antimalarial therapy in patients diagnosed with lupus nephritis: influence on outcomes and survival. Lupus 2008; 17:281–288.

27. Bultink IE, Hamann D, Seelen MA, *et al.* Deficiency of functional mannose-binding lectin is not associated with infections in patients with systemic lupus erythematosus. Arthritis Res Ther 2006; 8:R183.

28. Ruiz-Irastorza G, Olivares N, Ruiz-Arruza I, *et al.* Predictors of major infections in systemic lupus erythematosus. Arthritis Res Ther 2009; 11:R109.

29. Ruiz-Irastorza G, Ugarte A, Egurbide MV, *et al.* Antimalarials may influence the risk of malignancy in systemic lupus erythematosus. Ann Rheum Dis 2007; 66:815–817.

30. Fardet L, Nazareth I, Petersen I. Effects of chronic exposure of hydroxychloroquine/chloroquine on the risk of cancer, metastasis, and death: a population-based cohort study on patients with connective tissue diseases. Clin Epidemiol 2017; 9:545–554.

31. Ruiz-Irastorza G, Egurbide MV, Pijoan JI, *et al.* Effect of antimalarials on thrombosis and survival in patients with systemic lupus erythematosus. Lupus 2006; 15:577–583.

32. Alarcón G, McGwin G, Bertoli AM, *et al.* Effect of hydroxychloroquine on the survival of patients with systemic lupus erythematosus: data from LUMINA, a multiethnic US cohort (LUMINA L). Ann Rheum Dis 2007; 66:1168–1172.

33. Shinjo SK, Bonfa E, Wojdyla D, *et al.* Antimalarial treatment may have a time-dependent effect on lupus survival: data from a multinational Latin American inception cohort. Arthritis Rheum 2010; 62:855–862.

34. Zheng ZH, Zhang LJ, Liu WX, *et al.* Predictors of survival in Chinese patients with lupus nephritis. Lupus 2012; 21:1049–1056.

35. Götestam Skorpen CG, Hoeltzenbein M, Tincani A, *et al.* The EULAR points to consider for use of antirheumatic drugs before pregnancy, and during pregnancy and lactation. Ann Rheum Dis 2016; 75:795–810.

36. Flint J, Panchal S, Hurrell A, *et al.* BSR and BHPR guideline on prescribing drugs in pregnancy and breastfeeding-part I: standard and biologic disease modifying antirheumatic drugs and corticosteroids. Rheumatology (Oxford) 2016; 55:1693–1697.

37. Bermas BL, Kim SC, Huybrechts K, *et al.* Trends in use of hydroxychloroquine during pregnancy in systemic lupus erythematosus patients from 2001 to 2015. Lupus 2018; 961203317749046. doi: 10.1177/0961203317749046. [Epub ahead of print]

38. Leroux M, Desveaux C, Parcevaux M, *et al.* Impact of hydroxychloroquine on preterm delivery and intrauterine growth restriction in pregnant women with systemic lupus erythematosus: a descriptive cohort study. Lupus 2015; 24:1384–1391.

39. Kroese SJ, de Hair MJ, Limper M, *et al.* Hydroxychloroquine use in lupus patients during pregnancy is associated with longer pregnancy duration in preterm births. J Immunol Res 2017; 2017:2810202.

40. Izmirly PM, Kim MY, Llanos C, *et al.* Evaluation of the risk of anti-SSA/Ro-SSB/La antibody associated cardiac manifestations of neonatal lupus in fetuses of SLE mothers exposed to hydroxychloroquine. Ann Rheum Dis 2010; 69:1827–1830.

41. Izmirly PM, Costedoat-Chalumeau N, Pisoni CN. Maternal use of hydroxychloroquine is associated with a reduced risk of recurrent anti-SSA/Ro-antibody– associated cardiac manifestations of neonatal lupus. Circulation 2012; 126:76–82.

42. Barsalou J, Jaeggi E, Laskin CA, *et al.* Prenatal exposure to antimalarials
■ decreases the risk of cardiac but not noncardiac neonatal lupus: a single-centre cohort study. Rheumatology (Oxford) 2017; 56:1552–1559.

Hydroxychloroquine given to anti-Ro positive pregnant mothers protects the baby from congenital heart block, but not from other types of neonatal lupus.

43. Tselios K, Deeb M, Gladman DD, *et al.* Antimalarial-induced cardiomyopathy: a systematic review of the literature. Lupus 2018; 27:591–599.

44. Wolfe F, Marmor MF. Rates and predictors of hydroxychloroquine retinal toxicity in patients with rheumatoid arthritis and systemic lupus erythematosus. Arthritis Care Res (Hoboken) 2010; 62:775–784.

45. Marmor MF, Kellner U, Lai T, Lyons JS; American Academy of Ophthalmology. Revised recommendations on screening for chloroquine and hydroxychloroquine retinopathy. Ophthalmology 2011; 118:415–422.

46. Marmor MF, Kellner U, Lai T, *et al.,* American Academy of Ophthalmology.
■■ Recommendations on screening for chloroquine and hydroxychloroquine retinopathy (2016 revision). Ophthalmology 2016; 123:1386–1394.

Identification of factors associated to antimalarial maculopathy and most recent recommendations for preventive eye screening.

47. Marmor MF. Modern management of antimalarial usage and retinopathy. J Curr Ophthalmol 2017; 29:143–144.

48. Shulman S, Wollman J, Brikman S, *et al.* Implementation of recommendations for the screening of hydroxychloroquine retinopathy: poor adherence of rheumatologists and ophthalmologists. Lupus 2017; 26:277–281.

49. Costedoat-Chalumeau N, Amoura Z, Hulot JS, *et al.* Low blood concentration of hydroxychloroquine is a marker for and predictor of disease exacerbations in patients with systemic lupus erythematosus. Arthritis Rheum 2006; 54:3284–3290.

50. Costedoat-Chalumeau N, Galicier L, Aumaître O, *et al.,* Group PLUS. Hydroxychloroquine in systemic lupus erythematosus: results of a French multicentre controlled trial (PLUS Study). Ann Rheum Dis 2013; 72:1786–1792.

51. Costedoat-Chalumeau N, Houssiau F, Izmirly P, *et al.* A prospective interna-
■ tional study on adherence to treatment in 305 patients with flaring SLE: assessment by drug levels and self-administered questionnaires. Clin Pharmacol Ther 2017; doi: 10.1002/cpt.885. [Epub ahead of print]

A prospective study that underlines the difficulty in assessing nonadherence in the clinical practice and the utility of hydroxychloroquine blood levels in this setting.

52. Al-Rawi H, Maggot SJ, Williams FM, Wahie S. Steady-state pharmacokinetics of hydroxychloroquine in patients with cutaneous lupus erythematosus. Lupus 2018; 27:847–852.

53. Toubi E, Kessel A, Rosner I, *et al.* The benefit of combining hydroxychloroquine with quinacrine in the treatment of SLE patients. Lupus 2000; 9:92–95.

54. Ugarte A, Porta S, Rios R, *et al.* Combined mepacrine-hydroxychloroquine treatment in patients with systemic lupus erythematosus and refractory cutaneous and articular activity. Lupus 2018; 961203318768877. doi: 10.1177/0961203318768877. [Epub ahead of print]

55. Wallace DJ, Gudsoorkar VS, Weisman MH, Venuturupalli SR. New insights into mechanisms of therapeutic effects of antimalarial agents in SLE. Nat Rev Rheumatol 2012; 8:522–533.

Drug-induced lupus erythematosus: an update on drugs and mechanisms

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Purpose of review
Rapid introduction of newly developed drugs in the absence of clear understanding of the pathophysiologic mechanisms behind drug-induced lupus erythematosus (DILE) can sometimes make DILE difficult to recognize in clinical practice. The purpose of this review is to summarize drugs most recently reported to be involved in DILE and discuss the current landscape of diverse mechanisms involved.

Recent findings
A large number of proton pump inhibitor (PPI)-induced subacute cutaneous lupus erythematosus cases have been reported, suggesting a shift over time in the spectrum of drugs implicated in DILE. Twenty-two articles comprising 29 DILE case reports published within the last 2 years are summarized in this review, including 12 (41.4%) systemic DILE. Antitumor necrosis factor (anti-TNF) drugs were the most frequently (41.7%) reported to introduce systemic DILE in these cases. Chemotherapeutic drugs were the most common drug class (54.5%) involved in subacute cutaneous lupus erythematosus, with an observed higher incidence in female patients. Enhanced neutrophil extracellular trap (NET) formation induced by procainamide and hydralazine could be a new mechanism contributing to the pathogenesis of DILE.

Summary
The list of drugs implicated in triggering DILE is expanding as new drugs with novel mechanisms of action are being developed. It is important to recognize culprit drugs that may induce lupus erythematosus, as discontinuation usually results in improvement of drug-induced manifestations. Characterizing the mechanisms involved might help better understand the cause of idiopathic autoimmunity.

Keywords
autoimmunity, drug-induced lupus erythematosus, drugs, mechanisms

INTRODUCTION

Drug-induced lupus erythematosus (DILE) is a lupus-like autoimmune disorder, which usually occurs with chronic exposure to certain drugs (months to years) and resolves after cessation of the culprit medication. The recognition of DILE is usually attributed to Hoffman, who first reported lupus-like symptoms following sulfadiazine treatment in 1945 [1]. Later in 1985, hydrochlorothiazide was reported to induce subacute cutaneous lupus erythematosus (SCLE), which introduced the concept of drug-induced SCLE [2]. To date, over 100 drugs from more than 10 drug categories have been implicated in DILE [3,4], but only procainamide and hydralazine are regarded as two high-risk drugs with 20% [5] and 5–8% [6] risk of developing DILE, respectively. Fewer cases of DILE induced by these two drugs are being reported as their use in clinical practice declines, yet cases of DILE triggered by newer oncology drugs and biological modulators in patients with neoplastic and autoimmune diseases are expanding recently [7].

Similar to idiopathic lupus, DILE can be classified into three major forms: systemic DILE, drug-induced subacute cutaneous lupus erythematosus (DISCLE) and chronic cutaneous DILE. The latter two forms could also be defined as drug-induced cutaneous lupus erythematosus (DICLE). Systemic DILE is

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KEY POINTS

- New DILE cases published within the last 2-year period in PubMed database are summarized in this review.
- DISCLE associated with PPI and chemotherapeutic drugs deserves more attention owing to increasing numbers of case reports.
- Enhanced NET formation could be a new mechanism contributing to the pathogenesis of DILE.

characterized by mild arthralgia, myalgia, serositis and constitutional symptoms [8]. DISCLE is the most common subtype with predominant skin involvement and is more frequently seen in older female patients [9]. Chronic cutaneous DILE is rare and often associated with fluorouracil compounds [10]. Discoid skin lesions are more distinctly found in chronic cutaneous DILE than the other two subtypes. Patients exposed to different drugs would develop different forms of DILE, whose clinical manifestations and serological characteristics can extremely vary.

Guidelines proposed by Borchers *et al.* in 2007 [11] and further advanced by Xiao and Chang [12], could aid to confirm a DILE diagnosis to some extent. Notably, diagnosis of DILE must be made after overall examination, medication and history review, and comprehensive evaluation of the disease during the time course following causative drug exposure and withdrawal.

Recognizing the offending drug linked to DILE is the first and utmost step in DILE management. However, DILE can be easily overlooked in clinical practice given the following factors: Delayed insidious association between drug exposure and symptom onset; Rapid introduction of new drugs developed with limitations in predicting their long-term effect during treatment; and Lack of understanding of the pathophysiologic mechanisms in DILE. This review will summarize the spectrum of drugs linked to DILE and shape a current landscape of diverse mechanisms behind DILE, with an emphasis on updating drugs and mechanisms reported within the last 2 years.

DRUGS IMPLICATED IN DRUG-INDUCED LUPUS ERYTHEMATOSUS

Drugs associated with DILE have various chemical structures such as aromatic amines, hydrazine and sulfhydryl groups, indicating that no single unifying chemical configuration accounts for DILE [13]. Meanwhile, drugs that induce DILE possess distinguishable distribution patterns in different forms of DILE, most of which are well summarized in a wealth of literature [14–17].

In general, drugs involved in systemic DILE are identified in four categories, which are drugs definitely, probably, possibly and recently reported to induce DILE [15,16], or they can also be grouped into high, moderate, low or very low risk categories by the risk levels. The most common drugs causing systemic DILE are hydralazine (high risk), procainamide (high risk), isoniazid (moderate risk), minocycline (very low risk) and more recently reported tumour necrosis factor-α (TNF-α) inhibitors (very low risk) [4,11,18]. Drugs most likely to trigger SCLE include hydrochlorothiazide [2], calcium channel blockers and angiotensin-converting enzyme inhibitors [16]. Drugs such as proton-pump inhibitors (PPIs) [19,20▪,21▪], terbinafine [22–24], immunomodulators (leflunomide [25,26], TNF-α inhibitors [27]) and chemotherapeutic agents [28–30] can also induce SCLE. A population-based matched case–control study performed by Gronhagen *et al.* [31] confirmed association between certain suspected drugs and SCLE, with significantly increased odds ratio (OR) found for terbinafine (OR 52.9), TNF-α inhibitors (OR 8.0), antiepileptics (OR 3.4) and PPIs (OR 2.9). Chronic cutaneous DILE has usually been triggered by fluorouracil compounds or their modern derivatives such as capecitabine [32,33].

Systemic DILE induced by TNF-α inhibitors is well described in the literature and received widespread attention [17,34–37], while PPI-induced SCLE is worth more awareness in clinical practice, as PPI-associated SCLE cases have been increasingly reported in a large scale. PPIs, often prescribed to treat peptic ulcer and gastroesophageal reflux disease (GERD), reduce gastric acid secretion by inhibiting the K+/H+ ATPase pump in gastric parietal cells [38]. In a case–control study reported by Gronhagen *et al.* [31], 66 out of 234 SCLE cases from Sweden were found to be associated with PPIs. Four years later, in 2014, 24 patients with PPI-induced SCLE were identified in a retrospective medical chart review of 429 CLE patients from Denmark [39]. Most recently, a study by Michaelis *et al.* [20▪] revealed that, from August 2009 to May 2016 (case–control study from Sweden by Gronhangen *et al.* [31] was excluded), cases associated with PPIs were increased by 34.1% compared with all other medications, whereas reports in antihypertensive and antifungal medications decreased by 28.9 and 22.4%, respectively [20▪]. A recent retrospective chart review presenting 88 cases with DISCLE identified PPIs are one of the most common culprit drug classes involved [21▪]. Future efforts to investigate the mechanisms behind PPI-associated SCLE, which are currently unclear, are warranted.

SYSTEMATIC REVIEW OF DRUG-INDUCED LUPUS ERYTHEMATOSUS REPORTED IN THE LAST 2 YEARS

To investigate if there has been a shift in drugs implicated in triggering DILE within the last 2 years, we conducted a literature review. We searched PubMed for clinical case reports of DILE published from 1 January 2016 to 10 May 2018. Searches were performed with the phrase 'drug induced lupus'. Only case reports in English full text were included. Impact factors of publishing journals were ignored. Large case series of PPI-associated DISCLE in this timeframe [20▪,21▪] were discussed separately in this article, and thus were excluded in following literature analysis.

There were 29 cases of DILE reported in 22 articles (Table 1) [35▪,40–42,43▪–45▪,46,47,48▪,49–60], among which 12 (41.4%) cases were systemic DILE, 11 (37.9%) cases were DISCLE and six (20.7%) cases were DICLE without further differentiation into DISCLE or chronic cutaneous DILE. The 12 systemic DILE cases included nine female patients (75%) and three male patients (25%), with a mean age of 44 years (range 9–91). Anti-TNF-α drugs were the most frequently reported drugs to induce systemic DILE within the last 2 years (five cases; four were associated with infliximab and one with adalimumab). Of note, two systemic DILE cases respectively associated with infliximab and carbamazepine, occurred in paediatric population, which is less frequently seen in DILE, implying DILE should also be suspected in younger patients with long-term treatment of certain medications. All three cases of systemic DILE induced by hydralazine were with negative antinuclear antibody (ANA), as opposed to serologic findings of positive serum ANA in most hydralazine-induced lupus erythematosus patients, suggesting that diagnosis of hydralazine-induced lupus erythematosus shall not be ruled out if ANA was negative.

In 11 cases of DISCLE, there were 10 female patients and one male patient, with an average age of nearly 47.6 years (range 14–69, two patients without accurate age record). The highest drug class associated with DISCLE was chemotherapeutics, with six cases reported being induced by mitotane, gemcitabine, capecitabine, annastrozole, hydroxyurea and palbociclib. Mitotane, the antifibrotic drug prifenidone, and antiretroviral HIV therapy were newly identified as triggers of DISCLE, never described in previous DISCLE cases.

IgG treatment-induced cutaneous lupus erythematosus was reported in case series with DICLE in three female patients and three male patients (average age of 55 years, range 42–67).

MECHANISMS INVOLVED IN DRUG-INDUCED LUPUS ERYTHEMATOSUS

Despite that a variety of drugs within different classes and with different mechanisms of action have been associated with DILE, most studies exploring pathogenic mechanisms in DILE have been primarily focused on procainamide and hydralazine. Several mechanisms have been proposed, including genetic predisposition, drug biotransformation and epigenetic dysregulation in different immune cells. Mechanisms underlying the pathogenesis of DILE are summarized in Fig. 1.

Genetic predisposition

It is widely accepted that genetic susceptibility plays a role in development of DILE. Drugs such as procainamide, hydralazine and isoniazid contain a structure of aromatic amines or hydrazines, and are predominantly metabolized by acetylation utilizing N-acetyltransferase enzymes [13]. The majority of patients with procainamide or hydralazine-induced lupus erythematosus are found to be slow acetylators, who are more prone for autoantibodies accumulation after exposure to procainamide or hydralazine compared with fast acetylators [61–63]. Interestingly, the risk of developing DILE is about the same in patients with the same serum concentration of procainamide, regardless of the acetylator phenotype [64]. Unlike the findings in procainamide and hydralazine, isoniazid-implicated DILE seems to be less related to acetylator phenotype though isoniazid is also metabolized by acetylation [65,66]. In addition, associations between DILE occurrence and certain human leukocyte antigen (HLA), like HLA-DR2, HLA-DR3, class III C4A and C4B null complement alleles, have been suggested by some studies, but these findings were not always consistent [67–69]. The complement system might also play a role in the pathogenic mechanisms of DILE. Sim *et al.* [70,71] reported that hydralazine, penicillamine, isoniazid and metabolic products of procainamide could be potent inhibitors of the covalent binding reaction of complement component C4, which might inhibit the activation of complement component C3 in the classical complement pathway, hindering the clearance of immune complexes.

Drug biotransformation

Procainamide is oxidized by activated neutrophils resulting in the production of a toxic metabolite called procainamide hydroxylamine (PAHA). PAHA, together with myeloperoxidase (MPO) and reactive

Table 1. Summary of 29 case reports of DILE reported in the literature published on PubMed (January 2016–May 2018)

# Case	Sex/age (years)	Drug (doses)	Drug categories	DILE forms	Latency	Autoantibodies	Outcome symptom after drug removal	Ref.
1	M/39	Clozapine (25 mg/day)	Antipsychotics	Systemic DILE	8 days	ANA+	Remission	[40]
2	M/91	Minocycline (200 mg/day)	Antibiotics	Systemic DILE	2 years	ANA+, dsDNA+	Improvement	[41]
3	F/62	Trimethoprim/sulfamethoxazole	Antibiotics	Systemic DILE	1 week	ANA+, histone+, dsDNA-, SSA-, SSB-	Remission	[42]
4	M/21	Hydralazine (50 mg TID)	Antihypersentsitives	Systemic DILE	2 months	histone+, ANA-	Remission	[43▪]
5	F/36	Hydralazine (50 mg TID)	Antihypersentsitives	Systemic DILE	18 months	histone+, dsDNA-, ANA-, SSA-, SSB-,	Improvement	[44▪]
6	F/35	Hydralazine (10 mg, q8h)	Antihypersentsitives	Systemic DILE	4 weeks	histone+, ANA-, dsDNA-	Improvement	[45▪]
7	F/14	Infliximab	Immunomodulators: TNF-α inhibitors	Systemic DILE	7 months	ANA+, dsDNA+	Remission	[35▪]
8	F/64	Infliximab	Immunomodulators: TNF-α inhibitors	Systemic DILE	11 months	histone+, dsDNA-ANA+,	Remission	[35▪]
9	F/67	Infliximab	Immunomodulators: TNF-α inhibitors	Systemic DILE	3 months	ANA+, antidsDNA	Remission	[35▪]
10	F/48	Infliximab	Immunomodulators: TNF-α inhibitors	Systemic DILE	3 years	ANA+, dsDNA+	Remission	[46]
11	F/42	Adalimumab	Immunomodulators: TNF-α inhibitors	Systemic DILE	2 years	ANA+	Improvement	[47]
12	F/9	Carbamazepine (200 mg/day)	Anticonvulsives	Systemic DILE	3 years	ANA+, histone+	Remission	[48▪]
13	F/ in 60s	Mitotane (300 mg, TID)	Chemotherapeutics	DISCLE	1 month	ANA-, SSA-, SSB-	Remission	[49]
14	M/42	Interferon alpha-2a	Immunomodulators	DISCLE	< 24 weeks	ANA-, histone-, ds-DNA-,	n.d	[50]
15	F/63	Gemcitabine	Chemotherapeutics	DISCLE	2 weeks	SSA-, SSB-, histone-	Remission	[51]
16	F/50	Leflunomide (20 mg/day)	Immunomodulators	DISCLE	3 years	ANA+, histone ++, SSA+, SSB-, dsDNA-	Improvement	[52]
17	F/67	Capecitabine	Chemotherapeutics	DISCLE	6 weeks	ANA+, SSA+	Improvement	[53]
18	F/54	Pirfenidone	Novel Antifibrosis drug	DISCLE	8 weeks	dsDNA+, ANA histone-, SSA-, SSB-	Improvement	[54]
19	F/ 69	Anastrozole	Chemotherapeutics	DISCLE	16 months	SSA+, ANA-, SSB-	Improvement	[55]
20	F/ 14	Hydroxyurea (1500 mg/day)	Chemotherapeutics	DISCLE	5 years	ANA+, histone+, SSA-, SSB-	Improvement	[56]
21	F/ 50s	Palbociclib	Chemotherapeutics agents	DISCLE	2 months	ANA+, dsDNA-, SSA-, SSB-	Remission	[57]
22	F/ 35	Emtricitabine, rilpivirine, tenofovir disoproxil fumarate (combination)	Antiretroviral Therapy	DISCLE	3 years	ANA+, dsDNA+, histone+	Remission	[58]
23	F/ 34	Terbinafine (topical cream)	Antifungal drugs	DISCLE	A number of years	ANA+, SSA+	Remission	[59]
24	F/62	IVIg (1.3 g/kg/month)	Immunomodulators	DICLE	6 weeks	n.d	Improvement	[60]
25	F/45	IVIg (1.2 g/kg/month)	Immunomodulators	DICLE	6 months	n.d	Improvement	[60]
26	M/42	IVIg (1.3 g/kg/month)	Immunomodulators	DICLE	2 weeks	SSA+	Improvement	[60]
27	F/67	IVIg (1 g/kg/month)	Immunomodulators	DICLE	<3 weeks	ENA+	Improvement	[60]
28	M/54	SCIg (1.8 g/kg/month)	Immunomodulators	DICLE	22 months	ANA+, ENA-	Remission	[60]
29	M/60	IVIg (0.8 g/kg/month)	Immunomodulators	DICLE	6 months	ENA+	Improvement	[60]

ANA, antinuclear antibodies; dsDNA, antidouble-stranded DNA; ENA, extractable nuclear antigen antibodies; histone, antihistone antibodies; IVIg, intravenous immunoglobulin; n.d, not determined; SCIg, subcutaneous immunoglobulin; SSA, anti-Ro/SSA; SSB, anti-La/SSB.

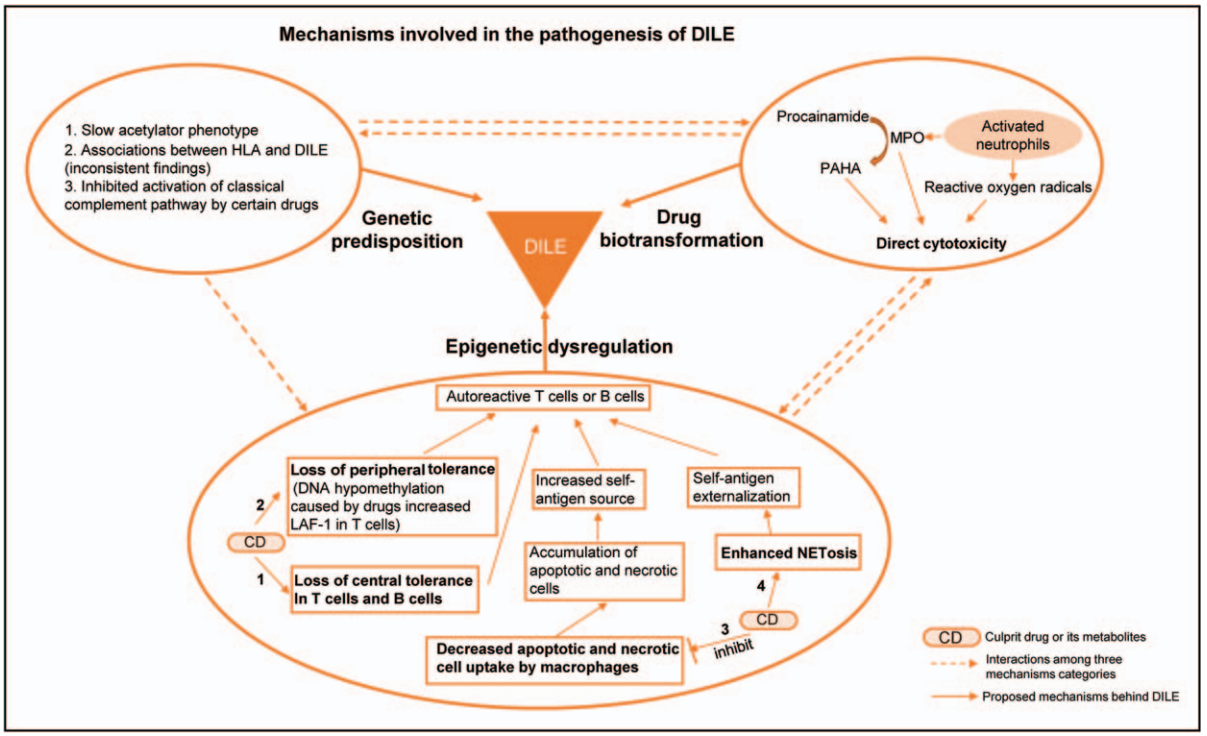

FIGURE 1. Mechanisms involved in the pathogenesis of drug-induced lupus erythematosus. Genetic predisposition, drug biotransformation and epigenetic dysregulation are three important components of current proposed pathogenic mechanisms of DILE. Instead of working independently, these factors are likely to interact with each other to cause DILE. Genetic predisposition: Studies revealing genetic predisposition could be summarized in three main aspects, listed in the left upper circle. Biotransformation: Procainamide undergoes neutrophil-mediated oxidative metabolism to produce procainamide hydroxylamine (PAHA). PAHA, myeloperoxidase (MPO), and reactive oxygen species contribute to direct cytotoxicity. Epigenetic dysregulation: Drugs and some drug metabolites exert epigenetic dysregulation on T cells and B cells (1,2), macrophages (3) and neutrophils (4), which eventually leads to autoreactive T cell and B cell generation, triggering DILE.

oxygen species released during oxidative metabolism of procainamide, contribute to the cytotoxicity [72–74]. In addition, autoantibodies against myeloperoxidase were found in the serum of DILE patients, which indirectly supported a role of myeloperoxidase-mediated metabolism in the development of DILE [75]. Other drugs, including hydralazine, quinidine, phenytoin, sulfone, penicillamine, chlorpromazine and isoniazid, undergo the biotransformation similar to procainamide, which generates reactive metabolites triggering DILE. On the contrary, drugs in small molecules can bind to proteins, a process called haptenization, then stimulate immune responses [14].

Epigenetic dysregulation in adaptive immune cells and other mechanisms of autoreactivity

Biotransformed culprit drugs or their metabolites have been reported to alter epigenetic properties of immune cells then ultimately lead to DILE. In early epigenetic mechanism studies of DILE, several mechanisms involving T cells or B cells were put forward. Hydralazine and procainamide were shown to inhibit T cell DNA methylation [76]. More specifically, procainamide acts as a competitive DNA methyltransferase inhibitor, while hydralazine prevents induction of DNA methyltransferase by inhibiting ERK signalling pathway [77,78]. DNA hypomethylation in T cells results in increased lymphocyte function associated antigen 1 (LFA-1) expression, which consequently induces autoreactivity. Adoptive transfer of these autoreactive T cells into mice caused a lupus-like disease [79,80].

Other studies suggest that PAHA, a procainamide metabolite, interferes with T cell central tolerance, resulting in the production of autoreactive T cells possibly triggering autoimmunity [81,82]. Similarly, hydralazine is able to subvert B cell tolerance and contributes to the generation of pathogenic autoreactivity by disrupting receptor editing via inhibition of the ERK signalling pathway [83]. Quinidine and procainamide at therapeutic range concentrations were reported to inhibit uptake of apoptotic thymocytes by macrophages, which could

render these accumulated cells a source for uncontrolled uptake of self-antigens in certain settings [84].

Sontheimer *et al.* [85] discussed an evidence likely pertaining to the pathogenesis of DISCLE, pointing out that drugs involved in DISCLE are capable of causing photosensitivity further amplifying cutaneous immune responses that give rise to an increase in local type I interferon production and downstream molecules such as chemokine (C-X-C motif) ligand 9 (CXCL9).

Role of NETosis and the innate immune system

More recently, a role for NETosis, a unique mechanism of neutrophil cell death, has been described in DILE. Neutrophil extracellular traps (NETs) are web-like structure containing nuclear DNA and cytosolic proteins secreted by activated neutrophils after specific stimuli [86]. Autoantigen-rich nuclear material and granular proteins can be externalized during NETosis, which subsequently induces autoimmunity [87]. In 2018, Irizarry-Caro *et al*. [88▪▪] described that procainamide and hydralazine, known to induce lupus erythematosus, promote NET formation via triggering neutrophil muscarinic receptors and increasing intracellular calcium flux *in vitro,* respectively, demonstrating the contribution of innate immune responses in the development of DILE. Interestingly, it was also pointed out in the same article that minocycline and clozapine, another two drugs less commonly associated with DILE, do not induce NETosis. Additional future experiments both *in vitro* and *in vivo* are suggested to confirm and characterize this mechanism of drug-induced NETosis in DILE [89▪].

CONCLUSION

This article summarizes the current knowledge in DILE, with an emphasis on recent developments in the field. We performed a systematic review for new cases of DILE reported over the last 2 years to highlight the observed shift in DILE-implicated drugs over time, though publication bias is an obvious limitation. This analysis highlighted drugs recently described to trigger DILE and rare cases of DILE in paediatric patients. DILE associated with PPIs and anti-TNF therapies might be more commonly encountered in current rheumatology practices than less used drugs such as procainamide and hydralazine. We expect a plethora of DILE reports in the future with the increasing use and expanding targets of immunotherapy in cancer patients, including check-point inhibitors.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ▪ of special interest
- ▪▪ of outstanding interest

1. Hoffman BJ. Sensitivity to sulfadiazine resembling acute disseminated lupus erythematosus. Arch Dermatol Syphilol 1945; 51:190–192.
2. Reed BR, Huff JC, Jones SK, *et al.* Subacute cutaneous lupus erythematosus associated with hydrochlorothiazide therapy. Ann Intern Med 1985; 103: 49–51.
3. Chang C, Gershwin ME. Drug-induced lupus erythematosus: incidence, management and prevention. Drug Saf 2011; 34:357–374.
4. Vasoo S. Drug-induced lupus: an update. Lupus 2006; 15:757–761.
5. Rubin RL. Drug-induced lupus. Toxicology 2005; 209:135–147.
6. Yokogawa N, Vivino FB. Hydralazine-induced autoimmune disease: comparison to idiopathic lupus and ANCA-positive vasculitis. Mod Rheumatol 2009; 19:338–347.
7. Bukhari M. Drug-induced rheumatic diseases: a review of published case reports from the last two years. Curr Opin Rheumatol 2012; 24:182–186.
8. Aylward PE, Tonkin AM, Bune A. Cardiac tamponade in hydrallazine-induced systemic lupus erythematosus. Aust N Z J Med 1982; 12:546–547.
9. Lowe GC, Henderson CL, Grau RH, *et al.* A systematic review of drug-induced subacute cutaneous lupus erythematosus. Br J Dermatol 2011; 164:465–472.
10. Marzano AV, Vezzoli P, Crosti C. Drug-induced lupus: an update on its dermatologic aspects. Lupus 2009; 18:935–940.
11. Borchers AT, Keen CL, Gershwin ME. Drug-induced lupus. Ann N Y Acad Sci 2007; 1108:166–182.
12. Xiao X, Chang C. Diagnosis and classification of drug-induced autoimmunity (DIA). J Autoimmun 2014; 48-49:66–72.
13. Yung RL, Richardson BC. Chapter 22: drug-induced lupus mechanisms A2. In: Robert LG, editor. Systemic lupus erythematosus (fifth edition). San Diego, CA: Academic Press; 2011. pp. 385–403.
14. Chang C, Gershwin ME. Drugs and autoimmunity: a contemporary review and mechanistic approach. J Autoimmun 2010; 34:J266–J275.
15. Sarzi-Puttini P, Atzeni F, Capsoni F, *et al.* Drug-induced lupus erythematosus. Autoimmunity 2005; 38:507–518.
16. Pretel M, Marques L, Espana A. Drug-induced lupus erythematosus. Actas Dermosifiliogr 2014; 105:18–30.
17. Araujo-Fernandez S, Ahijon-Lana M, Isenberg DA. Drug-induced lupus: including antitumour necrosis factor and interferon induced. Lupus 2014; 23:545–553.
18. Aguirre Zamorano MA, Lopez Pedrera R, Cuadrado Lozano MJ. [Drug-induced lupus]. Med Clin (Barc) 2010; 135:124–129.
19. Bracke A, Nijsten T, Vandermaesen J, *et al.* Lansoprazole-induced subacute cutaneous lupus erythematosus: two cases. Acta Derm Venereol 2005; 85:353–354.
20. ▪ Michaelis TC, Sontheimer RD, Lowe GC. An update in drug-induced subacute cutaneous lupus erythematosus. Dermatol Online J 2017; 23:.
This study demonstrates that cases of PPI-associated DISCLE increased greatly during August 2009 to May 2016 by analysing drugs triggering DILE reported in PubMed database.
21. ▪ Laurinaviciene R, Sandholdt LH, Bygum A. Drug-induced cutaneous lupus erythematosus: 88 new cases. Eur J Dermatol 2017; 27:28–33.
This article shows a clear trend that PPI-induced DISCLE replaces the predominance of antyhypertsensive drug in DISCLE to become the most common culprit drug category with a large case series of patients diagnosed as CLE during 2004-2014.
22. McKay DA, Schofield OM, Benton EC. Terbinafine-induced subacute cutaneous lupus erythematosus. Acta Derm Venereol 2004; 84:472–474.

23. Lorentz K, Booken N, Goerdt S, Goebeler M. Subacute cutaneous lupus erythematosus induced by terbinafine: case report and review of literature. J Dtsch Dermatol Ges 2008; 6:823–827; 823-8.
24. Callen JP, Hughes AP, Kulp-Shorten C. Subacute cutaneous lupus erythematosus induced or exacerbated by terbinafine: a report of 5 cases. Arch Dermatol 2001; 137:1196–1198.
25. Suess A, Sticherling M. Leflunomide in subacute cutaneous lupus erythematosus: two sides of a coin. Int J Dermatol 2008; 47:83–86.
26. Marzano AV, Ramoni S, Del Papa N, *et al.* Leflunomide-induced subacute cutaneous lupus erythematosus with erythema multiforme-like lesions. Lupus 2008; 17:329–331.
27. Costa MF, Said NR, Zimmermann B. Drug-induced lupus due to antitumor necrosis factor alpha agents. Semin Arthritis Rheum 2008; 37:381–387.
28. Guhl G, Diaz-Ley B, García-García C, *et al.* Chemotherapy-induced subacute lupus erythematosus. Lupus 2009; 18:859–860.
29. Weger W, Kränke B, Gerger A, *et al.* Occurrence of subacute cutaneous lupus erythematosus after treatment with fluorouracil and capecitabine. J Am Acad Dermatol 2008; 59((2 Suppl 1)):S4–S6.
30. Chen M, Crowson AN, Woofter M, *et al.* Docetaxel (taxotere) induced subacute cutaneous lupus erythematosus: report of 4 cases. J Rheumatol 2004; 31:818–820.
31. Gronhagen CM, Fored CM, Linder M, *et al.* Subacute cutaneous lupus erythematosus and its association with drugs: a population-based matched case-control study of 234 patients in Sweden. Br J Dermatol 2012; 167: 296–305.
32. Yoshimasu T, Hiroi A, Uede K, Furukawa F. Discoid lupus erythematosus (DLE)-like lesion induced by uracil-tegafur (UFT). Eur J Dermatol 2001; 11:54–57.
33. Merlin F, Prochilo T, Kildani B, *et al.* Discoid lupus erythematosus (DLE)-like lesions induced by capecitabine. Int J Colorectal Dis 2008; 23:715–716.
34. Quaresma MV, Bernardes Filho F, Oliveira FB, *et al.* Anti-TNF-alpha and hydralazine drug-induced lupus. An Bras Dermatol 2015; 90((3 Suppl 1)): 125–129.
35. ■ Shovman O, Tamar S, Amital H, *et al.* Diverse patterns of anti-TNF-alpha-induced lupus: case series and review of the literature. Clin Rheumatol 2018; 37:563–568.

Three cases of anti-TNF-α induced lupus erythematosus presenting distinct clinical symptoms are described in this study, one of which occurred in a 14-year-old paediatric patient.

36. Wetter DA, Davis MD. Lupus-like syndrome attributable to antitumor necrosis factor alpha therapy in 14 patients during an 8-year period at Mayo Clinic. Mayo Clin Proc 2009; 84:979–984.
37. Almoallim H, Al-Ghamdi Y, Almaghrabi H, Alyasi O. Anti-tumor necrosis factor-alpha induced systemic lupus erythematosus(). Open Rheumatol J 2012; 6:315–319.
38. Sachs G, Shin JM, Howden CW. Review article: the clinical pharmacology of proton pump inhibitors. Aliment Pharmacol Ther 2006; 23(Suppl 2):2–8.
39. Sandholdt LH, Laurinaviciene R, Bygum A. Proton pump inhibitor-induced subacute cutaneous lupus erythematosus. Br J Dermatol 2014; 170: 342–351.
40. Buzina N, Eterovic M. Life-threatening lupus-like syndrome associated with clozapine. J Clin Psychopharmacol 2016; 36:532–534.
41. Starobin D, Guller V, Gurevich A, Tal S. Minocycline induced lupus with yellow colored chylous exudative pleural effusion. Respir Med Case Rep 2017; 22:113–116.
42. Jose A, Cramer AK, Davar K, Gutierrez G. A case of drug-induced lupus erythematosus secondary to trimethoprim/sulfamethoxazole presenting with pleural effusions and pericardial tamponade. Lupus 2017; 26:316–319.
43. ■ Zeitjian V, Mehdizadeh A. ANA-negative hydralazine-induced pericardial effusion. Case Rep Med 2017; 2017:3521541.

A case report that describes a patient with pericardial effusion caused by hydralazine-associated DILE with a negative ANA.

44. ■ Iyer P, Dirweesh A, Zijoo R. Hydralazine induced lupus syndrome presenting with recurrent pericardial effusion and a negative antinuclear antibody. Case Rep Rheumatol 2017; 2017:5245904.

This is a special case of hydralazine-induced lupus erythematosus with a negative ANA in a female patient who developed recurrent pericardial effusion.

45. ■ Holman SK, Parris D, Meyers S, Ramirez J. Acute low-dose hydralazine-induced lupus pneumonitis. Case Rep Pulmonol 2017; 2017:2650142.

An uncommon clinical case of hydralazine-induced lupus erythematosus from a very low daily dose of hydralazine. It is worth to mention that a negative ANA is also observed in this case.

46. Magno Pereira V, Andrade C, Figueira R, *et al.* Infliximab-induced lupus: a case report. GE Port J Gastroenterol 2017; 24:84–88.
47. Lomicova I, Suchý D, Pizinger K, Cetkovská P. A case of lupus-like syndrome in a patient receiving adalimumab and a brief review of the literature on drug-induced lupus erythematosus. J Clin Pharm Ther 2017; 42: 363–366.
48. ■ Molina-Ruiz AM, Lasanta B, Barcia A, *et al.* Drug-induced systemic lupus erythematosus in a child after 3 years of treatment with carbamazepine. Australas J Dermatol 2017; 58:e20–e22.

A case report describing a child who developed carbamazepine-induced systemic lupus eryhematosus, which rarely occurrs in the paediatric population.

49. Mayor-Ibarguren A, Roldán-Puchalt MC, Gómez-Fernández C, *et al.* Subacute cutaneous lupus erythematosus induced by mitotane. JAMA Dermatol 2016; 152:109–111.
50. Reyes HA, Cativo EH, Sy AM. Drug-induced subacute cutaneous lupus erythematosus in a patient receiving therapy for chronic hepatitis C. Am J Ther 2016; 23:e1965–e1967.
51. Ben Zvi M, Vaknine H, Menczer J, *et al.* Gemcitabine-induced subacute cutaneous lupus erythematosus: a case report. Chemotherapy 2016; 61:236–239.
52. Singh H, Sukhija G, Tanwar V, *et al.* Rare occurrence of drug induced subacute cutaneous lupus erythematosus with leflunomide therapy. J Clin Diagn Res 2016; 10:OD06–OD07.
53. Kim WI, Kim JM, Kim GW. Subacute cutaneous lupus erythematosus induced by capecitabine: 5-FU was innocent. J Eur Acad Dermatol Venereol 2016; 30:e163–e164.
54. Kelly AS, De la Harpe Golden P, D'Arcy C, Lally A. Drug-induced lupus erythematosus secondary to pirfenidone. Br J Dermatol 2017. [Epub ahead of print]
55. Fisher J, Patel M, Miller M, Burris K. Anastrozole-induced subacute cutaneous lupus erythematosus. Cutis 2016; 98:E22–E26.
56. Yanes DA, Mosser-Goldfarb JL. A cutaneous lupus erythematosus-like eruption induced by hydroxyurea. Pediatr Dermatol 2017; 34:e30–e31.
57. Pinard J, Patel M, Granter SR, *et al.* Subacute cutaneous lupus erythematosus induced by palbociclib. J Cutan Med Surg 2018. [Epub ahead of print]
58. Mantis J, Bhavsar R, Abrudescu A. Drug-induced lupus erythematosus associated with antiretroviral therapy in a patient with human immunodeficiency virus: a case report. Cureus 2017; 9:e1661.
59. Ramachandran SM, Leventhal JS, Franco LG, *et al.* Topical drug-induced subacute cutaneous lupus erythematosus isolated to the hands. Lupus Sci Med 2017; 4:e000207.
60. Adrichem ME, Starink MV, van Leeuwen EMM, *et al.* Drug-induced cutaneous lupus erythematosus after immunoglobulin treatment in chronic inflammatory demyelinating polyneuropathy: a case series. J Peripher Nerv Syst 2017; 22:213–218.
61. Perry HM Jr, Tan EM, Carmody S, Sakamoto A. Relationship of acetyl transferase activity to antinuclear antibodies and toxic symptoms in hypertensive patients treated with hydralazine. J Lab Clin Med 1970; 76:114–125.
62. Strandberg I, Boman G, Hassler L, Sjöqvist F. Acetylator phenotype in patients with hydralazine-induced lupoid syndrome. Acta Med Scand 1976; 200:367–371.
63. Woosley RL, Drayer DE, Reidenberg MM, *et al.* Effect of acetylator phenotype on the rate at which procainamide induces antinuclear antibodies and the lupus syndrome. N Engl J Med 1978; 298:1157–1159.
64. Sonnhag C, Karlsson E, Hed J. Procainamide-induced lupus erythematosus-like syndrome in relation to acetylator phenotype and plasma levels of procainamide. Acta Med Scand 1979; 206:245–251.
65. Alarcon-Segovia D, Fishbein E, Alcala H. Isoniazid acetylation rate and development of antinuclear antibodies upon isoniazid treatment. Arthritis Rheum 1971; 14:748–752.
66. Evans DA, Bullen MF, Houston J, *et al.* Antinuclear factor in rapid and slow acetylator patients treated with isoniazid. J Med Genet 1972; 9:53–56.
67. Batchelor JR, Welsh KI, Tinoco RM, *et al.* Hydralazine-induced systemic lupus erythematosus: influence of HLA-DR and sex on susceptibility. Lancet 1980; 1:1107–1109.
68. Russell GI, Bing RF, Jones JA, *et al.* Hydralazine sensitivity: clinical features, autoantibody changes and HLA-DR phenotype. Q J Med 1987; 65:845–852.
69. Speirs C, Fielder AH, Chapel H, *et al.* Complement system protein c4 and susceptibility to hydralazine-induced systemic lupus erythematosus. Lancet 1989; 333:922–924.
70. Sim E, Dodds AW, Goldin A. Inhibition of the covalent binding reaction of complement component C4 by penicillamine, an antirheumatic agent. Biochem J 1989; 259:415–419.
71. Sim E, Gill EW, Sim RB. Drugs that induce systemic lupus erythematosus inhibit complement component C4. Lancet 1984; 2:422–424.
72. Rubin RL, Curnutte JT. Metabolism of procainamide to the cytotoxic hydroxylamine by neutrophils activated in vitro. J Clin Invest 1989; 83: 1336–1343.
73. Rubin RL. Autoantibody specificity in drug-induced lupus and neutrophil-mediated metabolism of lupus-inducing drugs. Clin Biochem 1992; 25: 223–234.
74. Jiang X, Khursigara G, Rubin RL. Transformation of lupus-inducing drugs to cytotoxic products by activated neutrophils. Science 1994; 266:810–813.
75. Nassberger L, Sjöholm AG, Jonsson H, *et al.* Autoantibodies against neutrophil cytoplasm components in systemic lupus erythematosus and in hydralazine-induced lupus. Clin Exp Immunol 1990; 81:380–383.
76. Cornacchia E, Golbus J, Maybaum J, *et al.* Hydralazine and procainamide inhibit T cell DNA methylation and induce autoreactivity. J Immunol 1988; 140:2197–2200.
77. Scheinbart LS, Johnson MA, Gross LA, *et al.* Procainamide inhibits DNA methyltransferase in a human T cell line. J Rheumatol 1991; 18:530–534.
78. Deng C, Lu Q, Zhang Z, *et al.* Hydralazine may induce autoimmunity by inhibiting extracellular signal-regulated kinase pathway signaling. Arthritis Rheum 2003; 48:746–756.

79. Yung R, Powers D, Johnson K, *et al.* Mechanisms of drug-induced lupus. II. T cells overexpressing lymphocyte function-associated antigen 1 become autoreactive and cause a lupuslike disease in syngeneic mice. J Clin Invest 1996; 97:2866–2871.
80. Quddus J, Johnson KJ, Gavalchin J, *et al.* Treating activated CD4+ T cells with either of two distinct DNA methyltransferase inhibitors, 5-azacytidine or procainamide, is sufficient to cause a lupus-like disease in syngeneic mice. J Clin Invest 1993; 92:38–53.
81. Rubin RL, Kretz-Rommel A. Initiation of autoimmunity by a reactive metabolite of a lupus-inducing drug in the thymus. Environ Health Perspect 1999; 107(Suppl 5):803–806.
82. Kretz-Rommel A, Duncan SR, Rubin RL. Autoimmunity caused by disruption of central T cell tolerance. A murine model of drug-induced lupus. J Clin Invest 1997; 99:1888–1896.
83. Mazari L, Ouarzane M, Zouali M. Subversion of B lymphocyte tolerance by hydralazine, a potential mechanism for drug-induced lupus. Proc Natl Acad Sci 2007; 104:6317–6322.
84. Ablin J, Verbovetski I, Trahtemberg U, *et al.* Quinidine and procainamide inhibit murine macrophage uptake of apoptotic and necrotic cells: a novel contributing mechanism of drug-induced-lupus. Apoptosis 2005; 10: 1009–1018.
85. Sontheimer RD, Henderson CL, Grau RH. Drug-induced subacute cutaneous lupus erythematosus: a paradigm for bedside-to-bench patient-oriented translational clinical investigation. Arch Dermatol Res 2009; 301:65–70.
86. Brinkmann V, Reichard U, Goosmann C, *et al.* Neutrophil extracellular traps kill bacteria. Science 2004; 303:1532–1535.
87. Grayson PC, Kaplan MJ. At the bench: neutrophil extracellular traps (NETs) highlight novel aspects of innate immune system involvement in autoimmune diseases. J Leukoc Biol 2016; 99:253–264.
88. ■■ Irizarry-Caro JA, Carmona-Rivera C, Schwartz DM, *et al.* Brief report: drugs implicated in systemic autoimmunity modulate neutrophil extracellular trap formation. Arthritis Rheumatol 2018; 70:468–474.
This study makes significant contribution indicating the inovlement of NETosis in the pathogenesis of DILE by studying several common offending drugs known to cause DILE. The findings suggest the involvement of the innate immune repose in DILE.
89. ■ Sawalha AH. Editorial: the innate and adaptive immune response are both involved in drug-induced autoimmunity. Arthritis Rheumatol 2018; 70:330–333.
This editorial summarizes the progress in our understanding of mechanisms involved in drug-induced autoimmunity and highlights the role played by epigenetic changes and the adaptive immune response, and NETosis and the innate immune response in the pathogenesis of DILE.

Autoimmune haemolytic anaemia and autoimmune thrombocytopenia in childhood-onset systemic lupus erythematosus: updates on pathogenesis and treatment

Jessica Neely and Emily von Scheven

Purpose of review
Autoimmune haemolytic anaemia (AIHA) and autoimmune thrombocytopenia are common complications of childhood-onset lupus, which may be life-threatening. A greater understanding of the pathogenesis of these haematologic manifestations will enhance our understanding of the biology of systemic lupus erythematosus (SLE) and inform the identification of novel treatments.

Recent findings
The mechanisms underlying AIHA and autoimmune thrombocytopenia are incompletely understood and likely multifactorial. Although the development of auto-antibodies is central to the disease process, recent studies have demonstrated the importance of cytokines in the underlying pathologic process. In-vitro and in-vivo evidence points to a role for IL17 in the pathogenesis of AIHA, which involves loss of tolerance to red cell auto-antigens and the development of autoantibodies. Sirolimus, an mTor inhibitor, has benefited patients with primary autoimmune cytopenias, possibly by stimulating T regulatory cells, and may also have efficacy for SLE-associated cytopenias. Similarly, low-dose recombinant human IL-2 therapy has shown promising results for improving platelet counts in patients with autoimmune thrombocytopenia, possibly by restoring the balance between T regulatory, T helper and Th17 cells.

Summary
The emergence of new agents directed at restoring immune dysregulation hold promise for the treatment of AIHA and autoimmune thrombocytopenia and should provide better tolerated alternatives to high-dose corticosteroids.

Keywords
autoimmune haemolytic anaemia, autoimmune thrombocytopenia, childhood-onset systemic lupus erythematosus

INTRODUCTION

Autoimmune haematologic manifestations are common among patients with childhood-onset systemic lupus erythematosus (SLE) and may be the presenting symptom [1]. Patients with SLE are at risk for a number of haematologic complications; however, we focus our discussion on autoimmune haemolytic anaemia (AIHA) and autoimmune thrombocytopenia, both of which are included in the 1997 Revised American College of Rheumatology (ACR) criteria for SLE [2] and the 2012 SLICC (Systemic Lupus International Collaborating Clinics) criteria [3]. In addition, AIHA and autoimmune thrombocytopenia are each given a weight of 4 points in the new proposed ACR/European League Against Rheumatism criteria [4] highlighting the need for rheumatologists to be familiar with the clinical presentation and diagnosis. Recent large-scale epidemiologic studies in South America demonstrated greater prevalence and severity of haematologic manifestations in childhood-onset SLE (cSLE) compared with adult-onset SLE (aSLE) [5–8]. Among these studies, the prevalence of AIHA in cSLE was found to be as high as 25%, and the prevalence of autoimmune thrombocytopenia as high as 25%. Comparison of cSLE to aSLE patients found that children with AIHA and autoimmune

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KEY POINTS

- Autoimmune cytopenias occur more frequently in childhood than adult SLE, affecting approximately 25% of children.
- Cytokine disturbances, including elevation of IL-17, appears to play an important role in pathogenesis.
- Agents that act to restore T-cell regulatory mechanisms, such as sirolimus and low-dose IL-2, hold promise as therapeutic agents.
- Thrombopoeitin receptor agonists may be helpful adjunctive therapy, in addition to immune modulation, for autoimmune thrombocytopenia.

thrombocytopenia had more constitutional symptoms, including more fever, weight loss and hepatosplenomegaly than adults [6,7]. In addition, children with AIHA had lower mean haemoglobin at diagnosis, 8.3 g/dl (range 2.2–10 g/dl) [7], and those with AT experienced more haemorrhagic manifestations and had higher incidence of acute renal failure, pericarditis and central nervous system involvement [6]. Accordingly, it is critical that Pediatric Rheumatologists remain up to date regarding the pathogenesis and treatment of autoimmune cytopenias in cSLE. In this review, we discuss the current understanding of the pathophysiology of AIHA and autoimmune thrombocytopenia in patients with SLE and summarize the most recent literature on treatment, with a focus on treatment of refractory disease. As many questions on this topic remain unanswered, we additionally discuss several forthcoming areas of research.

AUTOIMMUNE HAEMOLYTIC ANAEMIA: CLINICAL FEATURES AND PATHOGENESIS

Autoimmune haematologic manifestations in SLE are triggered by loss of tolerance to red blood cell (RBC) auto-antigens. Antibodies produced by autoreactive B cells, usually IgG, bind RBC antigens causing destruction of red cells either through complement fixation and removal by phagocytes or through antibody-dependent cell-mediated cytotoxicity by cytotoxic $CD8^+$ T cells in the reticuloendothelial system [9]. Clinically, AIHA can range from mild haemolysis with compensatory reticulocytosis to brisk, life-threatening haemolysis resulting in haemodynamic compromise. Common clinical symptoms include fatigue, pallor, dyspnoea on exertion and tachypnoea. The diagnosis is confirmed with a positive direct antiglobulin test (DAT), or Coombs test, and laboratory evidence of haemolysis, including anaemia, elevated lactate dehydrogenase (LDH), indirect bilirubin and reticulocyte count and depressed haptoglobin. Surprisingly, patients with active haemolysis are also at an increased risk of thrombotic events due to increased fibrin and thrombin generation, circulating microparticles and externalization of membrane phospholipids that contribute to a hypercoagulable state [10].

Current understanding of AIHA pathogenesis in SLE is derived from our understanding of both SLE and primary AIHA; however, the exact mechanisms by which tolerance to red cell antigens is lost remain unknown. The pathogenesis of SLE likely results from a variety of mechanisms, which ultimately lead to disease through a complex interplay of genetic factors, environment triggers and defects in innate and adaptive immunity that result in loss of self-tolerance, clonal expansion of autoreactive cells and loss of regulatory mechanisms [11,12]. Although autoreactive B cells are responsible for auto-antibody production, helper T cells are critical in promoting autoantibody production [12]. Th17 cells, the subset that produce IL-17, are elevated in patients with AIHA and appear to correlate with disease activity [13]. In in-vitro experiments using mouse models, the transfer of Th17 cells enhanced onset of AIHA, whereas neutralization of IL-17 inhibited development of AIHA [13]. Elevated levels of IL-17 have also been found in serum and tissues of patients with SLE and early evidence in animal models suggests an important role for IL-17 in organ damage and autoantibody production in SLE [14,15]. Another helper T cell subset, Treg cells, are vital in maintaining immune tolerance and mitigating the immune response. Decreased numbers of Treg cells have been found in both patients with SLE [15] and primary AIHA [16] leading to imbalance of immunoregulatory and inflammatory cytokines and promoting autoimmunity. Restoration of Treg cell balance is thus a promising therapeutic strategy [17▪▪].

The autoantibodies produced in AIHA are primarily of the IgG subtype in patients with SLE, though IgM antibodies leading to cold AIHA have also been reported. There has long been interest in the association between antiphospholipid antibodies (aPLs) and AIHA in patients with SLE. aPLs have been speculated to play a pathogenic role in AIHA through cross-reactivity with phospholipids on the erythrocyte membrane; however, conclusive evidence to support this claim is lacking [18]. Studies show an increased risk of developing AIHA in SLE patients positive for aPLs [19]. Likewise, there is a distinct subset of patients with primary APS without SLE that exhibit haematological manifestations

including AIHA and autoimmune thrombocytopenia [20]. Recently, a systematic review and meta-analysis investigated the risk of developing AIHA in aPL-positive compared with aPL-negative SLE patients and found a cumulative increased odds of AIHA of 3.22 [95% confidence interval (95% CI) 2.40–4.32] in aPL-positive patients, confirming the association [21], though the significance of this finding is not clear.

AUTOIMMUNE THROMBOCYTOPENIA: CLINICAL FEATURES AND PATHOGENESIS

Autoimmune thrombocytopenia in cSLE is defined as a platelet count less than 100 x 10^9/l in the absence of other causes. Thrombocytopenia is usually in the mild range (between 50 x 10^9/l and 100 x 10^9/l); however, severe thrombocytopenia with platelets less than 10 x 10^9/l can occur [22]. In general, patients with primary immune thrombocytopenia (ITP) have lower platelet counts and higher frequency of haemorrhagic manifestations compared to patients with SLE-associated autoimmune thrombocytopenia [23]. The frequency of severe thrombocytopenia in adults with SLE is roughly 20% [22] and in children with SLE roughly 13% [6]. The frequency of haemorrhagic manifestations in one cohort of children with SLE-associated autoimmune thrombocytopenia was 34% and most commonly included ecchymosis, epistaxis and gingival bleeding; however, 6% of children experienced diffuse alveolar haemorrhage [6]. Other severe haemorrhagic complications such as gastrointestinal bleeding or intracranial haemorrhage, while rare, may develop.

Similar to the pathobiology of primary ITP, SLE-associated autoimmune thrombocytopenia may also result from autoantibodies directed against platelet glycoproteins that leads to platelet opsonization and removal of platelets by the reticuloendothelial system [24]. The most commonly detected autoantibody is anti-GIIbIIIa, found in up to 76% of patients with SLE [25], though there are often multiple autoantibodies against various platelet glycoproteins secondary to epitope spreading. Testing for antiplatelet antibodies is not widely available in the clinical setting, and absence of detectable antibody does not rule out autoimmune thrombocytopenia in the SLE population. In addition to antibodies against platelet glycoproteins, there are likely additional pathways that lead to thrombocytopenia in SLE including antibodies directed against megakaryocytes or the thrombopoeitin receptor and aPL antibodies [24]. Although aPLs are associated with thrombocytopenia [19] and thrombocytopenia is a feature of APS, the biological significance of aPLs in autoimmune thrombocytopenia is also unclear.

In addition to antibody-mediated platelet destruction, defects in innate and adaptive immunity among patients with SLE may also contribute to the pathogenesis. Kang *et al*. [26] have shown that undifferentiated megakaryocytes, or MM cells, in the bone marrow serve as antigen-presenting cells (APCs) and induce Th17 cell responses in SLE-prone mice. This identification of the MM cell as novel APC provides insight into one possible link between SLE and autoimmune thrombocytopenia.

ADVANCES IN UNDERSTANDING DISEASE PATHOGENESIS: WHAT IS ON THE HORIZON?

Gaps in knowledge remain regarding the pathogenesis of these important autoimmune hematologic manifestations in patients with SLE. The underlying genetic factors that increase susceptibility to AIHA or autoimmune thrombocytopenia, and the immunology behind the loss of tolerance to red cell and platelet antigens remain to be explored. Researchers at the Puget Sound Blood Center have created a novel murine model that is easily manipulated to study how immune tolerance to RBC antigens is established and to test therapeutic and prevention strategies for AIHA [27]. A San Diego laboratory group has identified a genetic locus, Lbw2, that confers risk of developing antierythrocyte antibodies and AIHA. Whole genome sequencing is being used to determine candidate genes at this locus and explore how variation in these genes promotes autoantibody formation against RBCs [28]. A Stanford group is utilizing a novel DNA-barcoding technology termed 'antibody repertoire capture' to enable large-scale profiling of antibodies in patients with SLE to determine antibody profiles associated with disease subtypes and gain insight into the mechanistic role of antibodies in these subtypes [29]. These novel approaches hold promise for elucidating the pathogenesis of AIHA and autoimmune thrombocytopenia in patients with SLE and identifying therapeutic targets for those patients with refractory disease.

UPDATES ON TREATMENT OF AUTOIMMUNE HAEMOLYTIC ANAEMIA AND AUTOIMMUNE THROMBOCYTOPENIA IN SLE

In patients with SLE, treatment for secondary autoimmune cytopenias should be directed at treatment of the underlying SLE, after considering the severity of other organ manifestations. First-line treatment

includes corticosteroids in the form of pulse IV methylprednisolone or high-dose oral prednisone. Steroid-sparing agents include hydroxychloroquine [30], IVIG [30], mycophenolate mofetil (MMF) [31], azathioprine [32], danazol [30], cyclophosphamide [33], rituximab [34,35▪,36▪] and tacrolimus [37]. Initial response to steroid therapy is usually favourable, but a small fraction of patients develop refractory disease. Choice of a second or third-line therapeutic agent is more challenging but should be informed by the severity of the disease and urgency for response. Patients with chronic, low-level haemolysis may be treated with agents that take time to reach therapeutic levels, such as MMF, whereas patients with brisk haemolysis need faster acting agents. A summary of treatment studies specifically evaluating treatments for AIHA and autoimmune thrombocytopenia in SLE is presented in Table 1. Lastly, it should also be noted that patients with chronic or repeated episodes of haemolysis should receive folate replacement due to ongoing losses to prevent megaloblastic anaemia [38].

RITUXIMAB

Rituximab, a monoclonal anti-CD20 antibody, is effective as a second-line therapy for refractory disease and may even be a good initial therapeutic agent for patients with severe haemolysis. A retrospective cohort study evaluated the efficacy of rituximab in 24 patients with cSLE and refractory cytopenias, including AIHA, autoimmune thrombocytopenia or both [35▪]. All patients except one demonstrated response to therapy defined as platelet count more than 100 x 10^9/l with a median response time of 48 days. This response was maintained in the majority of patients, but 5 had relapse of cytopenias. Median time to relapse was 15–27 months indicating over a year of remission after a single course of rituximab. These findings are in accordance with an earlier study of nine patients with cSLE and autoimmune cytopenias who were treated with rituximab and found durable response in all four patients with AIHA and six patients treated for autoimmune thrombocytopenia (one patient had both) [34]. Two of the six patients with autoimmune thrombocytopenia experienced relapse and responded to re-treatment. Similar findings supporting efficacy of rituximab for autoimmune cytopenias are reported in the adult SLE population [36▪].

THROMBOPOEITIN RECEPTOR AGONISTS

Two thrombopoeitin receptor (TPO) agonists, romiplostim and eltrombopag, have been approved for the treatment of chronic ITP in adults, but the efficacy of these agents in treating autoimmune thrombocytopenia in SLE is an active area of study. These agents bind to TPO receptors on bone marrow megakaryocytes to increase platelet production. Evidence for efficacy in SLE is limited and confined to small retrospective studies [39], the largest of which includes 16 adult patients with SLE in the French adult ITP national network [40▪▪]. These 16 patients failed multiple second-line therapies but ultimately had response to at least romiplostim or eltrombopag. Unfortunately, three patients developed thromboses, two while on eltrombopag who had a history of aPLs or APS developed arterial thrombosis, and one patient receiving romiplostim without aPLs developed venous thrombosis. This has raised concern about the safety of these agents in patients with APS or other thrombotic risk factors beyond active SLE. In the appropriate population, these agents have the potential to provide significant steroid-sparing effect for patients requiring chronic steroid therapy for refractory thrombocytopenia [41]. Eltrombopag has been FDA approved for children with refractory chronic ITP, and the safety and efficacy of both romiplostim and eltrombopag have been evaluated in randomized clinical trials [42,43]. A recent systematic review found that these agents improve overall and durable platelet response compared with placebo and that both agents are well tolerated in the paediatric population [44▪]. There were no thrombotic events reported in this subgroup of patients. Studies are needed to evaluate the safety and efficacy of these agents for refractory ITP in cSLE patients with close monitoring in patients with APS, aPLs and history of thrombosis.

NOVEL TREATMENTS FOR REFRACTORY DISEASE

For patients who are refractory to or intolerant of traditional immune suppressant medications used for SLE, there are a few promising agents currently under study.

SIROLIMUS

Sirolimus, an mTor inhibitor that blocks activation of T and B cells and may allow for the differential survival and expansion of regulatory T cells, is an interesting therapeutic option for patients with SLE and refractory cytopenias. Efficacy of sirolimus has been reported in children with refractory primary ITP and Evans syndrome [45▪]. Recently, a multicentre open-label clinical trial investigated the use of sirolimus in children with refractory multilineage cytopenias based on successful treatment of

Table 1. Evidence regarding steroid-sparing agents for treatment of haematologic disease in SLE

Drug	Study	Year	Design	Population	Indication (*N*)	Outcome
Azathioprine	Treatment of isolated severe immune haemolytic anaemia associated with SLE: 26 cases	2006	Retrospective cohort study	Five aSLE	AIHA (3), ES (2)	OR 80% CR 60%
Cyclophosphamide	Intermittent cyclophosphamide for the treatment of autoimmune thrombocytopenia in SLE	1990	Retrospective cohort study	Seven aSLE	AT	CR 100%
Danazol	Treatment of severe immune thrombocytopenia associated with SLE: 59 cases	2002	Retrospective cohort study	18 aSLE	AT	OR 50% CR 39% all sustained
Hydroxychloroquine				11 aSLE	AT	OR 64% CR 36% all sustained
IVIG				31 aSLE	AT	OR 65% CR 39% none sustained
Mycophenolate mofetil	Mycophenolate mofetil for nonrenal manifestations of SLE: a systematic review	2007	Systematic review	10 aSLE	AT (3), AIHA (5) leukopenia (1), PRCA (1)	Authors concluded good response
Rituximab	Efficacy and safety of rituximab for SLE-associated immune cytopenias: A multicentre retrospective cohort study of 71 adults	2017	Retrospective cohort study	71 aSLE	AT (44), AIHA (16), ES (10), PRCA (1)	OR 86% CR 60.5%
	Rituximab therapy has a rapid and durable response for refractory cytopenia in childhood-onset SLE	2015	Retrospective cohort study	24 cSLE	AT (16), AIHA (5), ES (3)	CR 96%
	B-cell depletion for autoimmune thrombocytopenia and autoimmune haemolytic anaemia in paediatric SLE	2009	Retrospective cohort study	Nine cSLE	AT (5), AIHA (3), ES (1)	CR 100%
Sirolimus	Sirolimus is effective in relapsed/refractory autoimmune cytopenias: results of a prospective multiinstitutional trial	2016	Multicentre, prospective, open-label trial	Two2 cSLE (total $N=30$)	ES (2)	CR by 3 months and 12 months
Tacrolimus	Efficacy and safety of tacrolimus in SLE patients with refractory thrombocytopenia: a retrospective study	2017	Retrospective cohort study	20 aSLE	AT	OR 100% CR 75% at 6 months
TPO agonists (Eltrombopag and Romiplostin)	Eltrombopag as steroid-sparing therapy for immune thrombocytopenic purpura in SLE	2015	Case series	Three aSLE	AT	All three attained platelets >50K
	Effectiveness of thrombopoietin-receptor agonists in the treatment of refractory immune thrombocytopenia associated with SLE	2014	Case series	Two aSLE	AT	Both attained CR platelets >100K
	Thrombopoietin-receptor agonist in SLE-associated immune thrombocytopenia: results of the 16 patients from the French Cohort	2016	Retrospective cohort study	16 aSLE	AT	OR 93% to at least one agent

Complete response (CR) for AT: platelets >100 000. Partial response (PR) for AT: platelets >30K but <100K or doubling of platelet count. CR for AIHA: Hgb normalized.
aSLE, adult-onset SLE; cSLE, childhood-onset SLE; ES, Evans syndrome; OR, overall response: complete or partial response; PRCA, pure red cell aplasia; SLE, systemic lupus erythematosus.

cytopenias in children with autoimmune lymphoproliferative syndrome (ALPS) [46▪▪]. Two of 30 children in this trial had SLE complicated by Evans syndrome and had previously failed treatment with corticosteroids, IVIG and rituximab. Both patients achieved complete response by 3 and 12 months, respectively, and were maintained safely on sirolimus for 2.5 and 3 years at trial completion. More studies are needed to determine safety in patients with cSLE, the immunological consequences of sirolimus treatment, and if patients can discontinue use of sirolimus once remission is attained.

LOW-DOSE INTERLEUKIN-2

Low-dose recombinant human interleukin -2 (rhIL-2) is gaining interest as a therapeutic agent for SLE and may be effective at treating hematologic manifestations. Patients with SLE have imbalance of effector and regulatory $CD4^+$ T cells, and low-dose IL-2 restores this balance by selectively increasing Treg cells while decreasing follicular helper T cells and Th17 cells [17▪▪]. A trial of low-dose recombinant human IL-2 (rhIL-2) in 38 patients with aSLE showed restoration of balance between these T cell subsets, decreased disease activity in all patients and significant corticosteroid reduction compared with baseline [17▪▪]. In the four patients with thrombocytopenia, resolution of thrombocytopenia was observed by week 12. Furthermore, there were no infections observed in any patients and common side effects included only injection site reactions in 13.2% and influenza-like symptoms in 5.3%, both of which resolved without any intervention. Not surprisingly, several clinical trials are actively recruiting patients to evaluate the efficacy of rhIL-2 in multiple autoimmune diseases, including the TRANSREG trial [47], which is recruiting patients across 14 different autoimmune and autoinflammatory diseases, and ANEMIL, a clinical trial of low-dose IL-2 in patients with primary warm AIHA [48].

COMBINATION RITUXIMAB AND BELIMUMAB

A study of patients with primary AIHA refractory to rituximab found the presence of long-lived splenic autoreactive plasma cells as well as elevated levels of B-cell activating factor (BAFF) in the supernatant of spleen cells [49]. This has led to speculation that BAFF promotes maturation and survival of autoreactive long-lived plasma cells in the spleen and that belimumab following rituximab may inhibit maturation of these pathogenic cells. A recent proof of concept trial evaluating combination therapy with rituximab followed by belimumab in severe, refractory SLE demonstrated reduction of pathogenic autoantibodies and neutrophil extracellular traps (NETs), pieces of DNA that serve as antigens for autoantibodies in SLE [50]. Median SLEDAI score improved from 18 to 2, and 10 out of 11 patients with lupus nephritis flare demonstrated response, including four who had a complete response. A phase II clinical trial is currently underway to investigate the combination regimen of rituximab and belimumab in adults with persistent ITP [51]. Combination therapy with belimumab administration after rituximab may be beneficial for patients with SLE and refractory cytopenias who have failed rituximab monotherapy.

CONCLUSION

Haematologic disease, including AIHA and autoimmune thrombocytopenia, are important autoimmune manifestations present in roughly 25% of paediatric SLE, and, in some patients, may be refractory and dominate the clinical picture. Although there is some knowledge of the mechanisms behind the immune dysregulation that leads to loss of tolerance to red cell and platelet antigens, much remains to be learned. As we continue to learn more about the genetic and molecular underpinnings of SLE and begin to molecularly characterize the heterogeneous phenotypes of SLE, we will gain insights into the pathophysiology behind autoimmune hematologic disease. These advances will inform the development of targeted treatment strategies both for SLE patients with refractory cytopenias and those with primary AIHA and ITP.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ▪ of special interest
- ▪▪ of outstanding interest

1. Levy DM, Kamphuis S. Systemic lupus erythematosus in children and adolescents. Pediatr Clin North Am 2012; 59:345–364.

2. Hochberg MC. Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum 1997; 40:1725.
3. Petri M, Orbai A-M, Alarcón GS, *et al.* Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. Arthritis Rheum 2012; 64:2677–2686.
4. Tedeschi SK, Johnson S, Costenbader KH, *et al.* A multicriteria decision analysis for the development of new systemic lupus erythematosus classification criteria. ACR Meeting Abstracts. 2017 ACR/ARHP Annu. Meet. Abstr. Number 1589, San Diego, CA; 18 September 2017.
5. Ramírez Gómez LA, Uribe OU, Uribe OO, *et al.* Childhood systemic lupus erythematosus in Latin America. The GLADEL experience in 230 children. Lupus 2008; 17:596–604.
6. Esteves GCX, Gormezano NWS, Pereira OL, *et al.* Distinct clinical correlates of immune thrombocytopenic purpura at diagnosis of childhood-onset and adult SLE. Mod Rheumatol 2017; 1–5. [Epub ahead of print]
7. Gormezano NWS, Kern D, Pereira OL, *et al.* Autoimmune hemolytic anemia in systemic lupus erythematosus at diagnosis: differences between pediatric and adult patients. Lupus 2017; 26:426–430.
8. Lube GE, Ferriani MPL, Campos LMA, *et al.* Evans syndrome at childhood-onset systemic lupus erythematosus diagnosis: a large multicenter study. Pediatr Blood Cancer 2016; 63:1238–1243.
9. Barcellini W. New insights in the pathogenesis of autoimmune hemolytic anemia. Transfus Med Hemother 2015; 42:287–293.
10. Ataga KI. Hypercoagulability and thrombotic complications in hemolytic anemias. Haematologica 2009; 94:1481–1484.
11. Liu Z, Davidson A. Taming lupus: a new understanding of pathogenesis is leading to clinical advances. Nat Med 2012; 18:871–882.
12. Moulton VR, Suarez-Fueyo A, Meidan E, *et al.* Pathogenesis of human systemic lupus erythematosus: a cellular perspective. Trends Mol Med 2017; 23:615–635.
13. Xu L, Zhang T, Liu Z, *et al.* Critical role of Th17 cells in development of autoimmune hemolytic anemia. Exp Hematol 2012; 40:994–1004.e4.
14. Martin JC, Baeten DL, Josien R. Emerging role of IL-17 and Th17 cells in systemic lupus erythematosus. Clin Immunol 2014; 154:1–12.
15. Yang J, Chu Y, Yang X, *et al.* Th17 and natural Treg cell population dynamics in systemic lupus erythematosus. Arthritis Rheum 2009; 60:1472–1483. doi:10.1002/art.24499.
16. Ahmad E, Elgohary T, Ibrahim H. Naturally occurring regulatory T cells and interleukins 10 and 12 in the pathogenesis of idiopathic warm autoimmune hemolytic anemia. J Investig Allergol Clin Immunol 2011; 21:297–304.
17. ■■ He J, Zhang X, Wei Y, *et al.* Low-dose interleukin-2 treatment selectively modulates CD4+ T cell subsets in patients with systemic lupus erythematosus. Nat Med 2016; 22:991–993.

This is the largest study investigating the use of low-dose IL-2 treatment in patients with SLE and demonstrates reduced follicular T helper subsets and Th17 cells and coincident increases in the Treg population. This study also demonstrates that low-dose IL-2 reduces disease activity in SLE.

18. Comellas-Kirkerup L, Hernández-Molina G, Cabral AR. Antiphospholipid-associated thrombocytopenia or autoimmune hemolytic anemia in patients with or without definite primary antiphospholipid syndrome according to the Sapporo revised classification criteria: a 6-year follow-up study. Blood 2010; 116:3058–3063.
19. Ünlü O, Zuily S, Erkan D. The clinical significance of antiphospholipid antibodies in systemic lupus erythematosus. Eur J Rheumatol 2016; 3:75–84.
20. Zuily S, Andrade D, Erkan D, Tektonidou M. ACTION on behalf of A. FRI0314|Antiphospholipid Syndrome Alliance for Clinical Trials and International Networking (APS Action) clinical database and repository analysis: the impact of systemic lupus erythematosus on the clinical phenotype of antiphospholipid antibody-positive patients. Ann Rheum Dis 2016; 75:548.
21. Unlu O, Wahl D, Zuily S, *et al.* Increased risk of hemolytic anemia associated with antiphospholipid antibodies in patients with systemic lupus erythematosus: a systematic review and meta-analysis. ACR Meeting Abstracts. 2015 ACR/ARHP Annu. Meet. Abstr. Number 2193, San Francisco, CA; 29 September 2015.
22. Jung J-H, Soh M-S, Ahn Y-H, *et al.* Thrombocytopenia in systemic lupus erythematosus: clinical manifestations, treatment, and prognosis in 230 patients. Medicine (Baltimore) 2016; 95:e2818.
23. Liu Y, Chen S, Sun Y, *et al.* Clinical characteristics of immune thrombocytopenia associated with autoimmune disease: a retrospective study. Medicine (Baltimore) 2016; 95:e5565.
24. Cines DB, Liebman H, Stasi R. Pathobiology of secondary immune thrombocytopenia. Semin Hematol 2009; 46:S2–S14.
25. Kuwana M, Okazaki Y, Ikeda Y. Detection of circulating B cells producing anti-GPIb autoantibodies in patients with immune thrombocytopenia. PLoS One 2014; 9:e86943.
26. Kang H-K, Chiang M-Y, Ecklund D, *et al.* Megakaryocyte progenitors are the main APCs inducing Th17 response to lupus autoantigens and foreign antigens. J Immunol 2012; 188:5970–5980.
27. Hudson KE. Project information - NIH RePORTER - NIH research portfolio online reporting tools expenditures and results. Immune response to RBC antigens (n.d.) https://projectreporter.nih.gov/project_info_description.cfm?aid=9389295&icde=38394886&ddparam=&ddvalue=&ddsub=&cr=2&csb=default&cs=ASC&pball=. [Accessed 4 May 2018].
28. Kono DH. Project information - NIH RePORTER - NIH research portfolio online reporting tools expenditures and results. Genetics of autoimmune hemolytic anemia (n.d.). https://projectreporter.nih.gov/project_info_description.cfm?aid=9119065&icde=38396283 [Accessed 4 May 2018].
29. Robinson W. Project information - NIH RePORTER - NIH research portfolio online reporting tools expenditures and results. Large-scale sequencing and characterizing of autoantibody responses n.d. https://projectreporter.nih.gov/project_info_description.cfm?aid=9266356&icde=38394886. [Accessed 4 May 2018].
30. Papo T, Schaeffer A, Bierling P, *et al.* Treatment of severe immune thrombocytopenia associated with systemic lupus erythematosus. J Rheumatol 2002; 29:75–83.
31. Mok CC. Mycophenolate mofetil for nonrenal manifestations of systemic lupus erythematosus: a systematic review. Scand J Rheumatol 2007; 36: 329–337.
32. Gomard-Mennesson E, Ruivard M, Koenig M, *et al.* Treatment of isolated severe immune hemolytic anaemia associated with systemic lupus erythematosus: 26 cases. Lupus 2006; 15:223–231.
33. Boumpas DT, Barez S, Klippel JH, Balow JE. Intermittent cyclophosphamide for the treatment of autoimmune thrombocytopenia in systemic lupus erythematosus. Ann Intern Med 1990; 112:674–677.
34. Kumar S, Benseler SM, Kirby-Allen M, Silverman ED. B-cell depletion for autoimmune thrombocytopenia and autoimmune hemolytic anemia in pediatric systemic lupus erythematosus. Pediatrics 2009; 123: e159–e163.
35. ■ Olfat M, Silverman ED, Levy DM. Rituximab therapy has a rapid and durable response for refractory cytopenia in childhood-onset systemic lupus erythematosus. Lupus 2015; 24:966–972.

This is the largest study demonstrating efficacy of rituximab specifically for hematologic manifestations in pediatric SLE.

36. ■ Serris A, Amoura Z, Canouï-Poitrine F, *et al.* Efficacy and safety of rituximab for systemic lupus erythematosus-associated immune cytopenias. A multicenter retrospective cohort study of 71 adults. Am J Hematol 2018; 93: 424–429.

This is the largest study demonstrating efficacy of rituximab in a multicentre cohort of adult SLE patients with haematologic manifestations.

37. Li Y, Feng X. Efficacy and safety of tacrolimus in systemic lupus erythematosus patients with refractory thrombocytopenia: a retrospective study. Lupus 2018; 27:60–65.
38. Hill QA, Stamps R, Massey E, *et al.* Guidelines on the management of drug-induced immune and secondary autoimmune, haemolytic anaemia. Br J Haematol 2017; 177:208–220.
39. Magnano L, Enriquez H, Esteve J, *et al.* Effectiveness of thrombopoietin-receptor agonists in the treatment of refractory immune thrombocytopenia associated to systemic lupus erythematosus. J Rheumatol 2014; 41: 1895–1896.
40. ■■ Terriou L, Guitton Z, Hie M, *et al.* Thrombopoietin-receptor agonist in systemic lupus erythematosus associated immune thrombocytopenia: results of the 16 patients from the French cohort. Blood 2016; 128:2542.

This is the largest study evaluating the use of TPO-agonists in the treatment of refractory ITP in adult SLE patients. This study shows these agents are efficacious in the treatment of SLE-associated thrombocytopenia, however, also reports presence of thrombosis in three patients treated with these agents.

41. Maroun M-C, Ososki R, Andersen JC, Dhar JP. Eltrombopag as steroid sparing therapy for immune thrombocytopenic purpura in systemic lupus erythematosus. Lupus 2015; 24:746–750.
42. Tarantino MD, Bussel JB, Blanchette VS, *et al.* Romiplostim in children with immune thrombocytopenia: a phase 3, randomised, double-blind, placebo-controlled study. Lancet 2016; 388:45–54.
43. Grainger JD, Locatelli F, Chotsampancharoen T, *et al.* Eltrombopag for children with chronic immune thrombocytopenia (PETIT2): a randomised, multicentre, placebo-controlled trial. Lancet 2015; 386:1649–1658.
44. ■ Zhang J, Liang Y, Ai Y, *et al.* Thrombopoietin-receptor agonists for children with immune thrombocytopenia: a systematic review. Expert Opin Pharmacother 2017; 18:1543–1551.

This is a systematic review demonstrating efficacy and safety of TPO agonists in children with primary ITP.

45. ■ Jasinski S, Weinblatt ME, Glasser CL. Sirolimus as an effective agent in the treatment of immune thrombocytopenia (ITP) and Evans syndrome (ES). J Pediatr Hematol Oncol 2017; 39:420–424.

This is a retrospective analysis demonstrating safety and efficacy of sirolimus in children with refractory primary ITP and Evans syndrome.

46. ■■ Bride KL, Vincent T, Smith-Whitley K, *et al.* Sirolimus is effective in relapsed/refractory autoimmune cytopenias: results of a prospective multiinstitutional trial. Blood 2016; 127:17–28.

This is an open-label clinical trial evaluating sirolimus for children with refractory cytopenias secondary to autoimmune disease or ALPS and includes two children with refractory cytopenias secondary to SLE. This study demonstrates efficacy of sirolimus in these two paediatric SLE patients and no adverse events.

47. Klatzmann D (PI). Induction of regulatory T cells by low dose IL2 in autoimmune and inflammatory diseases - full text view. ClinicalTrials.gov (n.d.). https://clinicaltrials.gov/ct2/show/NCT01988506. [Accessed 4 May 2018].

48. Lazaro E (PI). Evaluating the interest of interleukine-2 for patients with active warm hemolytic anemia resistant to conventional treatment - full text view. ClinicalTrials.gov n.d. https://clinicaltrials.gov/ct2/show/NCT02389231?cond=autoimmune+hemolytic+anemia&rank=7 [Accessed 4 May 2018].
49. Mahévas M, Michel M, Vingert B, *et al.* Emergence of long-lived autoreactive plasma cells in the spleen of primary warm auto-immune hemolytic anemia patients treated with rituximab. J Autoimmun 2015; 62:22–30.
50. Kraaij T, Kamerling SWA, Teng YKO, *et al.* Synergetic B-cell immunomodulation with rituximab and belimumab combination treatment in severe, refractory SLE – 2017. ACR Meeting Abstract Number 890. ACR/ARHP Annu. Meet, San Diego, CA; 18 September 2017.
51. Mahevas M (PI). Efficacy and safety assessment of a treatment combining rituximab and belimumab in adults with persistent immune thrombocytopenia: full text view - ClinicalTrials.gov n.d. https://clinicaltrials.gov/ct2/show/NCT03154385 [Accessed 4 May 2018].

Haploinsufficiency of A20 and other paediatric inflammatory disorders with mucosal involvement

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Purpose of review
This review aims at summarizing the current knowledge of A20 haploinsufficiency and other paediatric inflammatory disorders with mucosal involvement.

Recent findings
A20 haploinsufficiency is a newly described autoinflammatory disease caused by loss-of-function mutations in *TNFAIP3* that result in the activation of the nuclear factor (NF)-kB pathway. Patients may present with dominantly inherited, early-onset systemic inflammation and a Behçet-like disease, or a variety of autoinflammatory and autoimmune features. In Behçet disease, recent literature provides insights into genetic susceptibility and emerging treatment options; in addition, the first paediatric classification criteria were published. Recent advances in periodic fever, aphthous stomatitis, pharyngitis and adenitis syndrome (PFAPA) suggest that the disease has a complex underlying genetic mechanism and in some cases is inherited in an autosomal dominant pattern with reduced penetrance phenotype in many family members. Activation of the pyrin inflammasome through the RoA signalling pathway uncovers an interesting molecular connection between hyperimmunoglobulinemia D syndrome and familial Mediterranean fever. The description of new monogenic types of inflammatory bowel disease (IBD) may provide novel insights into disease pathogenesis. Finally, recent studies highlighted the role of gut microorganisms and dysbiosis in IBD.

Summary
Monogenic diseases such as A20 haploinsufficiency may help to advance our understanding of disease pathogenesis and to develop targeted therapies for more common, multifactorial disorders with mucosal inflammation.

Keywords
autoinflammatory syndrome, Behçet disease, haploinsufficiency of A20, inflammatory bowel disease, TNFAIP3/A20

INTRODUCTION

Autoinflammatory diseases are a heterogeneous group of inherited conditions caused by abnormal activation of cells mediating innate immunity. Recently, the recognition of a new group of autoinflammatory disorders resulting from dysregulation in the ubiquitin pathway has expanded our understanding of molecular mechanisms underlying autoinflammation. To date, haploinsufficiency of A20 (HA20), OTULIN deficiencies (otulipenia) and linear ubiquitin chain assembly complex (LUBAC) deficiencies have been associated with dysregulated ubiquitination [1].

In this review, we summarize the current knowledge of HA20 and present recent findings in other paediatric inflammatory disorders with mucosal involvement that might be considered in the differential diagnosis of HA20.

PATHOGENESIS OF A20 HAPLOINSUFFICIENCY

Ubiquitination is a posttranslational protein modification and an important mechanism for the regulation of many processes of cell physiology, such as protein degradation or DNA repair. The process of ubiquitination is dynamic and reversible;

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KEY POINTS

- Mucosal inflammation may be the first and most prominent symptom of a variety of systemic autoinflammatory and autoimmune diseases, which must be recognized to allow for early diagnosis and appropriate management.
- Haploinsufficiency of A20 may be considered in patients with a disease phenotype mimicking Behçet disease or other more common, multifactorial diseases, particularly those presenting with mucosal inflammation, an early disease onset and a positive family history.
- Several newly described monogenic forms of early-onset inflammatory bowel disease may shed light into the pathogenesis of later childhood and adult onset inflammatory bowel disease.

ubiquitin is removed by deubiquitinases (DUBs), also known as deubiquitinating enzymes [2]. The protein A20, encoded by *TNFAIP3*, is a DUB with a critical function in the negative regulation of inflammation and immunity [3,4] (Fig. 1). A20 functions as a negative regulator of the NF-kB pathway by cleaving K63 and linear UB chains from target molecules, RIPK1 and IKKγ. Decreased expression of A20 in patients with HA20 results in impaired deubiquitination, increased phosphorylation of the IKK complex and increased degradation of inhibitor of kB (IkB). This leads to an activation of the NF-kB pathway, an increased expression of proinflammatory cytokines and systemic inflammation [5▪▪] (Fig. 1).

The mutations in *TNFAIP3* reported to date are shown in Fig. 2. In addition, genetic variants in *TNFAIP3* have been associated with increased susceptibility to different autoimmune and inflammatory diseases, including juvenile idiopathic arthritis (JIA), rheumatoid arthritis (RA), inflammatory bowel disease (IBD), systemic lupus erythematosus (SLE), type 1 diabetes, psoriasis, celiac disease and coronary artery disease [6–11]. As a negative regulator of the NF-kB pathway, TNFAIP3/A20 plays a critical role in various cellular mechanisms, including cell activation, cytokine signalling and apoptosis [12], and activation of the proinflammatory transcripts in immune cells are contributing to the pathophysiology of above-mentioned diseases.

In mice, the cell-specific ablation of A20 causes clinical phenotypes mimicking human diseases. Specific ablation of A20 in myeloid cells results in severe destructive polyarthritis resembling RA and enterocyte-specific A20 deficiency increases the susceptibility to intestinal inflammation [13,14]. Tissue-specific deletion of A20 in B cells or dendritic cells results in the production of autoantibodies and an SLE-like autoimmune disease [15,16]. Finally, A20-deficient (A20−/−) mice develop severe multiorgan inflammation and cachexia with early mortality [17]. A20 was initially thought to be required to terminate TNF-induced signals, but due to the severe inflammation observed in double-deficient mice, A20-TNF or A20-TNFR1, it has been postulated that A20 may be crucial for the regulation of

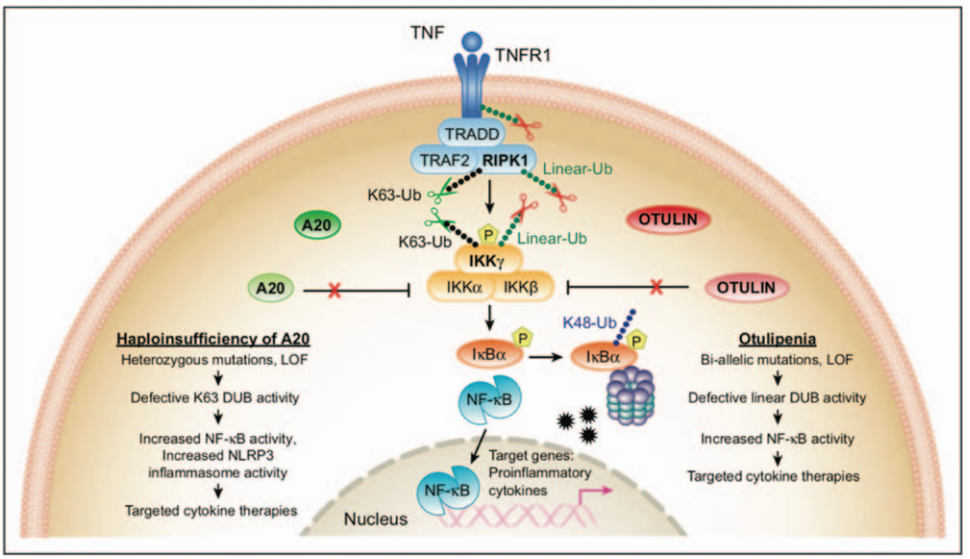

FIGURE 1. Proposed pathophysiologic mechanism of A20 haploinsufficiency (HA20). IKKγ, inhibitor of nuclear factor kappa B kinase subunit gamma; RIPK1, the death domain-containing protein kinase receptor-interacting protein 1; TNFR1, TNF receptor 1; TRADD, TNFR1-associated death domain protein. Reproduced with permission from [4].

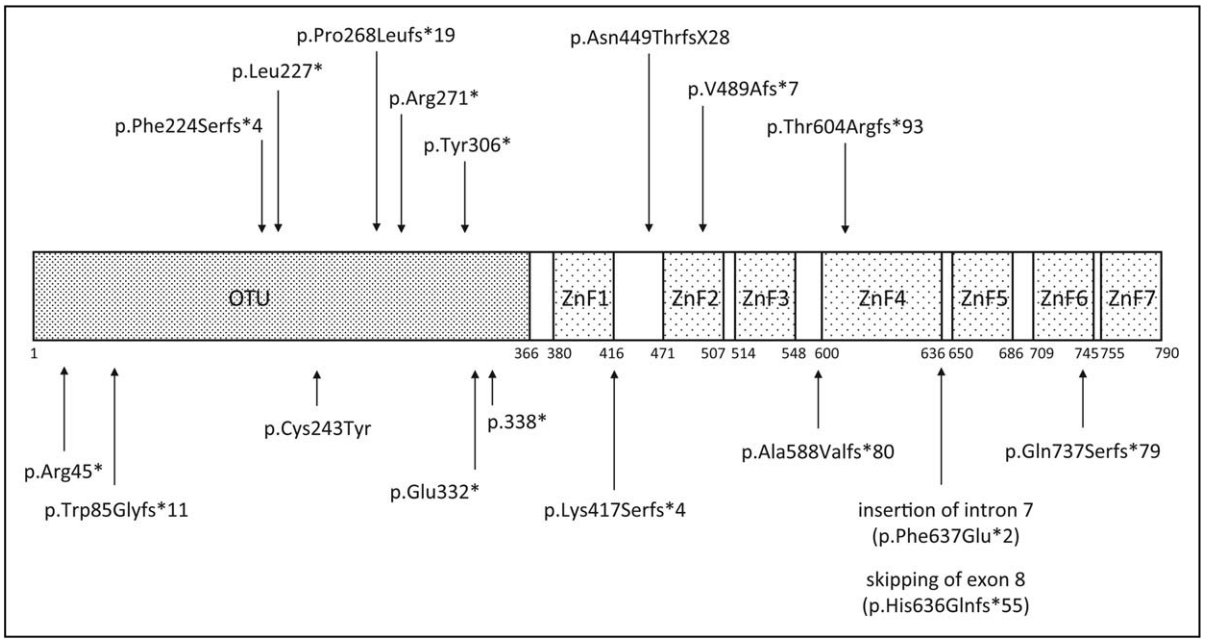

FIGURE 2. Schematic of domains in TNFAIP3/A20 and the location of mutations. The protein A20 consists of an N-terminal ovarian tumour (OTU) domain that mediates the DUB activity and seven zinc-finger (ZnF) domains that mediate A20 ubiquitin (Ub) E3 ligase activity. In addition to the loss-of-function mutations reported so far and shown in the figure, the complete deletion of one of the *TNFAIP3* alleles was described in a patient [27].

TNF-independent signals. A20 has also been shown to downregulate the activity of the NLRP3 inflammasome [18], and patients with HA20 had an increased NLRP3 activity [5▪▪]. In addition, A20 functions as a tumour-suppressor gene, and somatic mutations and deletions have been identified in B-cell and Hodgkin lymphomas [19].

To summarize, A20 functions as a ubiquitin-editing enzyme with a central role in the negative regulation of the NF-kB pathway. Failure to downregulate persistent NF-kB signalling in specific cells has been associated with various inflammatory and autoimmune disorders, and malignancies.

THE SPECTRUM OF CLINICAL PHENOTYPES ASSOCIATED WITH A20 HAPLOINSUFFICIENCY IS BROAD

Patients with HA20 present with dominantly inherited, early-onset systemic inflammation and a variety of autoinflammatory and autoimmune features (Table 1) [20▪,21▪,22–27].

The initially reported cohort included 16 patients from seven unrelated families [5▪▪,20▪]. The clinical picture was heterogeneous, and disease severity varied even within families, with some patients presenting with early-onset severe disease and others with later-onset only mildly affected. In most patients (56%), recurrent mucosal ulcers were the first symptom and, as a consequence, half of the patients were initially diagnosed with Behçet disease. Other diagnoses recorded prior to the recognition of HA20 included JIA and RA in patients with polyarthritis (25% of the patients), SLE, early-onset Crohn disease and periodic fever, aphthous stomatitis, pharyngitis and adenitis syndrome (PFAPA). Hence, it might be difficult to distinguish HA20 from other autoinflammatory and autoimmune diseases, especially early in the disease.

In this cohort, all patients developed recurrent painful ulcers of at least two sites during their disease course: oral (100%), genital (94%) and/or intestinal (six out of nine patients with gastrointestinal symptoms). Musculoskeletal symptoms including arthritis and intestinal involvement such as bloody diarrhoea were common (56% of patients). Recurrent fevers were reported in 50% of patients. The spectrum of cutaneous manifestations (50% of patients) ranged from pustular, folliculitis-like rashes and acne to dermal abscesses. Three patients (19%) suffered from severe ocular inflammation that manifested as treatment refractory anterior uveitis or retinal vasculitis resulting in chorioretinal scarring and macular fibrosis.

Immune cells from patients with HA20 showed evidence of increased expression of NF-kB target genes and elevated levels of several proinflammatory cytokines [interleukin (IL)-1β, tumour necrosis factor (TNF), IL-6, IL-9, IL-17, IL-18, interferon-gamma (IFNγ) and IP-10] [5▪▪].

Table 1. Clinical features of patients with A20 haploinsufficiency (HA20)

Characteristic features	Zhou *et al.* [5▪▪] Aeschlimann *et al.* [20▪] (*N* = 16)[a]	Kadowaki *et al.* [21▪] Ohnishi *et al.* [24] Shigemura *et al.* [25] Tagaki *et al.* [26] (*N* = 22)	Duncan *et al.* [23] (*N* = 1)	Berteau *et al.* [22] (*N* = 3)	Franco-Jarava *et al.* [27] (*N* = 1)
F/M	13/3	13/9	0/1	2/1	0/1
Age at disease onset	Mostly early-onset Range: neonate – 29 years	Mostly early-onset Range: neonate – 20 years	10 years	Early-onset Range: 6 months–6 years	2 months
Mucosal ulcers	16/16 Oral (*n* = 16) Genital (*n* = 15) GI (*n* = 6)	13/22 Oral (*n* = 13) Genital (*n* = 7) GI (*n* = 4) Palpebral conjunctiva (*n* = 1)	NR	3/3 Oral (*n* = 3) Genital (*n* = 2) GI (*n* = 2)	Oral Genital GI
Recurrent fever	7/16 Episodic	12/22 Episodic	NR	3/3 Episodic	Episodic
Musculoskeletal	9/16 Arthritis, arthralgia, myalgia	3/22 Arthritis, enthesitis, myalgia	NR	2/3 Arthritis, arthralgia	NR
Gastrointestinal	9/16 Abdominal pain, diarrhoea, IBD-like disease, colon perforation	8/22 Abdominal pain, diarrhoea, IBD-like disease, colon perforation	Enteropathy	3/3 Abdominal pain, diarrhoea, vomiting	Diarrhoea, duodenal inflammation
Cutaneous	8/16 Pustular or folliculitis-like rash, acne, cutaneous abscess	8/22 Psoriasis, pseudofolliculitis, EN-like lesions, severe local reaction to BCG vaccine, aquagenic acrokeratoderma	NR	1/3 Pseudofolliculitis, urticaria	Neutrophilic dermatosis
Cardiovascular	3/16 Pericarditis, venous thrombosis	1/22 Aortic valve insufficiency	NR	1/3 Thrombophlebitis	NR
Ocular	3/16 Anterior uveitis, retinal vasculitis	NR	NR	1/3 Episcleritis	NR
Autoimmune features	Thyroiditis, lupus nephritis, ITP, SLE-like features, autoantibodies	Hashimoto thyroiditis, Grave's disease, autoimmune hepatitis, ALPS-undefined, autoantibodies	Insulin-dependent diabetes, hepatitis, interstitial lung disease, cytopenias	Hashimoto thyroiditis, vitiligo, autoantibodies	NR

Table 1 (*Continued*)

Characteristic features	Zhou *et al.* [5▪▪] Aeschlimann *et al.* [20▪] (*N* = 16)[a]	Kadowaki *et al.* [21▪] Ohnishi *et al.* [24] Shigemura *et al.* [25] Tagaki *et al.* [26] (*N* = 22)	Duncan *et al.* [23] (*N* = 1)	Berteau *et al.* [22] (*N* = 3)	Franco-Jarava *et al.* [27] (*N* = 1)
Other	Possibly related to HA20: Periodontal disease, uterine fibroids, asthma, scoliosis, perinatal lacunar infarction, autism, recurrent bacterial and viral infections, lymphadenopathy, dental crowding secondary to small jaw, geographic tongue	IgA-vasculitis, nephrotic syndrome, chronic hepatitis, cervical lymph adenitis, aseptic meningitis, developmental disorder, craniopharyngeoma, Hodgkin lymphoma, severe local or febrile reaction to antipneumococcal vaccine, pharyngitis	NR	Local inflammatory swelling following antipneumococcal vaccine, pharyngitis	Recurrent respiratory tract infections during infancy, atypical coronary vasculitis Kawasaki-like disease, interstitial pulmonary infiltrates

ALPS, autoimmune lymphoproliferative syndrome; BCG, Bacillus Calmette–Guérin; EN, erythema nodosum; F/M, female/male; GI, gastrointestinal; IBD, inflammatory bowel disease; ITP, idiopathic thrombocytopenic purpura; NR, not reported; SLE, systemic lupus erythematosus.
[a]The initially described cohort by Zhou *et al.* [5▪▪] included 17 patients from seven families. One of these patients was thought to have HA20 based on her symptoms and the positive family history; however, she died years before genetic testing became available. This patient was subsequently excluded from the analysis [5▪▪,20▪].

Acute phase reactants were increased, especially during relapses. Apart from clinical features of autoimmunity, low-titre autoantibodies, such as antinuclear antibodies, anti-dsDNA or lupus anticoagulant, were observed in half of the patients. Although immunodeficiency has been described in other autoinflammatory diseases associated with dysregulation of the ubiquitin pathway [1], only one family was found to have any sign of immunodeficiency (IgG subclass deficiency). HLA-B51 may be positive (two out of five patients tested). Tissue biopsies revealed nonspecific chronic inflammation ($n = 6$ biopsies of various sites) or histologic findings consistent with pustules ($n = 2$), class V lupus nephritis ($N = 1$), or central nervous system (CNS) vasculitis ($N = 1$). The remaining two biopsies showed normal tissue. Thus, histology findings may be indistinguishable from those observed in other autoimmune or inflammatory diseases.

In accordance with the wide spectrum of clinical phenotypes and disease severity, treatment varied. Some patients with a milder, Behçet-like phenotype responded to colchicine, while others required immune-suppressive drugs including cytokine inhibitors (anti-TNF, anti-IL-1), which effectively suppressed systemic inflammation in seven out of nine patients. One patient with a SLE-resembling disease underwent autologous haematopoietic stem cell transplant but required reinitiation of various immunosuppressive agents following disease flare 18 months posttransplant. Given the recent recognition of HA20, clinical outcome has not yet been well investigated, but disease course was characterized by mainly recurrent systemic inflammation; one patient died from uncontrollable disease.

Despite the phenotypic heterogeneity, many patients in the initial cohort presented with a Behçet-like disease. This likely reflects some selection bias, as several cases were identified when additional patients with similar symptoms and a cohort of Turkish and Japanese patients with adult-onset Behçet disease were screened. To date, a large Japanese cohort and several case reports of patients with HA20 have been published, which corroborate the initially described findings and further expand on the clinical phenotype (Table 1) [21▪,22,23–27]. Kadowaki *et al.* [21▪] reported 22 patients from nine unrelated families with a genetic diagnosis of HA20. Although some patients developed a Behçet-like phenotype [24,25], the majority (60%) did not fulfil the International Study Group for Behçet disease 1990 criteria and were at first diagnosed with other autoinflammatory disorders such as PFAPA or familial Mediterranean fever (FMF). Consistent with observations from the initial

cohort [5▪▪,20▪], the disease presentation and severity varied even within families with the same genetic mutation suggesting possible additional effects with other environmental or genetic influences. Interestingly, many patients also revealed autoimmune features such as SLE-like disease, autoimmune hepatitis or Hashimoto thyroiditis. While an increased differentiation of both Th9 and Th17 cells was shown in patients included in the initial cohort [5▪▪], Kadowaki *et al.* [21▪] observed an increased expression of only Th17, but not Th9 cells. The authors therefore suggested a crucial role of Th17 cells in the pathogenesis of HA20. Consistent with this observation, Th17 has been associated with autoimmunity [21▪]. One patient with an autoimmune lymphoproliferative syndrome (ALPS)-like presentation further broadened the phenotypic spectrum [21▪,26]. This patient developed early-onset, recurrent fever, bilateral cervical lymphadenopathy, hepatosplenomegaly and a cutaneous rash suggestive of Kawasaki disease. His immunophenotyping showed an increase in double-negative T cells and a decrease in IgM memory B cells, suggestive of ALPS. But, unlike in ALPS patients, his central memory (TCM), naive, terminally differentiated effector memory T cells re-expressing CD45RA+ (TEMRA) and effector memory (TEM) subpopulations of CD3+ CD8+ T cells were normal [21▪,26]. He failed treatment with cyclosporine but responded to mycophenolate mofetil and prednisolone.

A20 haploinsufficiency was also described in a Spanish boy with psychomotor and growth delay and recurrent autoinflammatory attacks of fever, neutrophilic dermatosis, diarrhoea and mucosal ulcers [27]. Using comparative genomic hybridization (CGH) array, a deletion of 13.13 Mb was identified on chromosome 6 that involved the *TNFAIP3* gene. Thus, this is the first study of a patient with a complete deletion of one of the *TNFAIP3* alleles [27]. Finally, HA20 was reported in a French family with an early-onset, Behçet-like disease [22] and in a British boy with complex autoimmunity characterized by insulin-dependent diabetes, cytopenias, hepatitis, enteropathy and interstitial lung disease [23]. Prior to the diagnosis of HA20, the boy underwent haematopoietic stem cell transplantation and subsequently went into complete remission except for the diabetes [23].

To conclude, HA20 might be considered in patients with an early-onset, dominantly inherited disease with a broad spectrum of autoinflammatory and autoimmune features. The clinical presentation may resemble Behçet disease, JIA, IBD or a periodic fever syndrome and may be associated with recurrent mucosal ulcers, low-titre autoantibodies and a course refractory to standard treatment. Future identification of additional patients with HA20 will likely broaden the phenotypic spectrum.

OTHER INFLAMMATORY DISEASES WITH MUCOSAL INVOLVEMENT

Behçet disease

In contrast to HA20, various pathological pathways seem to be involved in Behçet disease [28]. Important new insights in the pathogenesis have been provided in a recent Immunochip gene-association study [29▪]. Using a large cohort of 1900 Turkish Behçet disease patients, 1779 controls and two replication cohorts of Iranian and Japanese patients, Takeuchi *et al.* [29▪] confirmed the *HLA-B51* locus as the strongest association for Behçet disease and identified new susceptibility loci shared between Behçet disease and IBD. In addition, immune-related risk alleles such as *IL1A-IL1B* and *FUT2* were associated with Behçet disease suggesting a potential role of innate immunity to pathogen response in Behçet disease susceptibility [29▪].

In 2017, the first classification criteria were proposed for paediatric Behçet disease [30]. These were the result of an international collaboration of paediatricians and the analysis of a large prospective cohort of 219 patients. According to these criteria, a child is classified as having Behçet disease if at least three of the following six items are present: recurrent oral aphthosis (at least three attacks per year), genital ulcers, cutaneous involvement (specified as necrotic folliculitis, acneiform lesions or erythema nodosum), ocular involvement (anterior uveitis, posterior uveitis, retinal vasculitis), neurological signs (with exception of isolated headaches) and vascular features (venous or arterial thrombosis or arterial aneurysm) [30]. The classification criteria still require validation by an independent cohort of children with Behçet disease but provide a helpful tool for future therapeutic trials.

Finally, new potential treatments have emerged. In an open-label study of adult Behçet disease patients, the IL-1 blocker anakinra was partially effective in the treatment of resistant oral and genital ulcers [31], and provided further evidence for the implication of the innate immune system in the pathogenesis of Behçet disease. Ustekinumab (anti-IL-12/anti-IL-23) significantly decreased the number of oral ulcers at week 12 in an open-label study of adult Behçet disease patients [32]. Furthermore, apremilast, an oral phospho-diesterase-4 inhibitor modulating several inflammatory pathways, effectively treated oral ulcers in a phase 2, placebo-controlled study of 111 adults with Behçet disease [33]. Despite promising results, the role of

these emerging therapies has yet to be determined for paediatric Behçet disease.

Periodic fever, aphthous stomatitis, pharyngitis and adenitis syndrome

PFAPA is considered to be the most common autoinflammatory disease in childhood [34]. Following several reports of familial clustering, two recent studies investigated the inheritance pattern of patients with PFAPA. Both found an autosomal-dominant inheritance with a reduced penetrance phenotype occurring in many family members [35,36]. This not only suggests a genetic predisposition for PFAPA but also the presence of additional, potentially environmental effects that may explain the observed reduced penetrance phenotype.

Over the past years, various genetic studies sought to identify causative mutations in PFAPA patients. Cheung *et al.* [37] recently found a significant association between polymorphism in *CARD8* and the risk of PFAPA. In a cohort of 82 unrelated PFAPA patients, frameshift mutations in *CARD8* were found in 14% of patients, compared with 3% of healthy controls ($P = 0.012$). Patients carrying *CARD8* mutations had significantly higher prevalence of symptoms outside of flares and oral aphthosis. CARD8 has been shown to negatively regulate the NLRP3 inflammasome activity. The authors found that mutations in CARD8 result in a decreased ability to bind NLRP3 and thus may lead to an increased inflammasome activity and IL-1β production [37].

Mevalonate kinase deficiency

The clinical picture of other autoinflammatory diseases such as hyperimmunoglobulinemia D syndrome (HIDS), the less severe type of mevalonate kinase deficiency (MKD), may also resemble HA20. A retrospective description of 114 MKD/HIDS patients from the Eurofever registry provided detailed information on disease phenotype and genotype [38]. Patients presented with early-onset attacks (86% had recurrent disease, 14% continuous disease with or without exacerbations), fever, gastrointestinal ($n = 112$), mucocutaneous ($n = 99$) and musculoskeletal symptoms ($n = 89$), lymphadenopathy ($n = 102$) and neurologic manifestations ($n = 43$). The study confirmed disease characteristics described in earlier reports, but found a significantly higher prevalence of amyloid A amyloidosis in patients with p.V377I/p.1268T compound heterozygosity [38]. An interesting study by Park *et al.* [39] elucidated an unexpected molecular connection between HIDS and FMF. The authors showed that the pyrin inflammasome is regulated by RhoA-dependent phosphorylation of pyrin and pyrin's following interaction with 14-3-3 proteins. Defects in prenylation, seen in HIDS, lead to RhoA inactivation and subsequent pyrin inflammasome activation [39].

Inflammatory bowel disease

Besides HA20, TGF-β1 deficiency and ALPI deficiency were recently recognized as monogenenic causes of severe infantile IBD [40,41]. Biallelic loss-of-function mutations in the transforming growth factor (*TGF*)-*β1* gene were found to cause early-onset IBD and encephalopathy [40]. This study showed the critical and nonredundant role of TGF-β1 in the development and homeostasis of intestinal immunity and the CNS in humans [40]. Parlato *et al.* [41] reported compound heterozygous loss-of-function mutations in *ALPI* as a cause of severe intestinal inflammation and autoimmunity. This study emphasized the crucial role of ALPI (alkaline phosphatase, intestine) in the maintenance of gut homeostasis and provided strong rationale for oral ALPI treatment not only in patients with monogenic ALPI deficiency but also common polygenic forms of IBD [41].

Studies of human gut microbiota provided insights into the pathogenesis of various chronic inflammatory diseases, especially IBD. The Integrative Human Microbiome Project examined the dynamics of the gut microbiome in children and adults with IBD (Crohn disease $n = 59$, ulcerative colitis $n = 34$) and 24 controls over a 1-year period [42]. The authors found that some bacterial species were universally present, and that loss of these organisms in disease results in dysbiosis with more far-reaching consequences than previously thought. This suggests a critical role of these specific organisms in the maintenance of gut health. Other species were metagenomically present, but had low or nonexistent gene expression, indicating that they are inactive in the gut. Finally, the authors described disease-specific microbial characteristics that were more pronounced or only detectable at the transcript level. These results provided novel insights into the pathogenic role of gut microbial dysfunction in IBD [42,43].

CONCLUSION

HA20 is characterized by an early-onset systemic inflammation and a wide range of inflammatory and autoimmune features with variable disease severity. In light of the clinical data available so far, genetic testing for mutations in *TNFAIP3* may

be considered in patients presenting with a dominantly inherited, early-onset inflammatory disorder that may resemble Behçet disease or other autoinflammatory and autoimmune diseases, particularly those associated with mucosal involvement. Future studies will help to further describe the clinical phenotypes of patients with HA20.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Aksentijevich I, Zhou Q. NF-kappaB pathway in autoinflammatory diseases: dysregulation of protein modifications by ubiquitin defines a new category of autoinflammatory diseases. Front Immunol 2017; 8:399.
2. Komander D, Clague MJ, Urbe S. Breaking the chains: structure and function of the deubiquitinases. Nat Rev Mol Cell Biol 2009; 10:550–563.
3. Harhaj EW, Dixit VM. Regulation of NF-kappaB by deubiquitinases. Immunol Rev 2012; 246:107–124.
4. Aksentijevich I, McDermott MF. Lessons from characterization and treatment of the autoinflammatory syndromes. Curr Opin Rheumatol 2017; 29:187–194.
5. ■■ Zhou Q, Wang H, Schwartz DM, *et al.* Loss-of-function mutations in TNFAIP3 leading to A20 haploinsufficiency cause an early-onset autoinflammatory disease. Nat Genet 2016; 48:67–73.

This article describes a new monogenic autoinflammatory disease.

6. Boonyasrisawat W, Eberle D, Bacci S, *et al.* Tag polymorphisms at the A20 (TNFAIP3) locus are associated with lower gene expression and increased risk of coronary artery disease in type 2 diabetes. Diabetes 2007; 56:499–505.
7. Fung EY, Smyth DJ, Howson JM, *et al.* Analysis of 17 autoimmune disease-associated variants in type 1 diabetes identifies 6q23/TNFAIP3 as a susceptibility locus. Genes Immun 2009; 10:188–191.
8. Nair RP, Duffin KC, Helms C, *et al.* Genome-wide scan reveals association of psoriasis with IL-23 and NF-kappaB pathways. Nat Genet 2009; 41:199–204.
9. Thomson W, Barton A, Ke X, *et al.* Rheumatoid arthritis association at 6q23. Nat Genet 2007; 39:1431–1433.
10. Trynka G, Zhernakova A, Romanos J, *et al.* Coeliac disease-associated risk variants in TNFAIP3 and REL implicate altered NF-kappaB signalling. Gut 2009; 58:1078–1083.
11. Wellcome Trust Case Control Consortium. Genome-wide association study of 14,000 cases of seven common diseases and 3,000 shared controls. Nature 2007; 447:661–678.
12. Beyaert R, Heyninck K, Van Huffel S. A20 and A20-binding proteins as cellular inhibitors of nuclear factor-kappa B-dependent gene expression and apoptosis. Biochem Pharmacol 2000; 60:1143–1151.
13. Matmati M, Jacques P, Maelfait J, *et al.* A20 (TNFAIP3) deficiency in myeloid cells triggers erosive polyarthritis resembling rheumatoid arthritis. Nat Genet 2011; 43:908–912.
14. Vereecke L, Sze M, Mc Guire C, *et al.* Enterocyte-specific A20 deficiency sensitizes to tumor necrosis factor-induced toxicity and experimental colitis. J Exp Med 2010; 207:1513–1523.
15. Chu Y, Vahl JC, Kumar D, *et al.* B cells lacking the tumor suppressor TNFAIP3/A20 display impaired differentiation and hyperactivation and cause inflammation and autoimmunity in aged mice. Blood 2011; 117:2227–2236.
16. Tavares RM, Turer EE, Liu CL, *et al.* The ubiquitin modifying enzyme A20 restricts B cell survival and prevents autoimmunity. Immunity 2010; 33:181–191.
17. Lee EG, Boone DL, Chai S, *et al.* Failure to regulate TNF-induced NF-kappaB and cell death responses in A20-deficient mice. Science 2000; 289: 2350–2354.
18. Duong BH, Onizawa M, Oses-Prieto JA, *et al.* A20 restricts ubiquitination of pro-interleukin-1beta protein complexes and suppresses NLRP3 inflammasome activity. Immunity 2015; 42:55–67.
19. Honma K, Tsuzuki S, Nakagawa M, *et al.* TNFAIP3/A20 functions as a novel tumor suppressor gene in several subtypes of non-Hodgkin lymphomas. Blood 2009; 114:2467–2475.
20. ■ Aeschlimann FA, Batu ED, Canna SW, *et al.* A20 haploinsufficiency (HA20): clinical phenotypes and disease course of patients with a newly recognised NF-kB-mediated autoinflammatory disease. Ann Rheum Dis 2018; 77:728–735.

This article provides a detailed description of the clinical presentation of patients with A20 haploinsufficiency.

21. ■ Kadowaki T, Ohnishi H, Kawamoto N, *et al.* Haploinsufficiency of A20 causes autoinflammatory and autoimmune disorders. J Allergy Clin Immunol 2018; 141:1485–1488.

This article reports the largest cohort of A20 haploinsufficiency to date with additional clinical and laboratory data.

22. Berteau F, Rouvière B, Nau A. Response to: 'A20 haploinsufficiency (HA20): clinical phenotypes and disease course of patients with a newly recognised NF-kB-mediated autoinflammatory disease'. Ann Rheum Dis 2018. [Epub ahead of print]
23. Duncan CJA, Dinnigan E, Theobald R, *et al.* Early-onset autoimmune disease due to a heterozygous loss-of-function mutation in TNFAIP3 (A20). Ann Rheum Dis 2018; 77:783–786.
24. Ohnishi H, Kawamoto N, Seishima M, *et al.* A Japanese family case with juvenile onset Behcet's disease caused by TNFAIP3 mutation. Allergol Int 2017; 66:146–148.
25. Shigemura T, Kaneko N, Kobayashi N, *et al.* Novel heterozygous C243Y A20/TNFAIP3 gene mutation is responsible for chronic inflammation in autosomal-dominant Behcet's disease. RMD Open 2016; 2:e000223.
26. Takagi M, Ogata S, Ueno H, *et al.* Haploinsufficiency of TNFAIP3 (A20) by germline mutation is involved in autoimmune lymphoproliferative syndrome. J Allergy Clin Immunol 2017; 139:1914–1922.
27. Franco-Jarava C, Wang H, Martin-Nalda A, *et al.* TNFAIP3 haploinsufficiency is the cause of autoinflammatory manifestations in a patient with a deletion of 13Mb on chromosome 6. Clin Immunol 2018; 191:44–51.
28. Yazici H, Seyahi E, Hatemi G, Yazici Y. Behcet syndrome: a contemporary view. Nat Rev Rheumatol 2018; 14:119.
29. ■ Takeuchi M, Mizuki N, Meguro A, *et al.* Dense genotyping of immune-related loci implicates host responses to microbial exposure in Behcet's disease susceptibility. Nat Genet 2017; 49:438–443.

This study extends on shared susceptibility genes for Behçet disease and Crohn disease and reports a number of immune-related loci, thereby providing novel insights into the pathogenesis of Behçet disease.

30. Kone-Paut I, Shahram F, Darce-Bello M, *et al.* Consensus classification criteria for paediatric Behcet's disease from a prospective observational cohort: PEDBD. Ann Rheum Dis 2016; 75:958–964.
31. Grayson PC, Yazici Y, Merideth M, *et al.* Treatment of mucocutaneous manifestations in Behcet's disease with anakinra: a pilot open-label study. Arthritis Res Ther 2017; 19:69.
32. Mirouse A, Barete S, Monfort JB, *et al.* Ustekinumab for Behcet's disease. J Autoimmun 2017; 82:41–46.
33. Hatemi G, Melikoglu M, Tunc R, *et al.* Apremilast for Behcet's syndrome: a phase 2, placebo-controlled study. N Engl J Med 2015; 372:1510–1518.
34. Harel L, Hashkes PJ, Lapidus S, *et al.* The first international conference on periodic fever, aphthous stomatitis, pharyngitis, adenitis syndrome. J Pediatr 2018; 193:265–274; e3.
35. Di Gioia SA, Bedoni N, von Scheven-Gete A, *et al.* Analysis of the genetic basis of periodic fever with aphthous stomatitis, pharyngitis, and cervical adenitis (PFAPA) syndrome. Sci Rep 2015; 5:10200.
36. Manthiram K, Nesbitt E, Morgan T, Edwards KM. Family history in periodic fever, aphthous stomatitis, pharyngitis, adenitis (PFAPA) syndrome. Pediatrics 2016; 138:pii: e20154572.
37. Cheung MS, Theodoropoulou K, Lugrin J, *et al.* Periodic fever with aphthous stomatitis, pharyngitis, and cervical adenitis syndrome is associated with a CARD8 variant unable to bind the NLRP3 inflammasome. J Immunol 2017; 198:2063–2069.
38. Ter Haar NM, Jeyaratnam J, Lachmann HJ, *et al.* The phenotype and genotype of mevalonate kinase deficiency: a series of 114 cases from the Eurofever registry. Arthritis Rheumatol 2016; 68:2795–2805.
39. Park YH, Wood G, Kastner DL, Chae JJ. Pyrin inflammasome activation and RhoA signaling in the autoinflammatory diseases FMF and HIDS. Nat Immunol 2016; 17:914–921.
40. Kotlarz D, Marquardt B, Baroy T, *et al.* Human TGF-beta1 deficiency causes severe inflammatory bowel disease and encephalopathy. Nat Genet 2018; 50:344–348.
41. Parlato M, Charbit-Henrion F, Pan J, *et al.* Human ALPI deficiency causes inflammatory bowel disease and highlights a key mechanism of gut homeostasis. EMBO Mol Med 2018; 10:pii: e8483.
42. Schirmer M, Franzosa EA, Lloyd-Price J, *et al.* Dynamics of metatranscription in the inflammatory bowel disease gut microbiome. Nat Microbiol 2018; 3:337–346.
43. Lavelle A, Sokol H. Gut microbiota: beyond metagenomics, metatranscriptomics illuminates microbiome functionality in IBD. Nat Rev Gastroenterol Hepatol 2018; 15:193–194.

Systemic juvenile idiopathic arthritis and macrophage activation syndrome: update on pathogenesis and treatment

Shima Yasin and Grant S. Schulert

Purpose of review
The past decade has seen substantial progress in defining the cause and pathogenesis of the chronic childhood arthropathy systemic juvenile idiopathic arthritis (SJIA) and its related complication macrophage activation syndrome (MAS). The purpose of this review is to describe and synthesize advances in this field, particularly since 2016, with the potential to transform clinical practice.

Recent findings
Newly developed MAS classification criteria have been further studied and validated in other diseases and populations, as well as a recently proposed score to distinguish MAS from familial hemophagocytic lymphohistiocytosis. There has also been substantial progress toward understanding the genetic underpinnings of SJIA and MAS, both through targeted study of specific genes and the results of a large genome-wide association study. The immunopathogenesis of SJIA has been further elucidated through several studies regarding the proinflammatory cytokines interleukin-18, interferon (IFN)γ, and how their interplay impacts emergence of MAS. Finally, big data studies integrating genomic information with immunophenotypes have potential to provide novel insights into disease mechanisms in SJIA.

Summary
Collectively, these research advances have significant implications regarding the classification and diagnosis of SJIA and MAS, and support a next generation of biologic treatments including kinase inhibitors and targeted interleukin-18 or IFNγ blockade.

Keywords
autoinflammatory, IFNγ, interleukin-18, pediatric rheumatology

INTRODUCTION

Systemic juvenile idiopathic arthritis (SJIA) is a distinctive and severe form of chronic childhood arthritis, distinguished from other subtypes of juvenile idiopathic arthritis (JIA) by its predominance of systemic inflammation and extraarticular features, including daily spiking fevers, rash, lymphadenopathy, hepatosplenomegaly, and serositis [1]. Many rheumatologists now view SJIA as having features of an autoinflammatory disorder, with substantial innate immune dysregulation, and less resembling an autoimmune disease as characterizes other subtypes of JIA [2,3]. SJIA is also distinguished by the risk of patients to develop macrophage activation syndrome (MAS), a life-threatening episode of overwhelming inflammation, with fevers, cytopenias, coagulopathy, liver, and central nervous system dysfunction, and a systemic cytokine storm [4]. MAS has strong phenotypic similarity to the rare genetic disorders hemophagocytic lymphohistiocytosis (HLH), where defects in perforin granule mobilization causes impaired lymphocyte cytolytic activity [5]. Although MAS can occur in the setting of a broad spectrum of rheumatic diseases, in pediatric rheumatology it is most commonly reported in SJIA [6].

The past decade has seen substantial progress in defining the immunopathogenesis of SJIA and MAS, which has in turn supported the introduction of

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KEY POINTS

- Newly developed classification criteria for MAS in SJIA have been further studied and validated in other diseases and populations, showing both utility and key limitations.
- Large genome-wide association studies in SJIA clearly show that this disease is genetically distinct from other forms of JIA.
- Several studies support key roles for the proinflammatory cytokines interleukin-18, IFNγ in the immunopathogenesis of SJIA.

cytokine targeted therapy against interleukin-1 and interleukin-6, transforming the management of SJIA. However, substantial questions remain regarding the genetic basis of immune dysfunction in SJIA, the optimal clinical management strategy, and cause and approach to MAS. In this review, we will examine several key recent advances in SJIA and MAS, with a primary focus on new findings published since 2016.

DIAGNOSING AND DISTINGUISHING MACROPHAGE ACTIVATION SYNDROME

There is no single diagnostic marker for MAS in SJIA, and early diagnosis requires high index of suspicion and relies on a combination of clinical features and laboratory findings. In 2016, new classification criteria were developed by the European League Against Rheumatism/American College of Rheumatology/Pediatric Rheumatology International Trials Organization collaborative (Table 1) [7]. Shimizu and colleagues performed a validation study with these criteria utilizing 65 Japanese patients with and without MAS [8]. This study demonstrated very high diagnostic performance in full-blown MAS, but only 63.6% sensitivity at MAS onset. Ahn and colleagues evaluated the clinical significance of this criteria designed for SJIA patients in adult onset still's disease (AOSD) [9▪]. In a retrospective review of 26 patients with bone marrow results consistent with MAS, only 18 were classified as MAS. When comparing prognosis of both groups, in-hospital mortality was only observed in patients classified as MAS.

Due to resemblance in pathogenesis, clinical, and laboratory features, differentiation between MAS and familial HLH or other diseases can be challenging, and as such several groups initiated development and validation of new diagnostic scores for MAS. Focusing on MAS in SJIA patients, Minoia and colleagues developed MAS/familial HLH (MH) score to assist with differentiation from familial HLH (Table 1) [10▪]. The validity was subsequently tested, where Minoia and colleagues proposed an MH score of at least 60 as cutoff for familial HLH. Interestingly, the study found that the average values of ferritin and lactate dehydrogenase (LDH) were higher in MAS compared with familial HLH. Separately, Batu and colleagues [11] evaluated the previously developed Hscore for secondary HLH for use in rheumatic diseases (Table 1) [12]. This

Table 1. Diagnostic and classification scores for MAS, secondary HLH, and familial HLH.

2016 MAS Classification Criteria [7]	HScore for secondary HLH [12]	MH Score to differentiate MAS from familial HLH [10▪]
A febrile patient with known or suspected SJIA is classified as having MAS if:	Known immunosuppression: 0 (no) or 18 (yes)	Age of onset: 0 (>1.6 years), 37 (≤1.6)
(1) Ferritin >684 ng/ml	Fever: 0 (<38.4), 33 (38.4–39.4), 49 (>39.4)	PMN count: 0 (>1.4×10^9/l), 37 (≤1.4)
AND	Organomegaly: 0 (no), 23 (liver or spleen), 38 (both)	Fibrinogen: 0 (>131 mg/dl), 15 (≤131)
(2) Any 2 of the following:	Cytopenias: 0 (0–1 line), 24 (2 lines), 34 (3 lines)	Splenomegaly: 0 (no), 12 (yes)
Platelets ≤181×10^9/l	Ferritin: 0 (<2000 ng/ml), 35 (2–6000), 50 (>6000)	Platelets: 0 (>78×10^9/l), 11 (≤78)
AST >48 U/l	Triglycerides[a]: 0 (<133 mg/dl), 44 (133–354), 64 (>354)	Hemoglobin: 0 (>8.3 g/dl), 11 (≤8.3)
Triglycerides >156 mg/dl	Fibrinogen: 0 (>250 mg/dl), 30 (≤250)	Best cutoff: 60 (≥60 indicative of familial HLH)
Fibrinogen ≤360 mg/dl	AST: 0 (<30 U/l), 19 (≥30)	
	Hemophagocytosis: 0 (no) 35 (yes)	
	Best cutoff: 169 (≥169 indicative of reactive HLH)	

[a]Triglycerides reported as mmoles/l; converted to mg/dl for comparison.

work identified better sensitivity and specificity when using a higher cutoff at 190.5 compared with 169 proposed by the original study.

Use of cytokine-targeted biologics against interleukin-1 and interleukin-6 poses another challenge when applying MAS diagnostic criteria. It has been suggested by some studies that use cytokine-targeted biologics namely tocilizumab might mask MAS symptoms [13], but it remains unknown whether these medications alter clinical and laboratory features used for classification criteria. With this in mind, a recent comprehensive literature review evaluated the performance of 2016 MAS classification criteria in patients with SJIA who developed MAS while on biologic therapy [14▪]. The study demonstrated that patients treated with tocilizumab were less likely to meet 2016 classification criteria when having MAS, as they were less likely to be febrile and had lower ferritin levels in comparison to historic cohorts. Likewise, patients treated with canakinumab had lower ferritin levels but had no difference in clinical features in comparison to historic prebiologic cohorts.

MAS in rheumatic diseases is not strictly associated with SJIA or AOSD, but rather has been reported in several other autoimmune conditions including systemic lupus erythematosus (SLE) [15]. In a recent study, Borgia and colleagues studied features and treatment of MAS in childhood-SLE (cSLE) [16]. They observed higher rates of cytopenias and elevated dsDNA but no difference in cSLE features between those who developed MAS compared with patients without MAS. Also, concomitant diagnosis of SLE and MAS carried higher mortality rate in the cohort. Liu and colleagues demonstrated similar result in their case–control study, where fever was the most common clinical feature [17]. This study highlighted the importance of LDH in addition to hyperferritenemia, with optimal cutoff values for diagnosis using ferritin less than 662.5 ng/ml and LDH less than 359 U/ml. Finally, Ahn and colleagues evaluated the use of 2016 MAS classification criteria in patients with febrile SLE [18] and found that those classified as MAS had significantly higher mortality rate. This demonstrates both the importance of considering MAS in febrile SLE patients and usefulness of using the 2016 classification criteria in identifying patients at high risk of poor outcome.

GENETICS OF SYSTEMIC JUVENILE IDIOPATHIC ARTHRITIS AND MACROPHAGE ACTIVATION SYNDROME

Historically, genetic investigations into SJIA have been limited by the relatively small number of patients studied, often from a single geographic region, and as such have offered inconsistent results that in general have not been reproduced or validated. To overcome this challenge and examine SJIA genetic risks in an unbiased manner, the International Childhood Arthritis Genetics (INCHARGE) consortium performed a genome-wide association study using DNA from 982 SJIA patients from multiple continents. Initial results of this effort were reported in 2015, and for the first time established the major histocompatability complex (MHC) class II locus as a bona fide risk allele for SJIA [19]. These findings implicate a role for adaptive immunity in disease pathogenesis and challenge the emerging paradigm of SJIA as a purely autoinflammatory condition [2]. Ombrello and INCHARGE colleagues recently followed-up those initial findings with a more detailed description of the genetic architecture of SJIA [20▪▪]. Here, they identified 24 additional genetic loci with significant or suggestive genome-wide association with SJIA. The strongest non-MHC association was found in an intergenic region of chromosome 1, which overlapped a large cluster of transcription factor-binding sites. Other suggestive associations were found near genes encoding the histone deacetylase *HDAC9*, transcription factors including *KLF17*, and several zinc-finger proteins. Notably, none of these loci have been previously implicated in inflammatory arthritis and utilizing several approaches the authors failed to identify any evidence of shared genetic architecture between SJIA and other forms of JIA. These genetic findings have recently been integrated with clinical and demographic data to support the distinction of SJIA and its adult counterpart AOSD from other chronic arthropathies [21]. Taken together, these findings by Ombrello and colleagues offer a roadmap for further pathophysiologic research into the cause of SJIA and support its reclassification as a distinct entity from other forms of JIA.

Along with such robust, genome-wide approaches to the genetics of SJIA, a recently described monogenic cause of SJIA may also hold clues to disease pathogenesis. In 2015, Wakil and colleagues reported five consanguineous families with familial recurrence of classic SJIA, all found to have recessively inherited variants in *LACC1* (*c13orf31*) [22]. Subsequently *LACC1* variants have been reported in children with both systemic and nonsystemic JIA, as well as inflammatory bowel disease [23,24]. *LACC1* encodes a protein termed fatty acid metabolism-immunity nexus (FAMIN) which functions as a central metabolic regulator for macrophages [25]. Recent work has further demonstrated that FAMIN associates with the pattern recognition receptor NOD2, where it has key roles in optimizing innate immune signaling [26].

Finally, the variability in risk for MAS among patients with SJIA—a majority never experience overt MAS, whereas others have recurrent episodes—strongly supports a genetic component for this complication. This is mostly strikingly observed in children with gain-of-function variants in the NLRC4 inflammasome (see below). However, several studies have found that approximately one-third of patients with MAS in the setting of SJIA carry rare heterozygous variants in causative genes for familial HLH, such as perforin and related granule trafficking proteins [27–30]. This notion is expanded further by the recent demonstration of functional noncoding variants in *UNC13D* in patients with recurrent MAS [31]. Several recent studies have also shown that such hypomorphic variants in HLH-associated genes may lead to partial loss of lymphocyte cytolytic function [31–33], seemingly lowering the threshold for MAS in inflammatory settings such as SJIA. Future studies are needed to show in a prospective fashion whether such genetic variants can indeed predict risk for MAS in children with SJIA.

INTERLEUKIN-18, INTERFERONγ, AND PATHOGENESIS OF MACROPHAGE ACTIVATION SYNDROME

Cytokines of the interluekin-1 family are felt to be central to the pathogenesis of SJIA, most clearly illustrated by the dramatic response in many patients to therapies such as recombinant interleukin-1RA (anakinra). There is accumulating evidence for a key role in particular for interleukin-18 as driver of both SJIA and potentially its association with MAS. This was vividly illustrated by the recent description of patients carrying gain-of-function mutations in the inflammasome NLRC4, leading to persistently elevated interleukin-18 levels and recurrent MAS [34,35]. Several recent reports have demonstrated markedly elevated serum interleukin-18 in children with SJIA. Brachat and colleagues examined early transcriptional and biomarker changes in SJIA patients upon treatment with canakinumab [36▪]. Although serum interleukin-6 and inflammatory gene expression profiles markedly improved as soon as 3 days after treatment initiation, serum interleukin-18 remained persistently elevated, with only mild decreases after 2 months of treatment. In addition, there were no significant differences in interleukin-18 levels between patients with active and inactive SJIA up to day 197 of treatment, suggesting persistent interleukin-18 levels disconnected from disease activity status. These findings are expanded by Canna and colleagues who find similarly high levels of interleukin-18 in SJIA, but even further elevated in the setting of MAS [37▪▪]. More strikingly, they find that the natural antagonist interleukin-18-binding protein is only modestly higher in MAS, leading to elevated free interleukin-18 in both SJIA and in particular MAS. This free interleukin-18 elevation is also linked pathologically to MAS, as mice overexpressing interleukin-18 had more severe disease in the TLR9 mouse model of MAS [37▪▪]. Similarly, mice lacking interleukin-18-binding protein had severe manifestations of MAS including cytopenias, hyperferritinemia, and bone marrow hemophagocytosis [38▪], further supporting a central role for interleukin-18 signaling in MAS pathogenesis. This is highlighted by the recent successful use of recombinant interleukin-18-binding protein in the treatment of a child with recurrent MAS due to NLRC4 gain-of-function mutation [39], as well as the results of a small phase 2 trial in AOSD [40].

How mechanistically does interleukin-18 drive inflammation in SJIA and MAS? Recent work has shown that along with elevated interleukin-18, children with active SJIA have high serum interleukin-17A, a key mediator of chronic arthritis [41]. Indeed, γ/δ T cells from SJIA patients exhibited elevated levels of interleukin-17 production, which was inhibited by anti-interleukin-1 treatment. High levels of interleukin-18 could drive interleukin-17 production from donor γ/δ T cells, in particular when combined with other SJIA-associated inflammatory mediators such as interleukin-1β and S100A12. This work suggests a mechanism through which interleukin-18 production could lead to a chronic, T-cell-mediated arthritis in children with SJIA.

However, the pathologic role for interleukin-18 in MAS may be linked to its function as a driver of IFNγ production. Although there is little evidence to support IFNγ production in SJIA without MAS, it is increasingly clear that IFNγ is central to the emergence of MAS in SJIA [4]. There is strong evidence for production of IFNγ itself and IFNγ-induced proteins in the tissue during MAS [42,43]. A recent large international study of SJIA patients found that serum levels of IFNγ and IFNγ-induced chemokines such as CXCL9 were significantly higher in SJIA patients with active MAS than in patients with active SJIA without signs of MAS [44▪]. Indeed, IFNγ may be a useful therapeutic target in MAS as well. Prencipe *et al.* examined this in mice transgenic for human interleukin-6, which when challenged with lipopolysaccharide manifest clinical features of MAS [45]. These mice also showed substantial activation of the IFNγ pathway as determined by *ifng* gene expression, STAT1 activation in the tissues, and IFNγ-induced chemokine production. Treatment with an anti-IFNγ monoclonal antibody reversed this activation, as well as ameliorated clinical and laboratory features of MAS. Supported by these and

other findings, clinical trials of an anti-IFNγ monoclonal antibody in MAS have recently begun (NCT03311854).

The precise relationship between interleukin-18, IFNγ production, and emergence of MAS in patients with SJIA remains to be determined. In particular, if children with SJIA have marked and chronic elevations in interleukin-18, why does only a subset develop MAS? de Jager and colleagues have reported that natural killer (NK) cells from children with SJIA failed to produce IFNγ upon interleukin-18 stimulation, due in part to impaired interleukin-18 receptor phosphorylation [46]. These findings were recently confirmed and expanded by Put *et al.*, who showed NK cells from SJIA patients had defective interleukin-18-induced IFNγ production along with other alterations in balance between activating and inhibitory receptors [47▪]. Together this supports a model where in SJIA, NK cells (and possibly other immune effectors) are refractory to effects of interleukin-18 including IFNγ production, but this somehow is altered in MAS (Fig. 1). Indeed, Canna and colleagues have recently demonstrated that compared with familial or infection-triggered HLH, MAS is distinguished by unusually high ratios of interleukin-18 to CXCL9 (as biomarker of IFNγ activity) [37▪▪], supporting a hypothesis of immune cells refractory to interleukin-18.

CONCLUSION

The last several years have seen tremendous advances in understanding of the genetics and pathogenesis of SJIA and MAS. In turn, these findings are leading to better approaches to classification and diagnosis, and new targeted treatment approaches particularly toward MAS. However, significant questions remain unanswered regarding the immunopathogenesis of SJIA, and in particular how environmental stimuli interact with genetic risk factors to trigger disease. Future approaches that integrate functional, genomic, and epigenetic information will be highly informative in this regard. Cepika and colleagues recently used large data sets such as this to probe the biology of SJIA [48▪▪]. Here, transcriptional profiling, flow cytometry, and multiplex cytokine analysis was integrated to define distinct modules of immune response traits to a variety of inflammatory stimuli. These modules and traits were then utilized to identify immune differences between blood from SJIA patients with inactive disease and controls. Further work showed that sorted monocytes from patients with inactive SJIA had altered expression of several interleukin-1β regulatory proteins. Taken together, approaches such as this demonstrate how combining experimental and computational resources can be used to dissect the pathogenesis of SJIA and MAS. Future approaches, particularly those integrating

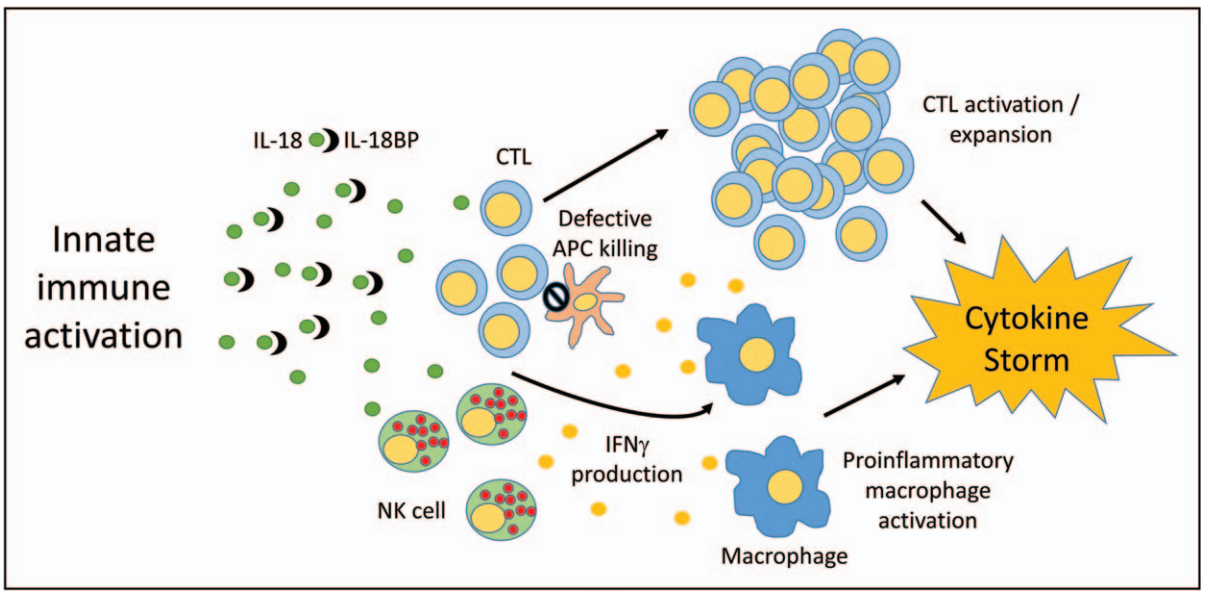

FIGURE 1. Interleukin-18, IFNγ, and emergence of MAS. Innate immune activation in SJIA is associated with markedly elevated interleukin-18. Even during active disease, this is balanced by high levels of interleukin-18-binding protein, and NK cell cytokine hyporesponsiveness. However, emergence of MAS is associated with further interleukin-18 increases, overwhelming interleukin-18-binding protein and allowing substantial free interleukin-18. This, possibly along with other factors including defective antigen-presenting cell (APC) killing, drives IFNγ production from NK cells and cytotoxic lymphocytes (CTL). In turn, IFNγ induces proinflammatory activation of macrophages in tissues including liver and bone marrow. Together with CTL activation and proliferation, this macrophage activation can lead to a 'cytokine storm' and fulminant MAS.

recent genomic findings, may lead the way to untangle the pathogenesis of this life-threatening disorder.

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Conflicts of interest

G.S.S. received consulting fees from Novartis. S.Y. has no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Petty RE, Southwood TR, Manners P, *et al.* International League of Associations for Rheumatology classification of juvenile idiopathic arthritis: second revision, Edmonton, 2001. J Rheumatol 2004; 31:390–392.
2. Nigrovic PA. Autoinflammation and autoimmunity in systemic juvenile idiopathic arthritis. Proc Natl Acad Sci USA 2015; 112:15785–15786.
3. Mellins ED, Macaubas C, Grom AA. Pathogenesis of systemic juvenile idiopathic arthritis: some answers, more questions. Nat Rev Rheumatol 2011; 7:416–426.
4. Schulert GS, Grom AA. Pathogenesis of macrophage activation syndrome and potential for cytokine- directed therapies. Annu Rev Med 2015; 66:145–159.
5. Filipovich AH. The expanding spectrum of hemophagocytic lymphohistiocytosis. Curr Opin Allergy Clin Immunol 2011; 11:512–516.
6. Grom AA, Horne A, De Benedetti F. Macrophage activation syndrome in the era of biologic therapy. Nat Rev Rheumatol 2016; 12:259–268.
7. Ravelli A, Minoia F, Davì S, *et al.* 2016 Classification Criteria for Macrophage Activation Syndrome Complicating Systemic Juvenile Idiopathic Arthritis: a European League Against Rheumatism/American College of Rheumatology/Paediatric Rheumatology International Trials Organisation Collaborat. Arthritis Rheumatol (Hoboken, NJ) 2016; 68:566–576.
8. Shimizu M, Mizuta M, Yasumi T, *et al.* Validation of classification criteria of macrophage activation syndrome in Japanese patients with systemic juvenile idiopathic arthritis. Arthritis Care Res (Hoboken) 2017; DOI: 10.1002/acr.23482.
9. ■ Ahn SS, Yoo B-W, Jung SM, *et al.* Application of the 2016 EULAR/ACR/PRINTO Classification Criteria for Macrophage Activation Syndrome in Patients with Adult-onset Still Disease. J Rheumatol 2017; 44:996–1003.

This study discussed the expansion of 2016 MAS criteria beyond sJIA and its use in AOSD in order to identify patients at high risk for a poor outcome.

10. ■ Minoia F, Bovis F, Davì S, *et al.* Development and Initial Validation of the Macrophage Activation Syndrome/Primary Hemophagocytic Lymphohistiocytosis Score, a diagnostic tool that differentiates primary hemophagocytic lymphohistiocytosis from macrophage activation syndrome. J Pediatr 2017; 189:72–78.

The authors developed and validated a criteria by which to discriminate primary HLH and MAS.

11. Batu ED, Erden A, Seyhoğlu E, *et al.* Assessment of the HScore for reactive haemophagocytic syndrome in patients with rheumatic diseases. Scand J Rheumatol 2017; 46:44–48.
12. Fardet L, Galicier L, Lambotte O, *et al.* Development and validation of the HScore, a score for the diagnosis of reactive hemophagocytic syndrome. Arthritis Rheumatol (Hoboken, NJ) 2014; 66:2613–2620.
13. Shimizu M, Nakagishi Y, Kasai K, *et al.* Tocilizumab masks the clinical symptoms of systemic juvenile idiopathic arthritis-associated macrophage activation syndrome: the diagnostic significance of interleukin-18 and interleukin-6. Cytokine 2012; 58:287–294.
14. ■ Schulert GS, Minoia F, Bohnsack J, *et al.* Effect of biologic therapy on clinical and laboratory features of macrophage activation syndrome associated with systemic juvenile idiopathic arthritis. Arthritis Care Res 2018; 70:409–419.

This comprehensive literature review clearly demonstrates the changes in the natural history of the SJIA and MAS and alterations in clinical and laboratory features after use of cytokine targeted biologics.

15. Stephan JL, Kone-Paut I, Galambrun C, *et al.* Reactive haemophagocytic syndrome in children with inflammatory disorders. A retrospective study of 24 patients. Rheumatol 2001; 40:1285–1292.
16. Borgia RE, Gerstein M, Levy DM, *et al.* Features, treatment, and outcomes of macrophage activation syndrome in childhood-onset systemic lupus erythematosus. Arthritis Rheumatol (Hoboken, NJ) 2018; 70:616–624.
17. Liu A-C, Yang Y, Li M-T, *et al.* Macrophage activation syndrome in systemic lupus erythematosus: a multicenter, case-control study in China. Clin Rheumatol 2018; 37:93–100.
18. Ahn SS, Yoo B-W, Jung SM, *et al.* In-hospital mortality in febrile lupus patients based on 2016 EULAR/ACR/PRINTO classification criteria for macrophage activation syndrome. Semin Arthritis Rheum 2017; 47:216–221.
19. Ombrello MJ, Remmers EF, Tachmazidou I, *et al.* HLA-DRB1*11 and variants of the MHC class II locus are strong risk factors for systemic juvenile idiopathic arthritis. Proc Natl Acad Sci USA 2015; 112:15970–15975.
20. ■■ Ombrello MJ, Arthur VL, Remmers EF, *et al.* Genetic architecture distinguishes systemic juvenile idiopathic arthritis from other forms of juvenile idiopathic arthritis: clinical and therapeutic implications. Ann Rheum Dis 2017; 76:906–913.

This further analysis of the INCHARGE GWAS described 24 genetic loci with significant or suggestive associations with SJIA. In general, these loci had not previously been associated with inflammatory arthritis, providing strong genetic evidence for a clear distinction between SJIA and other forms of JIA.

21. Nigrovic PA, Raychaudhuri S, Thompson SD. Review: genetics and the classification of arthritis in adults and children. Arthritis Rheumatol 2018; 70:7–17.
22. Wakil SM, Monies DM, Abouelhoda M, *et al.* Association of a mutation in lacc1 with a monogenic form of systemic juvenile idiopathic arthritis. Arthritis Rheumatol 2015; 67:288–295.
23. Kallinich T, Thorwarth A, von Stuckrad S-L, *et al.* Juvenile arthritis caused by a novel FAMIN (LACC1) mutation in two children with systemic and extended oligoarticular course. Pediatr Rheumatol 2016; 14:63.
24. Assadi G, Saleh R, Hadizadeh F, *et al.* LACC1 polymorphisms in inflammatory bowel disease and juvenile idiopathic arthritis. Genes Immun 2016; 17:261–264.
25. Cader MZ, Boroviak K, Zhang Q, *et al.* C13orf31 (FAMIN) is a central regulator of immunometabolic function. Nat Immunol 2016; 17:1046–1056.
26. Lahiri A, Hedl M, Yan J, Abraham C. Human LACC1 increases innate receptor-induced responses and a LACC1 disease-risk variant modulates these outcomes. Nat Commun 2017; 8:15614.
27. Vastert SJ, van Wijk R, D'Urbano LE, *et al.* Mutations in the perforin gene can be linked to macrophage activation syndrome in patients with systemic onset juvenile idiopathic arthritis. Rheumatol 2010; 49:441–449.
28. Kaufman KM, Linghu B, Szustakowski JD, *et al.* Whole-exome sequencing reveals overlap between macrophage activation syndrome in systemic juvenile idiopathic arthritis and familial hemophagocytic lymphohistiocytosis. Arthritis Rheumatol 2014; 66:3486–3495.
29. Zhang M, Behrens EM, Atkinson TP, *et al.* Genetic defects in cytolysis in macrophage activation syndrome. Curr Rheumatol Rep 2014; 16:439.
30. Bracaglia C, Sieni E, Da Ros M, *et al.* Mutations of familial hemophagocytic lymphohistiocytosis (FHL) related genes and abnormalities of cytotoxicity function tests in patients with macrophage activation syndrome (MAS) occuring in systemic juvenile idiopathic arthritis (sJIA). Pediatr Rheumatol Online J 2014; 12:53.
31. Schulert GS, Zhang M, Husami A, *et al.* Novel UNC13D intronic variant disrupting a NF(B enhancer in a patient with recurrent macrophage activation syndrome and systemic juvenile idiopathic arthritis. Arthritis Rheumatol 2018; DOI: 10.1002/art.40438.
32. Schulert GS, Zhang M, Fall N, *et al.* Whole-exome sequencing reveals mutations in genes linked to hemophagocytic lymphohistiocytosis and macrophage activation syndrome in fatal cases of H1N1 influenza. J Infect Dis 2016; 213:1180–1188.
33. Zhang M, Bracaglia C, Prencipe G, *et al.* A heterozygous RAB27A mutation associated with delayed cytolytic granule polarization and hemophagocytic lymphohistiocytosis. J Immunol 2016; 196:2492–2503.
34. Canna SW, de Jesus AA, Gouni S, *et al.* An activating NLRC4 inflammasome mutation causes autoinflammation with recurrent macrophage activation syndrome. Nat Genet 2014; 46:1140–1146.
35. Romberg N, Al Moussawi K, Nelson-Williams C, *et al.* Mutation of NLRC4 causes a syndrome of enterocolitis and autoinflammation. Nat Genet 2014; 46:1135–1139.
36. ■ Brachat AH, Grom AA, Wulffraat N, *et al.* Early changes in gene expression and inflammatory proteins in systemic juvenile idiopathic arthritis patients on canakinumab therapy. Arthritis Res Ther 2017; 19:13.

This study demonstrated that although gene expression signatures and serum interleukin-6 rapidly respond to treatment, interleukin-18 levels remain persistently elevated in SJIA regardless of response to canakinumab.

37. ■■ Weiss ES, Girard-Guyonvarc'h C, Holzinger D, *et al.* Interleukin-18 diagnostically distinguishes and pathogenically promotes human and murine macrophage activation syndrome. Blood 2018; 131:1442–1455.

This work clearly shows that interleukin-18 levels, and in particular an imbalance between interleukin-18 and interleukin-18-binding protein leading to free interleukin-18, distinguishes MAS. Furthermore, it shows that NLRC4 gain-of-function variants also cause elevated serum interleukin-18, and that elevated free interleukin-18 in mice worsens MAS.

38. Girard-Guyonvarc'h C, Palomo J, Martin P, *et al.* Unopposed IL-18 signaling ■ leads to severe TLR9-induced macrophage activation syndrome in mice. Blood 2018; 131:1430–1441.
This report demonstrates the important role of free interleukin-18 in MAS using a well-characterized mouse model.
39. Canna SW, Girard C, Malle L, *et al.* Life-threatening NLRC4-associated hyperinflammation successfully treated with IL-18 inhibition. J Allergy Clin Immunol 2017; 139:1698–1701.
40. Gabay C, Fautrel B, Rech J, *et al.* Open-label, multicentre, dose-escalating phase II clinical trial on the safety and efficacy of tadekinig alfa (IL-18BP) in adult-onset Still's disease. Ann Rheum Dis 2018; 77:840–847.
41. Kessel C, Lippitz K, Weinhage T, *et al.* Proinflammatory cytokine environments can drive interleukin-17 overexpression by (γ/δ T cells in systemic juvenile idiopathic arthritis. Arthritis Rheumatol (Hoboken, NJ) 2017; 69:1480–1494.
42. Put K, Avau A, Brisse E, *et al.* Cytokines in systemic juvenile idiopathic arthritis and haemophagocytic lymphohistiocytosis: tipping the balance between interleukin-18 and interferon-γ. Rheumatology (Oxford) 2015; 54: 1507–1517.
43. Billiau AD, Roskams T, Van Damme-Lombaerts R, *et al.* Macrophage activation syndrome: characteristic findings on liver biopsy illustrating the key role of activated, IFN-gamma-producing lymphocytes and IL-6- and TNF-alpha-producing macrophages. Blood 2005; 105:1648–1651.
44. Bracaglia C, de Graaf K, Pires Marafon D, *et al.* Elevated circulating levels of ■ interferon-γ and interferon-γ-induced chemokines characterise patients with macrophage activation syndrome complicating systemic juvenile idiopathic arthritis. Ann Rheum Dis 2017; 76:166–172.
This large study demonstrates that serum IFNγ and IFNγ-induced chemokines distinguish MAS from active SJIA without MAS, supporting their utility as biomarkers.
45. Prencipe G, Caiello I, Pascarella A, *et al.* Neutralization of IFN-γ reverts clinical and laboratory features in a mouse model of macrophage activation syndrome. J Allergy Clin Immunol 2018; 141:1439–1449.
46. de Jager W, Vastert SJ, Beekman JM, *et al.* Defective phosphorylation of interleukin-18 receptor beta causes impaired natural killer cell function in systemic-onset juvenile idiopathic arthritis. Arthritis Rheum 2009; 60:2782–2793.
47. Put K, Vandenhaute J, Avau A, *et al.* Inflammatory gene expression profile and ■ defective interferon-γ and granzyme K in natural killer cells from systemic juvenile idiopathic arthritis patients. Arthritis Rheumatol 2017; 69:213–224.
This work represents the most thorough characterization of NK cells in SJIA and confirms cellular hyporesponsiveness to interleukin-18.
48. Cepika A-M, Banchereau R, Segura E, *et al.* A multidimensional blood ■■ stimulation assay reveals immune alterations underlying systemic juvenile idiopathic arthritis. J Exp Med 2017; 214:3449–3466.
This ambitious work integrates transcriptional, cytometric, and cytokine analysis of whole blood to define functional modules associated with inflammatory responses. These are then used to characterize immune responses in SJIA and define new pathways altered in SJIA monocytes.

Update on the genetics of nonbacterial osteomyelitis in humans

Allison J. Cox and Polly J. Ferguson

Purpose of review
To summarize the current advances in our understanding or the genetic basis of nonbacterial osteomyelitis.

Recent findings
Chronic recurrent multifocal osteomyelitis (CRMO) is a complex genetic disorder. Past discoveries identified several single gene defects (*LPIN2*, *Pstpip2* and *IL1RN*) that cause IL-1-mediated sterile multifocal osteomyelitis. Recently Lorden *et al.*'s studies show that LIPIN2 deficiency can activate the NLRP3 inflammasome through alterations in the function of P2X7 receptor providing evidence that Majeed syndrome is an NLRP3 inflammasomopathy. New gene discoveries include the identification of *FBLIM1* as a CRMO susceptibility gene. Mutations in *FBLIM1* were found in a consanguineous family with CRMO. *Fblim1* is one of the most significantly differentially expressed gene in bone from chronic multifocal osteomyelitis (*cmo*) mice, plays a role in IL-10-driven anti-inflammatory responses, and is involved in the physiology of bone remodeling. Lastly, new data on the putative CRMO susceptibility locus on chromosome 18 is presented here. Using Sanger sequencing, rather than microsatellite analysis, the DS18S60 susceptibility region could not be replicated in a larger cohort.

Summary
CRMO occurs in humans, nonhuman primates, dogs and mice. There is a genetic component to disease but the genetic basis has only been identified for a small percentage of all cases.

Keywords
autoinflammatory, chronic recurrent multifocal osteomyelitis, *FBLIM1*, *LPIN2*, *Pstpip2*

INTRODUCTION

Chronic recurrent multifocal osteomyelitis (CRMO) is a rare inflammatory disorder that targets the bone [1▪,2,3▪]. Affected individuals present with bone pain, which may be associated with localized swelling and warmth over the affected bone(s). It occurs predominantly in children with mean age of diagnosis at 9 years of age, although it can present in adulthood [2,4]. There is no diagnostic test. Laboratory studies are typically normal or demonstrate mild elevations in inflammatory markers in the blood [5,6]. Plain films can be normal early in disease, but typically show osteolytic lesions with surrounding sclerosis in the long bones near the growth plate [7,8]. Mandibular and clavicular lesions may be predominately sclerotic [7,9]. The vertebrae may be involved and vertebral collapse may occur secondary to inflammation [7,10]. Whole body magnetic resonance imaging is used to document the number and location of the bone lesions as well as response to therapy [11–13]. Inflammation is present in the bone without any detected pathogen [14]. There are no Food and Drug Administration (FDA)-approved medications for the treatment of CRMO. Treatment is with anti-inflammatory medications used off-label; initially with nonsteroidal anti-inflammatory drugs (NSAID) with escalation to bisphosphonates or biologics in those with disease refractory to NSAIDs [1▪,15,16▪▪,17,18]. Evidence suggests that CRMO is an autoinflammatory disease [19–21]. Individuals with CRMO are at much higher risk of developing psoriasis, inflammatory bowel disease or inflammatory arthritis than the general population [6,22]. The cause of CRMO largely

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KEY POINTS

- CRMO is a genetically heterogeneous disorder.
- Syndromic forms of CRMO account for <1% of all CRMO cases.
- *LPIN2*, *IL1RN* and *FBLIM1* have been implicated in human CRMO.
- The chromosome 18 CRMO susceptibility locus could not be replicated in a cohort of nearly 90 CRMO trios.

remains unknown but evidence suggests a significant genetic component to the disease.

EVIDENCE THAT CHRONIC RECURRENT MULTIFOCAL OSTEOMYELITIS HAS A GENETIC COMPONENT

CRMO was defined by Giedion *et al.* in 1972 and shortly thereafter, literature reports surfaced describing pedigrees with multiple affected family members, including affected sibling pairs and parent–child dyads [6,20]. CRMO is associated with other inflammatory disorders with a documented genetic component such as psoriasis, inflammatory bowel disease and inflammatory arthritis [22–25]. Two different groups reported that nearly 50% of the time, there is a family history of psoriasis, inflammatory bowel disease or inflammatory arthritis in the first and second degree relatives of individuals with CRMO [6,26]. There are two human syndromes and multiple inbred animal models with sterile multifocal osteomyelitis as a prominent phenotype [27–32].

Syndromic forms of chronic recurrent multifocal osteomyelitis

There are two rare syndromic forms of CRMO, Majeed syndrome and Deficiency of IL-1 Receptor Antagonist (DIRA) that typically affect individuals at a very young age [28,30,31]. CRMO typically presents before 2 years in Majeed syndrome and in infancy in DIRA. The skin may also be involved in these syndromes; Sweet syndrome has been reported with Majeed syndrome and pustulosis is common in DIRA [27,30,31]. An unusual dyserythropoietic anemia is seen in Majeed syndrome; it is typically microcytic and is congenital [27]. Majeed syndrome is caused by homozygous mutations in *LPIN2* and DIRA is caused by recessive loss-of-function mutations in *IL1RN* [28,30,31]. Patients with both syndromes respond well to IL-1-blocking agents [30,31,33]. However, the vast majority of cases of CRMO are nonsyndromic [34]. It is likely that CRMO is a heterogeneous disorder with as yet undefined gene–environment interaction resulting in disease [3▪].

Majeed syndrome as an NLRP3 inflammasomopathy

Majeed syndrome is a recessive disorder caused by loss of function mutations in *LPIN2* [28,35]. Individuals with Majeed syndrome develop dyserythropoietic anemia and early-onset severe *CRMO*. Fourteen cases of Majeed syndrome have been reported; all caused by private homozygous mutations in consanguineous families from the Middle East, India and Spain [28,33,35,36▪,37▪]. Most mutations are nonsense or splice site mutations predicted to result in a lack of protein, however, one mutation is a missense mutation that changes a highly conserved serine residue at amino acid 734 to a leucine (Ser734Leu) [29]. *LPIN2* encodes the protein LIPIN2, which is a phosphatidate phosphatase (PAP) that plays an important role in glycerolipid synthesis and can play a role in regulation of gene expression (transcriptional co-activator activity) [38▪▪]. Donkor *et al.* [39] demonstrated *in vitro* that the Ser734Leu mutant protein no longer has phosphatidate phosphatase activity but retains its PGC-1α/PPARα transcriptional coactivator activity. This suggests that the PAP activity of LIPIN2 is important in the pathophysiology of Majeed syndrome. There is a *Lpin2*-deficient mouse, which has increased PAP activity in the liver, likely because of compensatory increase in Lipin1 or Lipin3 protein expression. The mice do not develop osteomyelitis but do have a mild anemia, which is microcytic and with elevated platelet counts [40]. Unexpectedly, *Lpin2*-deficient mice develop disease of the cerebellum late in life manifested as tremor and ataxia [40]. This neurologic phenotype has not been reported in any humans with Majeed syndrome but long-term follow-up is lacking.

It remains unclear why LIPIN2 deficiency in humans targets the bone but recent discoveries help to establish that the NLRP3 inflammasome is involved. Herlin *et al.* demonstrated the importance of IL-1β in human Majeed syndrome. Two brothers with Majeed syndrome had prompt resolution of bone inflammation and reduction of blood inflammatory markers when treated with IL-1 inhibitors but not tumor necrosis factor (TNF) inhibitors. Blocking either the IL-1 receptor or blocking with an IL-1β-specific antibody both produced sustained clinical improvement demonstrating that Majeed syndrome is an IL-1β-driven disease. However, how LIPIN2 deficiency results in over-production of IL-1β remained unknown. Recently, Lorden *et al.*

[41▪▪] performed a set of elegant experiments using both human and mouse macrophages that demonstrated that LIPIN2 deficiency causes IL-1β dysregulation through NLRP3 inflammasome activation. They showed that Lipin2 is important in several key events in NLRP3 inflammasome activation, including inhibiting the activation and sensitization of the P2X7 purinergic receptor, apoptosis-associated speck-like protein with a caspase recruitment domain (ASC) oligomerization, and caspase-1 processing. Reduced levels of cellular cholesterol accompanied Lipin2 under-expression, and correction of the cholesterol concentration normalized both the currents through the P2X7 receptor and IL-1β production by the inflammasome machinery [41▪▪].

FBLIM1 AS A CHRONIC RECURRENT MULTIFOCAL OSTEOMYELITIS SUSCEPTIBILITY GENE

Cox *et al.* [42▪▪] identified a homozygous mutation in *FBLIM1* in a child with CRMO born to consanguineous parents using a whole exome sequencing. *FBLIM1* encodes the protein Filamin-Binding LIM Protein 1 (FBLP-1), a protein that had previously been shown to be important in bone remodeling [43]. FBLP-1 is a regulator of the cytoskeleton; it acts as an anchor for cell-extracellular matrix (ECM) adhesion proteins and filamin-containing actin filaments, and is involved in integrin activation [44–47]. The mutation identified in the child with CRMO was in the filamin domain of FBPL-1 [42▪▪]. Mice deficient of FBLP-1 have increased RANKL (receptor activator of nuclear factor kappa-B ligand) expression, increased osteoclast activation and are severely osteopenic [43]. As part of an IL-10-mediated anti-inflammatory response in macrophages, *FBLIM1* was shown to be regulated by STAT3 and significantly upregulated consistent with its role as a gene with anti-inflammatory properties [48]. Additional support for *Fblim1* in sterile osteomyelitis comes from the *Pstpip2*-deficient chronic multifocal osteomyelitis (*cmo*) mouse. When comparing gene expression profiles in the bones of diseased cmo mice from unaffected cmo.IL1R-/- mice that lack a functional IL-1 receptor, *Fblim1* was the most differentially expressed gene (down regulated 20-fold) [42▪▪]. Another unrelated individual with CRMO was found to have a frameshift mutation on one FBLIM1 allele and a enhancer variant in the other allele that altered enhancer activity [42▪▪]. This data supports *FBLIM1* as a CRMO susceptibility gene.

ROLE OF CHROMOSOME 18 IN CHRONIC RECURRENT MULTIFOCAL OSTEOMYELITIS

In 2002, Golla *et al.* [49] reported a CRMO-associated locus on chromosome 18 in humans in the region that is syntenic to the mouse *cmo* locus. A rare allele (166 bp size fragment) for the CA microsatellite D18S60 (rs111655813) in the region was found to segregate significantly with CRMO in an analysis of 27 family trios from Germany [reported to be transmitted to the affected child from a heterozygous parent 11 out of 12 times ($P = 0.003$)] [49]. A haplotype encompassing the genes *RANK, PIGN* and *KIAA1468* was also significantly enriched in the CRMO cohort but sequencing did not reveal any disease-associated variants [49]. This finding had not been replicated and its significance remained unknown.

Therefore, we sought to replicate the previous association in our cohort of 84 CRMO trios but discovered that other polymorphisms were present in the repeat region including rs3220079, a SNP abutting the CA repeat, which complicated the analysis. Therefore, to accurately genotype this locus, we utilized Sanger sequencing to call genotypes based on the number of CA repeats rather than using microsatellite analysis. We found that the 166 bp microsatellite amplicon is the equivalent of 20 CA repeats. In dbSNP, the number of CA repeats (and their allele frequencies) for D18S60 are 15 (1.8%), 16 (78.5%), 17 (8.9%), 20 (5.4%), 21 (1.8%) and 23 (3.6%). The 5.4% frequency of the (CA)20-repeat allele also corresponds to the 5% frequency of the 166 bp amplicon utilized for the transmission disequilibrium test (TDT) reported by Golla *et al.* [49]. Using Sanger sequencing rather than gel electrophoresis to genotype followed by a TDT, we found no association between CRMO and

Table 1. Transmission disequilibrium test on 84 trios

Variant ID	Allele	Number of heterozygous parents	Number of alleles transmitted	Number of alleles not transmitted	Two-tailed *P*-value
D18S60/rs3220079	CA (20)	27	12	15	0.7011
D18S60/rs3220079	CA (17)	41	19	22	0.7552
rs12326680	C	60	28	32	0.6989
rs201716532	– (GA deleted)	21	9	12	0.6636

inheritance of D18S60 in our cohort (Table 1 and supplementary material, http://links.lww.com/COR/A40).

CONCLUSION

CRMO in humans is a complex genetic disorder. Several genes have been identified that can cause sterile osteomyelitis in human and animal models including *LPIN2*, *IL1RN*, *Pstpip2* (mouse) and *FBLIM1*. Yet mutations in these genes are found in a small proportion of CRMO cases suggesting other as yet unidentified genes are playing a role. The chromosome 18 locus identified in a German cohort by TDT was not replicated in a larger cohort of CRMO patients (North American and predominantly Caucasian) making it unlikely that a CRMO susceptibility gene is harbored in that region. Next generation sequencing methods are being utilized to identify susceptibility genes; the results of which have not been published. Knowing the genes and inflammatory pathways that are driving the disease in CRMO will help improve diagnosis and treatment.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. ■ Hofmann SR, Kapplusch F, Girschick HJ, *et al.* Chronic recurrent multifocal osteomyelitis (CRMO): presentation, pathogenesis, and treatment. Curr Osteoporos Rep 2017; 15:542–554.
This article is a nice review on CRMO.
2. Wipff J, Costantino F, Lemelle I, *et al.* A large national cohort of French patients with chronic recurrent multifocal osteitis. Arthritis Rheumatol 2015; 67:1128–1137.
3. ■ Cox AJ, Zhao Y, Ferguson PJ. Chronic recurrent multifocal osteomyelitis and related diseases-update on pathogenesis. Curr Rheumatol Rep 2017; 19:18.
Recent review on pathogenesis.
4. Schultz C, Holterhus PM, Seidel A, *et al.* Chronic recurrent multifocal osteomyelitis in children. Pediatr Infect Dis J 1999; 18:1008–1013.
5. El-Shanti HI, Ferguson PJ. Chronic recurrent multifocal osteomyelitis: a concise review and genetic update. Clin Orthop Relat Res 2007; 462: 11–19.
6. Jansson A, Renner ED, Ramser J, *et al.* Classification of nonbacterial osteitis: retrospective study of clinical, immunological and genetic aspects in 89 patients. Rheumatology (Oxford) 2007; 46:154–160.
7. Khanna G, Sato TS, Ferguson P. Imaging of chronic recurrent multifocal osteomyelitis. Radiographics 2009; 29:1159–1177.
8. Jurriaans E, Singh NP, Finlay K, Friedman L. Imaging of chronic recurrent multifocal osteomyelitis. Radiol Clin North Am 2001; 39:305–327.
9. Girschick HJ, Krauspe R, Tschammler A, Huppertz HI. Chronic recurrent osteomyelitis with clavicular involvement in children: diagnostic value of different imaging techniques and therapy with nonsteroidal anti-inflammatory drugs. Eur J Pediatr 1998; 157:28–33.
10. Kahn MF. Chronic recurrent multifocal osteomyelitis. Association with vertebra plana. J Bone Joint Surg Am 1990; 72:305–306.
11. Guerin-Pfyffer S, Guillaume-Czitrom S, Tammam S, Kone-Paut I. Evaluation of chronic recurrent multifocal osteitis in children by whole-body magnetic resonance imaging. Joint Bone Spine 2012; 79:616–620.
12. Kennedy MT, Murphy T, Murphy M, *et al.* Whole body MRI in the diagnosis of chronic recurrent multifocal osteomyelitis. Orthop Traumatol Surg Res 2012; 98:461–464.
13. Roderick M, Shah R, Finn A, Ramanan AV. Efficacy of pamidronate therapy in children with chronic nonbacterial osteitis: disease activity assessment by whole body magnetic resonance imaging. Rheumatology (Oxford) 2014; 53:1973–1976.
14. Girschick HJ, Raab P, Surbaum S, *et al.* Chronic nonbacterial osteomyelitis in children. Ann Rheum Dis 2005; 64:279–285.
15. Zhao Y, Laxer RM, Ferguson PJ. Treatment advances in chronic non-bacterial osteomyelitis and other autoinflammatory bone conditions. Curr Treat Options in Rheum 2017; 3(1):17–32.
16. ■■ Zhao Y, Wu EY, Oliver MS, *et al.,* CNO/CRMO study group the CARRA SVARD subcommittee. Consensus treatment plans for chronic nonbacterial osteomyelitis refractory to nonsteroidal anti-inflammatory drugs and/or with active spinal lesions. Arthritis Care Res (Hoboken) 2017; doi: 10.1002/acr.23462. [Epub ahead of print]
Description of consensus treatment plans for patients that need more than NSAID for treatment.
17. Beck C, Morbach H, Beer M, *et al.* Chronic nonbacterial osteomyelitis in childhood: prospective follow-up during the first year of anti-inflammatory treatment. Arthritis Res Ther 2010; 12:R74.
18. Gleeson H, Wiltshire E, Briody J, *et al.* Childhood chronic recurrent multifocal osteomyelitis: pamidronate therapy decreases pain and improves vertebral shape. J Rheumatol 2008; 35:707–712.
19. Morbach H, Hedrich CM, Beer M, Girschick HJ. Autoinflammatory bone disorders. Clin Immunol 2013; 147:185–196.
20. Ferguson PJ, El-Shanti HI. Autoinflammatory bone disorders. Curr Opin Rheumatol 2007; 19:492–498.
21. Ferguson PJ, Sandu M. Current understanding of the pathogenesis and management of chronic recurrent multifocal osteomyelitis. Curr Rheumatol Rep 2012; 14:130–141.
22. Laxer RM, Shore AD, Manson D, *et al.* Chronic recurrent multifocal osteomyelitis and psoriasis–a report of a new association and review of related disorders. Semin Arthritis Rheum 1988; 17:260–270.
23. Paller AS, Pachman L, Rich K, *et al.* Pustulosis palmaris et plantaris: its association with chronic recurrent multifocal osteomyelitis. J Am Acad Dermatol 1985; 12(5 Pt 2):927–930.
24. Carpenter E, Jackson MA, Friesen CA, *et al.* Crohn's-associated chronic recurrent multifocal osteomyelitis responsive to infliximab. J Pediatr 2004; 144:541–544.
25. Bjorksten B, Gustavson KH, Eriksson B, *et al.* Chronic recurrent multifocal osteomyelitis and pustulosis palmoplantaris. J Pediatr 1978; 93:227–231.
26. Ferguson PJ, Brown DR, Sundel R, *et al.* Chronic inflammatory disorders in first and second degree relatives of children with chronic recurrent multifocal osteomyelitis. Arthritis Rheum 2005; 52:747.
27. Majeed HA, Al-Tarawna M, El-Shanti H, *et al.* The syndrome of chronic recurrent multifocal osteomyelitis and congenital dyserythropoietic anaemia. Report of a new family and a review. Eur J Pediatr 2001; 160:705–710.
28. Ferguson PJ, Chen S, Tayeh MK, *et al.* Homozygous mutations in LPIN2 are responsible for the syndrome of chronic recurrent multifocal osteomyelitis and congenital dyserythropoietic anaemia (Majeed syndrome). J Med Genet 2005; 42:551–557.
29. Ferguson PJ, Bing X, Vasef MA, *et al.* A missense mutation in Pstpip2 is associated with the murine autoinflammatory disorder chronic multifocal osteomyelitis. Bone 2006; 38:41–47.

30. Aksentijevich I, Masters SL, Ferguson PJ, *et al.* An autoinflammatory disease with deficiency of the interleukin-1-receptor antagonist. N Engl J Med 2009; 360:2426–2437.

31. Reddy S, Jia S, Geoffrey R, *et al.* An autoinflammatory disease due to homozygous deletion of the IL1RN locus. N Engl J Med 2009; 360: 2438–2444.

32. Grosse J, Chitu V, Marquardt A, *et al.* Mutation of mouse Mayp/Pstpip2 causes a macrophage autoinflammatory disease. Blood 2006; 107: 3350–3358.

33. Herlin T, Fiirgaard B, Bjerre M, *et al.* Efficacy of anti-IL-1 treatment in Majeed syndrome. Ann Rheum Dis 2013; 72:410–413.

34. Beck C, Girschick HJ, Morbach H, *et al.* Mutation screening of the IL-1 receptor antagonist gene in chronic nonbacterial osteomyelitis of childhood and adolescence. Clin Exp Rheumatol 2011; 29:1040–1043.

35. Al-Mosawi ZS, Al-Saad KK, Ijadi-Maghsoodi R, *et al.* A splice site mutation confirms the role of LPIN2 in Majeed syndrome. Arthritis Rheum 2007; 56:960–964.

36. ▪ Rao AP, Gopalakrishna DB, Bing X, Ferguson PJ. Phenotypic variability in Majeed syndrome. J Rheumatol 2016; 43:1258–1259.
First case report of Majeed syndrome presenting in the second decade.

37. ▪ Pinto-Fernandez C, Seoane-Reula ME. Efficacy of treatment with IL-1RA in Majeed syndrome. Allergol Immunopathol (Madr) 2017; 45:99–101.
New report of Majeed syndrome from Spain treated with human recombinant IL-1 receptor antagonist.

38. ▪▪ Zhang P, Reue K. Lipin proteins and glycerolipid metabolism: roles at the ER membrane and beyond. Biochim Biophys Acta 2017; 1859(9 pt B): 1583–1595.
Helpful review of the function of lipin proteins including Lipin2, which is involved in Majeed syndrome.

39. Donkor J, Zhang P, Wong S, *et al.* A conserved serine residue is required for the phosphatidate phosphatase activity but not the transcriptional coactivator functions of lipin-1 and lipin-2. J Biol Chem 2009; 284:29968–29978.

40. Dwyer JR, Donkor J, Zhang P, *et al.* Mouse lipin-1 and lipin-2 cooperate to maintain glycerolipid homeostasis in liver and aging cerebellum. Proc Natl Acad Sci U S A 2012; 109:E2486–E2495.

41. ▪▪ Lorden G, Sanjuan-Garcia I, de Pablo N, *et al.* Lipin-2 regulates NLRP3 inflammasome by affecting P2X7 receptor activation. J Exp Med 2016; 511-528.
Key article demonstrating that Lipin2 regulates the NLRP3 inflammasome.

42. ▪▪ Cox AJ, Darbro BW, Laxer RM, *et al.* Recessive coding and regulatory mutations in FBLIM1 underlie the pathogenesis of chronic recurrent multifocal osteomyelitis (CRMO). PLoS One 2017; 12:e0169687.
This article describes mutations in FBLIM1 in human CRMO and presents supporting data for the role of FBLIM1 in CRMO from murine and in-vitro models.

43. Xiao G, Cheng H, Cao H, *et al.* Critical role of filamin-binding LIM protein 1 (FBLP-1)/migfilin in regulation of bone remodeling. J Biol Chem 2012; 287:21450–21460.

44. Tu Y, Wu S, Shi X, *et al.* Mig-2 link focal adhesions to filamin and the actin cytoskeleton and function in cell shape modulation. Cell 2003; 113:37–47.

45. Das M, Ithychanda SS, Qin J, Plow EF. Migfilin and filamin as regulators of integrin activation in endothelial cells and neutrophils. PLoS One 2011; 6:e26355.

46. Volmering S, Block H, Boras M, *et al.* The neutrophil Btk signalosome regulates integrin activation during sterile inflammation. Immunity 2016; 44:73–87.

47. Gkretsi V, Zhang Y, Tu Y, *et al.* Physical and functional association of migfilin with cell-cell adhesions. J Cell Sci 2005; 118(Pt 4):697–710.

48. Hutchins AP, Poulain S, Miranda-Saavedra D. Genome-wide analysis of STAT3 binding in vivo predicts effectors of the anti-inflammatory response in macrophages. Blood 2012; 119:e110–e119.

49. Golla A, Jansson A, Ramser J, *et al.* Chronic recurrent multifocal osteomyelitis (CRMO): evidence for a susceptibility gene located on chromosome 18q21.3-18q22. Eur J Hum Genet 2002; 10:217–221.

Spondyloarthritis: new insights into clinical aspects, translational immunology and therapeutics

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Purpose of review
The spondyloarthopathies (SpA), which encompass related diseases that were originally viewed as autoimmune, are now known to have a strong innate immune or autoinflammatory initiation phase characterized by disease localization to tissue-specific sites based on the nuances and microanatomy and immunology of those sites. This review covers recent translational advances in the field of SpA.

Recent findings
Imaging studies in SpA continue to add support for the pivotal role of enthesitis in disease initiation and expression. Although in its infancy, there is growing evidence for microbiotal intestinal dysbiosis in ankylosing spondylitis and psoriatic arthritis. The role of cytokines beyond tumour necrosis factor (TNF) continues to grow with support for the interleukin (IL)-23/17 axis being key to disease and emergent evidence for the importance of the IL-36 pathway. The treatment of inflammatory bowel disease (IBD) with vedolizumab an $\alpha 4\beta 7$-integrin blocker has been associated with arthritis flares and small molecules with Janus kinase inhibition appear to be as effective as the anti-TNFs. The disparate response of different domains in SpA points towards immunological heterogeneity even within what was considered a homogeneous disease.

Summary
The clinical aspects and translational immunology and therapeutics of SpA continue to evolve and indicate the complexity of diagnosis and treatment of these conditions.

Keywords
ankylosing spondylitis, entheses, enthesitis, interleukin-12, interleukin -17, interleukin -23, Janus kinase inhibitors (JAKi), psoriatic arthritis, spondyloathripathies, tumour necrosis factor inhibitor

INTRODUCTION

The spondyloarthropathies (SpA) are a group of diseases that share several characteristics, including genetic predisposition, shared clinical manifestation, a characteristic nonautoimmune multiorgan disease distribution and a pivotal role for cytokines in disease [1]. SpA include ankylosing spondylitis (AS), reactive arthritis (ReA), psoriatic arthritis (PsA) and inflammatory bowel disease (IBD) related arthropathy (IBD arthropathy) [1]. Enthesitis is an important pathologic feature of SpA and believed to represent the primary pathogenic process in such group of diseases [2]. Indeed, experimental models have shown several SpA-like diseases that initiate at entheses and are linked to nail disease as well as dactylitis, two important entheseal-associated conditions in humans [3]. In addition, there is a strong association between SpA and HLA-B27, yet the mechanism by which the latter impacts on the pathogenesis and induction of SpA is still controversial [4]. In the last few years, great advances occurred regarding the treatment of SpA attributed to a better understanding of the pathogenesis and immune pathways involved in these diseases [5]. In this review, we briefly highlight the recent insights concerning the imaging, immune pathways, new therapies and the role of microbiome in SpA group of diseases.

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KEY POINTS

- Spondyloarthritis (SpA) is a heterogeneous group of disorders, although share many aspects, including immunogenetic pathways, clinical features and treatment.
- Imaging continues to show that entheses is a key player in the development of SpA.
- Microbiome seems to impact the development and course of SpA and may have some future potential therapeutic effects.

IMAGING

MRI and ultrasonography have transformed our recognition of enthesitis at clinically inaccessible sites as a pivotal pathogenic process in SpA group of diseases [6]. Recent ultrasound studies have shown that the accessory pulleys, a type of mini-enthesis, are thickened in PsA individuals with a history of dactylitis and this phenomenon has been likened to a deep Koebner phenomenon [7]. Imaging studies using ultrasound have showed the robustness of an earlier ultrasound showing that imaging of the flexor tendon compartment can differentiate between RA and PsA hand involvement [8].

Regarding CT, recently, it has been shown that CT-based syndesmophytes measurements had very good longitudinal validity and better sensitivity to change than radiography or MRI [9]. The introduction of MRI of the sacroiliac joints (SIJs) has led to a significant change in understanding and recognition of the disorder. A recent study has suggested to repeat MRI scans within a 12-week period in HLA-B27 positive men with inflammatory back pain increasing the probability to detect new radiological findings in comparison to those without HLA-B27 [10]. Moreover, it has been reported that in AS patients, bone oedema on MRI imaging is highly associated with low bone mineral density [11].

THERAPEUTIC DISSECTION OF THE SPONDYLOARTHROPATHIES PATHOGENESIS

An interesting development in SpA has been the advent of vedolizumab, an α4β7-integrin mAb that blocks gut-specific homing of lymphocytes and is used for the treatment of IBD. Whilst it might be expected that such a strategy, by dampening or completely abrogating gut inflammation, may treat associated extraintestinal SpA, the situation is more complex. Indeed, there is evidence for abrogation of IBD-associated SpA with vedolizumab [12]. However, recent reports of a flare in SpA or new onset of SpA related arthritis, usually mild, have been reported on cases on therapy. Reports are also emerging for the need for concomitant antitumour necrosis factor (anti-TNF) agents to control SpA-related arthropathy in individuals who need concomitant vedolizumab to control the gut-related arthritis [13].

Studies are also being reported on difficult to treat psoriatic arthritis and psoriasis. Recent series combined with historical case reports indicate that a small subgroup of psoriatic cases have a disease wherein skin involvement is resistant to anti-TNF, but responds to IL-23/17 axis blockade and conversely arthritis in the same cases are sensitive to anti-TNF therapy but resistant to anti-IL23/17 pathway blockers [14].

These findings in SpA, related IBD and psoriasis represent remarkable clinical experiments. They point towards complex sub-compartmentalization and functional devolution of the immune system within individual patients, as completely different strategies are needed to control the overall disease burden. These findings hark back to earlier studies of the effectiveness of sulphasalaisine for gut but not axial disease and the effectiveness of etanercept for arthritis but not gut disease and also the recent effectiveness for anti-IL-17A for skin and SpA arthropathy but not IBD.

NEW THERAPIES FOR SPONDYLOARTHROPATHIES

Borrowing therapies from the RA arena have led to some recent failures and also to some recent successes. Neither ustekinumab or guselkumab were effective in RA but appear effective in PsA peripheral arthropathy [15]. Indeed, Ustekinumab, an IL-12/23 p40 inhibitor, decreases clinical manifestations of peripheral arthritis, dactylitis, enthesitits and psoriasis [16], and Guselkumab a IL-23p19 inhibitor, has been shown to improve symptoms in PsA [17]. With regards to IL-17 inhibitors, Secukinumab was found to improve clinical symptoms in both AS and PsA [18,19]. Ixekizumab, which is another IL-17 inhibitor, has also been shown to improve disease activity in PsA [20]. Bimekizumab, an inhibitor of both IL-17A and IL-17F, has been demonstrated to improve clinical symptoms of both AS and PsA and is superior to inhibition of IL-17 or IL-17F alone [21]. However, the IL-17A inhibitors, like IL-23 blockers, have limited efficacy in RA and are not being developed for this indication.

Tofocitinib, a Janus kinase inhibitor, has proven to be effective in PsA with numerically higher ACR20 responses than adalimumab for the 10 mg

dosing regimen [22▪▪]. Another study utilizing tofacitinib in TNF failures in PsA also showed efficacy [23▪▪]. Therefore, there is considerable optimism that insights into pathogenesis were translated into better treatments for patients with SpA. Finally, the efficacy of the PDE4 blocker, Apremilast in PsA, but not RA, also supports the emergent understanding of the immunological disparity between RA and PsA [24,25].

IMMUNOLOGICAL PATHWAYS AND CYTOKINES INVOLVED IN SPONDYLOARTHROPATHIES

The IL-23/IL-17 axis is associated with SpA including AS and PsA [26–28]. IL-17A is a cytokine produced by lymphocytes of the adaptive and innate immune system, including T helper-17 cells (Th17), IL-17-producing CD8(+)T cells (Tc17), γδ T cells and type 3 innate lymphoid cells (ILC3) [29]. All of these IL-17A producing populations differentiate following stimulation from a range of cytokines, the most prominent of these is IL-17A itself; however, IL-23 appears to be a core driver of this process [30]. The IL-17 family consists of six cytokines, A, B, C, D, E and F. The critical role of IL-17A in inflammatory arthritis is well established and the effect of the closely related family member IL-17F has also recently been described in a model of inflammatory arthritis [31].

In AS, IL-17 secreting cells have been shown to be present in the facet joints [32] and AS susceptibility is linked to polymorphisms in the IL-23 receptor (IL-23R) [33] and in STAT3, which IL-23 signals through [34]. However, a recent study revealed the various IL-23R polymorphisms do not correlate with serum IL-17 levels in AS patients [35], although serum levels of both IL-17 and IL-23 are increased [36]. Despite these observations, no serum biomarkers beyond C-reactive protein (CRP) were found to have robust applicability in AS. In PsA, IL-17 expressing cells are found in the synovial fluid with the most significant increase associated with Tc17 cells [37–39]. Here, polymorphisms in the IL-23R are also associated with the disease [40] and IL-23p19 mRNA and synovial IL-23 expression correlate with swollen joint count and CRP [41].

Also, of great interest is the recent discovery of a new cytokine, IL-39 [42,43]. IL-39 is a IL-12 family member cytokine consisting of IL-23p19 and Epstein–Barr virus induced 3 (EBI3) chains. IL-39 has been assigned a role in murine models of lupus, but the existence of the cytokine is yet to be confirmed in humans [42,43]. If IL-39 has a role in SpA, it could be speculated that IL-39 actions can be inhibited by existing IL-23p19 blockers such as guselkumab, which could potentially explain differences in efficacy between IL-12p40 blockers and IL-23p19 blockers in disease. This is an area of research that is ripe for exploration in the coming years.

IL-36 family cytokines consisting of IL-36α, IL-36β and IL-36γ are members of the wider IL-1 family and have been associated with psoriasis skin immunopathology [44,45] and their effects are blocked by the naturally occurring IL-36 antagonist (IL-36Ra). The importance of IL-36 associated inflammation in psoriasis was further heightened by the discovery that loss of function in the IL-36Ra is associated with generalized pustular psoriasis (GPP) [46]. This condition has since been named DITRA (deficiency of the interleukin IL-36 receptor antagonist). IL-36 family cytokines have also been implicated in both mouse models and humans in a range of other diseases, including skin inflammatory diseases, IBD, renal fibrosis, respiratory infection and chronic obstructive pulmonary disease (COPD) [47–53]. IL-36 has been assigned a role as a regulator of the IL-23/IL-17 axis [54,55]; it acts on APCs (macrophages and dendritic cells) to induce IL-23. This finding has also been replicated on human immune cells *in vitro* [56,57]. A recent phenotyping study identified a number of individuals with no functioning IL-36 receptor (IL-36R). These individuals maintained normal immune function, suggesting therapeutic blockage of IL-36 may be possible [58]. IL-36 is yet to be implicated in human SpA. IL-37 and IL-38 are anti-inflammatory IL-1 family cytokines that mediate immunosuppressive effects in murine models of skin and joint inflammation [59–61]. These cytokines can attenuate inflammation driven by IL-1, IL-36 and TLR-agonists [62,63]. *IL-38* gene polymorphisms are associated with both AS and PsA [64–66]. Collectively, with IL-39, these cytokines form an interesting group of 'thirty somethings' that may help elucidate the heterogeneity of SpA in the coming years.

THE ROLE OF GUT MICROBIOME IN SPONDYLOARTHROPATHIES

In recent years, a rapid development of analytical techniques has occurred, allowing greater understanding into the role of the human microbiome and its relation to various disorders. There is a great deal of interest in the field and potential for better understanding to deliver improved diagnosis and treatment of SpA. Several studies in the last few years showed that the gut microbiome composition differs in SpA patients in comparison to healthy individuals. Recently, a HLA-B27/β_2m-transgenic rat model of SpA found that HLA-B27 expression profoundly impacts the intestinal metabolome [67]. In

addition, an increased abundance of *Ruminococcus gnavus* in SpA patients, as compared with both RA patients and healthy controls, has been detected and significantly correlated with disease activity in patients with a history of IBD [68].

It is well known that around half of AS patients have subclinical terminal ileitis [69]. Costello *et al.* [70] showed different microbial communities in the terminal ileum of AS patients in comparison to healthy controls. There was a decrease in the abundance of Veillonellaceae and Prevotellaceae, and an increase of the abundance of Lachnospiraceae, Ruminococcaceae, Rikenellaceae, Porphyromonadaceae and Bacteroidaceae [70]. Furthermore, HLA-B27 transgenic rats do not develop most of the AS features in a germ-free environment, whereas transferring them to a conventional rat colony leads to development of classical AS symptoms [71]. Another study demonstrated that the gut of AS patients harbour a higher prevalence of sulphate-reducing bacteria, which are implicated in the pathogenesis of IBD [72]. Moreover, previous studies have showed that subclinical Klebsiella infection is involved in the pathogenesis of AS and IBD [73,74].

Faecal samples from patients with psoriasis and PsA showed a decrease in the abundance of Coprococcus species; PsA patients also showed a significant reduction in Akkermansia, Ruminococcus and Pseudobutyrivibrio, a microbiota profile previously described in IBD [75]. A recent study found that individuals with ReA had a significantly higher abundance of Erwinia and Pseudomonas, and those with enthesitis had a higher prevalence of Campylobacter, whereas individuals with radiographic sacroiliitis and uveitis were enriched in Ruminococcaceae and Erwinia, respectively [76]. With regards to IBD, ulcerative colitis patients with arthritis had a higher prevalence of Staphylococcus, Klebsiella and Proteus in stool cultures [77]. To summarize, the complex interplay between the gut microbiome and SpA pathogenesis is still poorly understood, widely differing findings have been reported in different SpA-related manifestations (Fig. 1) and the field is still in its infancy.

NEW BONE FORMATION IN SPONDYLOARTHROPATHIES

It has been established that male sex, HLA-B27 status, smoking, baseline radiographic damage, elevation in CRP and MRI positivity for both fatty corner lesions and corner bone marrow oedema predict propensity for spinal fusion progression. It is now clear that spinal bone formation in AS and SpA is postinflammation tissue remodelling reaction rather than an uncoupled process from inflammation, which might be the case in diffuse idiopathic skeletal hyperostosis (DISH). Several

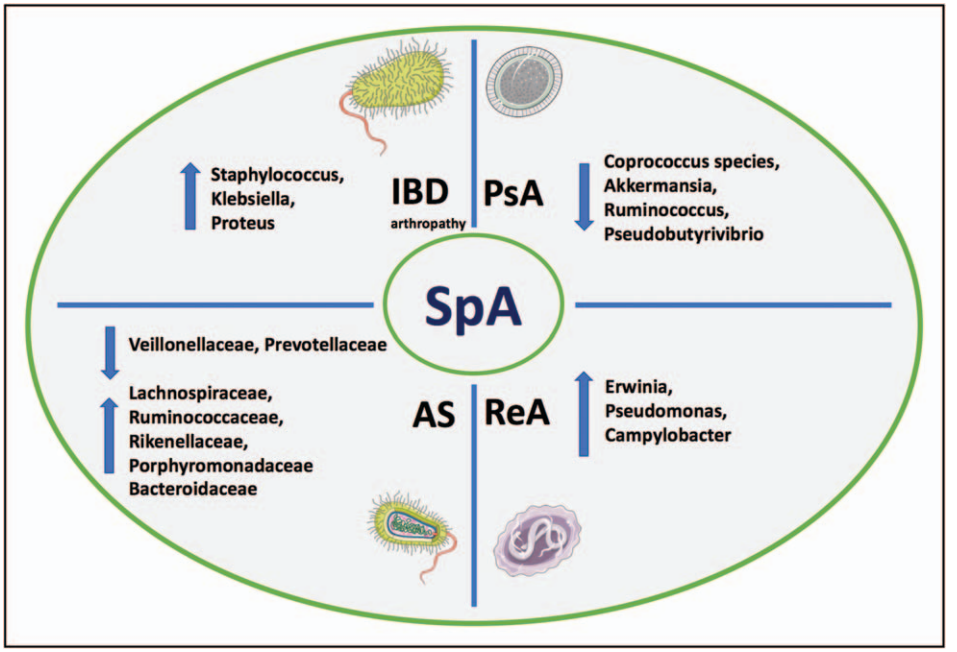

FIGURE 1. Despite the numerous genetic and immunological and clinical overlaps in SpA, research into the microbiome thus far has indicated many differences rather than common unifying microbial signatures. Whether this represents chance or potential informatics related biases or genuine fundamental differences awaits elucidation. Features of this figure are reproduced from https://smart.servier.com (Servier Medical Art by Servier is licensed under a Creative Commons Attribution 3.0 Unported License), and were changed in terms of shape and size.

studies have attempted to provide a serum biomarker that could robustly predict spinal fusion progression, but this has proved elusive. A recent study has suggested that macrophage migration inhibitory factor (MIF) may be one such marker that can predict the risk of fusion [78]. MIF is an enigmatic polyfunctional molecule that has been considered as a cytokine, a hormone and a growth factor. Baseline serum MIF levels were reported to be elevated in AS and also found to independently predict AS progression [78]. The same study showed MIF-induced TNF production in monocytes and β-catenin driven osteoblasts maturation and mineralization, which collectively point towards both inflammation and new bone formation. Clearly, further work and independent replication is needed to determine whether MIF measurement with or without CRP is superior to CRP alone in predicting radiological progression. This could ultimately help direct therapy towards biological agents in individuals with early-onset axial SpA.

On the same theme, recent studies of low-dose CT have shown this to be a useful method for the more sensitive measurement of new bone formation in the spine and especially the thoracic spine in AS [79]. There is a need for further studies in the translational setting to evaluate the effect of both TNF and IL-17A blockade on new bone formation in SpA. Mechanistically, blocking IL-17A but not blocking IL-22, which is also downstream of IL-23 signalling, might be predicted to accelerate new bone formation; IL-22 does indeed accelerate new bone formation *in vitro* [80▪▪]. However, the effect of IL-17A on bone formation seems to be highly contextual and there is a growing body of evidence that IL-17 also stimulates bone formation *in vivo* and *in vitro* [81,82]. In addition, there is no evidence for an acceleration of new bone formation under IL-17A therapy. Our expectation is that the early initiation of strategies that switch off spinal osteitis in SpA will have a profound impact on the retardation of new bone formation.

CONCLUSION

The term SpA encompasses a heterogeneous group of disorders that despite the great advances made concerning pathogenesis understanding is still in its infancy. Some of the major poorly understood aspects include systemic and local factors that trigger innate immune activation at the enthesis, the cell types involved in this and how this leads to extensive joint inflammation (Fig. 2). Future basic and clinical research should hasten the development of efficacious treatments for individuals that are currently difficult to treat.

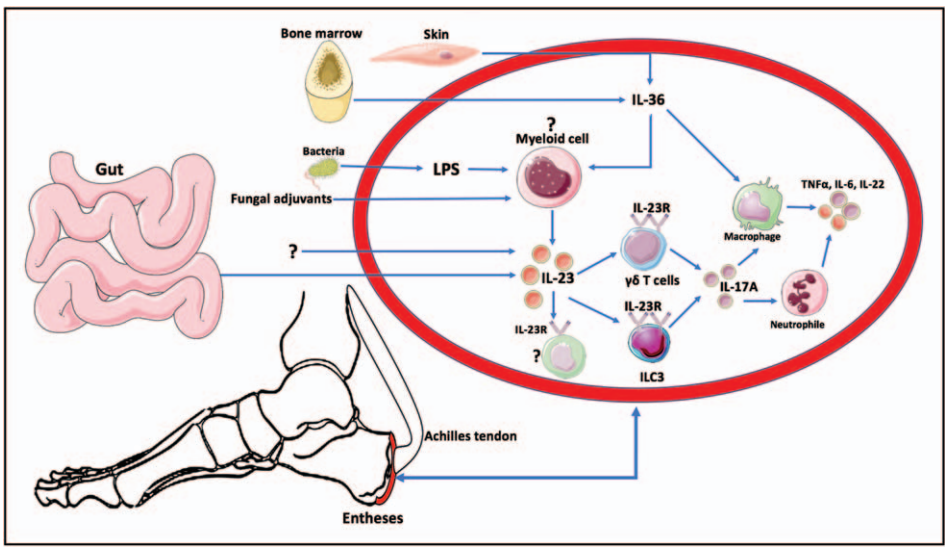

FIGURE 2. Experimentally, there is evidence pointing towards inflammation or inflammation triggering molecules originating outside the enthesis including gut and skin and other sites, that might trigger enthesitis, but these are poorly defined in man (Question 1 (Q1). In animal models, this has been ascribed to systemic overexpression of IL-23 [3]. Several other pattern recognition receptor (PRR) ligands may also gain access to the enthesis including lipopolysaccharide and fungal adjuvants, for example (Q2). The enthesis resident myeloid cells, or enthesis infiltrating cells, that respond to exogenous stimuli or to local microdamage remain poorly defined (Q3) [83]. Beyond gamma delta T cells and ILCs, the innate and adaptive cell populations at the enthesis remain poorly defined (Q4) [84]. The precise interplay and cytokine hierarchy at the enthesis is not well understood (Q5) [Features of this figure are reproduced from https://smart.servier.com (Servier Medical Art by Servier is licensed under a Creative Commons Attribution 3.0 Unported License)], and were changed in terms of shape and size.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. van Tubergen A, Weber U. Diagnosis and classification in spondyloarthritis: identifying a chameleon. Nat Rev Rheumatol 2012; 8:253–261.
2. Schett G, Lories RJ, D'Agostino MA, *et al.* Enthesitis: from pathophysiology to treatment. Nat Rev Rheumatol 2017; 13:731–741.
3. Sherlock JP, Joyce-Shaikh B, Turner SP, *et al.* IL-23 induces spondyloarthropathy by acting on ROR-gammat+ CD3+CD4-CD8- entheseal resident T cells. Nat Med 2012; 18:1069–1076.
4. Lim CSE, Sengupta R, Gaffney K. The clinical utility of human leucocyte antigen B27 in axial spondyloarthritis. Rheumatology (Oxford) 2017. [Epub ahead of print]
5. Dubash S, McGonagle D, Marzo-Ortega H. New advances in the understanding and treatment of axial spondyloarthritis: from chance to choice. Ther Adv Chronic Dis 2018; 9:77–87.
6. Watad A, Eshed I, McGonagle D. Lessons learned from imaging on enthesitis in psoriatic arthritis. Isr Med Assoc J 2017; 19:708–711.
7. Tinazzi I, McGonagle D, Aydin SZ, *et al.* 'Deep Koebner' phenomenon of the flexor tendon-associated accessory pulleys as a novel factor in tenosynovitis and dactylitis in psoriatic arthritis. Ann Rheum Dis 2018; 77:922–925.
8. Tinazzi I, McGonagle D, Zabotti A, *et al.* Comprehensive evaluation of finger flexor tendon entheseal soft tissue and bone changes by ultrasound can differentiate psoriatic arthritis and rheumatoid arthritis. Clin Exp Rheumatol 2018. [Epub ahead of print]
9. Tan S, Yao J, Flynn JA, *et al.* Quantitative syndesmophyte measurement in ankylosing spondylitis using CT: longitudinal validity and sensitivity to change over 2 years. Ann Rheum Dis 2015; 74:437–443.
10. Sengupta R, Marzo-Ortega H, McGonagle D, *et al.* Short-term repeat magnetic resonance imaging scans in suspected early axial spondyloarthritis are clinically relevant only in HLA-B27-positive male subjects. J Rheumatol 2018; 45:202–205.
11. Wang D, Hou Z, Gong Y, *et al.* Bone edema on magnetic resonance imaging is highly associated with low bone mineral density in patients with ankylosing spondylitis. PLoS One 2017; 12:e0189569.
12. Bethge J, Meffert S, Ellrichmann M, *et al.* Combination therapy with vedolizumab and etanercept in a patient with pouchitis and spondylarthritis. BMJ Open Gastroenterol 2017; 4:e000127.
13. Roblin X, Paul S, Ben-Horin S. Co-treatment with golimumab and vedolizumab to treat severe UC and associated spondyloarthropathy. J Crohns Colitis 2018; 12:379–380.
14. Gniadecki R, Bang B, Sand C. Combination of antitumour necrosis factor-alpha and antiinterleukin-12/23 antibodies in refractory psoriasis and psoriatic arthritis: a long-term case-series observational study. Br J Dermatol 2016; 174:1145–1146.
15. Smolen JS, Agarwal SK, Ilivanova E, *et al.* A randomised phase II study evaluating the efficacy and safety of subcutaneously administered ustekinumab and guselkumab in patients with active rheumatoid arthritis despite treatment with methotrexate. Ann Rheum Dis 2017; 76:831–839.
16. Kavanaugh A, Puig L, Gottlieb AB, *et al.* Efficacy and safety of ustekinumab in psoriatic arthritis patients with peripheral arthritis and physician-reported spondylitis: posthoc analyses from two phase III, multicentre, double-blind, placebo-controlled studies (PSUMMIT-1/PSUMMIT-2). Ann Rheum Dis 2016; 75:1984–1988.
17. Deodhar A, Gottlieb A, Boehncke W-H, *et al.* OP0218 efficacy and safety results of guselkumab, an antiil23 monoclonal antibody, in patients with active psoriatic arthritis over 24 weeks: a phase 2a, randomized, double-blind, placebo-controlled study. [Oral presentation]. Ann Rheum Dis 2017; 76.(Suppl 2).
18. Baeten D, Sieper J, Braun J, *et al.* Secukinumab, an interleukin-17A inhibitor, in ankylosing spondylitis. N Engl J Med 2015; 373:2534–2548.
19. Mease P, McInnes IB. Secukinumab: a new treatment option for psoriatic arthritis. Rheumatol Ther 2016; 3:5–29.
20. Mease PJ, van der Heijde D, Ritchlin CT, *et al.* Ixekizumab, an interleukin-17A specific monoclonal antibody, for the treatment of biologic-naive patients with active psoriatic arthritis: results from the 24-week randomised, double-blind, placebo-controlled and active (adalimumab)-controlled period of the phase III trial SPIRIT-P1. Ann Rheum Dis 2017; 76:79–87.
21. Glatt S, Baeten D, Baker T, *et al.* Dual IL-17A and IL-17F neutralisation by bimekizumab in psoriatic arthritis: evidence from preclinical experiments and a randomised placebo-controlled clinical trial that IL-17F contributes to human chronic tissue inflammation. Ann Rheum Dis 2018; 77:523–532.
22. ■■ Mease P, Hall S, FitzGerald O, *et al.* Tofacitinib or adalimumab versus placebo for psoriatic arthritis. N Engl J Med 2017; 377:1537–1550.
This is the first clinical trial to show that Tofacitinib is better than TNFi in psoriatic arthritis.
23. ■■ Gladman D, Rigby W, Azevedo VF, *et al.* Tofacitinib for psoriatic arthritis in patients with an inadequate response to TNF inhibitors. N Engl J Med 2017; 377:1525–1536.
This clinical trial shows that Tofacitinib is useful for psoriatic arthritis in patients with an inadequate response to TNF inhibitors.
24. Reed M, Crosbie D. Apremilast in the treatment of psoriatic arthritis: a perspective review. Ther Adv Musculoskelet Dis 2017; 9:45–53.
25. Ritchlin CT, Krueger JG. New therapies for psoriasis and psoriatic arthritis. Curr Opin Rheumatol 2016; 28:204–210.
26. Raychaudhuri SP, Raychaudhuri SK. Mechanistic rationales for targeting interleukin-17A in spondyloarthritis. Arthritis Res Ther 2017; 19:51.
27. Raychaudhuri SK, Saxena A, Raychaudhuri SP. Role of IL-17 in the pathogenesis of psoriatic arthritis and axial spondyloarthritis. Clin Rheumatol 2015; 34:1019–1023.
28. Lubberts E. The IL-23-IL-17 axis in inflammatory arthritis. Nat Rev Rheumatol 2015; 11:415–429.
29. Papotto PH, Ribot JC, Silva-Santos B. IL-17(+) gammadelta T cells as kick-starters of inflammation. Nat Immunol 2017; 18:604–611.
30. Gaffen SL, Jain R, Garg AV, Cua DJ. IL-23-IL-17 immune axis: discovery, mechanistic understanding, and clinical testing. Nat Rev Immunol 2014; 14:585–600.
31. Sarkar S, Justa S, Brucks M, *et al.* Interleukin (IL)-17A, F and AF in inflammation: a study in collagen-induced arthritis and rheumatoid arthritis. Clin Exp Immunol 2014; 177:652–661.
32. Appel H, Maier R, Wu P, *et al.* Analysis of IL-17(+) cells in facet joints of patients with spondyloarthritis suggests that the innate immune pathway might be of greater relevance than the Th17-mediated adaptive immune response. Arthritis Res Ther 2011; 13:R95.
33. Reveille JD, Sims AM, Danoy P, *et al.* Genome-wide association study of ankylosing spondylitis identifies non-MHC susceptibility loci. Nat Genet 2010; 42:123–127.
34. Davidson SI, Liu Y, Danoy PA, *et al.* Association of STAT3 and TNFRSF1A with ankylosing spondylitis in Han Chinese. Ann Rheum Dis 2011; 70:289–292.
35. Nossent JC, Sagen-Johnsen S, Bakland G. IL23R gene variants in relation to IL17A levels and clinical phenotype in patients with ankylosing spondylitis. Rheumatol Adv Pract 2018; 2:6.
36. Mei Y, Pan F, Gao J, *et al.* Increased serum IL-17 and IL-23 in the patient with ankylosing spondylitis. Clin Rheumatol 2011; 30:269–273.
37. Leijten EF, van Kempen TS, Boes M, *et al.* Brief report: enrichment of activated group 3 innate lymphoid cells in psoriatic arthritis synovial fluid. Arthritis Rheumatol 2015; 67:2673–2678.
38. Menon B, Gullick NJ, Walter GJ, *et al.* Interleukin-17+CD8+ T cells are enriched in the joints of patients with psoriatic arthritis and correlate with disease activity and joint damage progression. Arthritis Rheumatol 2014; 66:1272–1281.
39. Raychaudhuri SP, Raychaudhuri SK, Genovese MC. IL-17 receptor and its functional significance in psoriatic arthritis. Mol Cell Biochem 2012; 359(1–2):419–429.
40. Zhu KJ, Zhu CY, Shi G, Fan YM. Association of IL23R polymorphisms with psoriasis and psoriatic arthritis: a meta-analysis. Inflamm Res 2012; 61:1149–1154.
41. Celis R, Planell N, Fernandez-Sueiro JL, *et al.* Synovial cytokine expression in psoriatic arthritis and associations with lymphoid neogenesis and clinical features. Arthritis Res Ther 2012; 14:R93.
42. Wang X, Wei Y, Xiao H, *et al.* A novel IL-23p19/Ebi3 (IL-39) cytokine mediates inflammation in Lupus-like mice. Eur J Immunol 2016; 46:1343–1350.
43. Wang X, Zhang Y, Wang Z, *et al.* Anti-IL-39 (IL-23p19/Ebi3) polyclonal antibodies ameliorate autoimmune symptoms in lupus-like mice. Mol Med Rep 2018; 17:1660–1666.
44. Gabay C, Towne JE. Regulation and function of interleukin-36 cytokines in homeostasis and pathological conditions. J Leukoc Biol 2015; 97:645–652.
45. Ainscough JS, Macleod T, McGonagle D, *et al.* Cathepsin S is the major activator of the psoriasis-associated proinflammatory cytokine IL-36γ. Proc Natl Acad Sci 2017; 114:E2748–E2757.
46. Onoufriadis A, Simpson MA, Pink AE, *et al.* Mutations in IL36RN/IL1F5 are associated with the severe episodic inflammatory skin disease known as generalized pustular psoriasis. Am J Hum Genet 2011; 89:432–437.

47. Nishida A, Hidaka K, Kanda T, *et al.* Increased expression of interleukin-36, a member of the interleukin-1 cytokine family, in inflammatory bowel disease. Inflamm Bowel Dis 2016; 22:303–314.
48. Chi H-H, Hua K-F, Lin Y-C, *et al.* IL-36 signaling facilitates activation of the NLRP3 inflammasome and IL-23/IL-17 axis in renal inflammation and fibrosis. J Am Soc Nephrol 2017; 28:2022–2037.
49. Scheibe K, Backert I, Wirtz S, *et al.* IL-36R signalling activates intestinal epithelial cells and fibroblasts and promotes mucosal healing in vivo. Gut 2017; 66:823–838.
50. Kovach M, Levanen B, Andersson A, *et al.* Late Breaking Abstract-IL-36 receptor agonists contribute to a pro-inflammatory milieu in smokers with and without COPD. Eur Respir J 2017; 50:OA4846.
51. Ogawa Y, Kim J-C, Morasso M, Udey M. Macrophages regulate IL-17-associated skin inflammation in mice with Dlx3-deficient keratinocytes. J Dermatol Sci 2016; 84:e2.
52. Aoyagi T, Newstead MW, Zeng X, *et al.* IL-36 receptor deletion attenuates lung injury and decreases mortality in murine influenza pneumonia. Mucosal Immunol 2017; 10:1043–1055.
53. Di Caprio R, Balato A, Caiazzo G, *et al.* IL-36 cytokines are increased in acne and hidradenitis suppurativa. Arch Dermatol Res 2017; 309:673–678.
54. Carrier Y, Ma H-L, Ramon HE, *et al.* Inter-regulation of Th17 cytokines and the IL-36 cytokines in vitro and in vivo: implications in psoriasis pathogenesis. J Invest Dermatol 2011; 131:2428–2437.
55. Vigne S, Palmer G, Lamacchia C, *et al.* IL-36R ligands are potent regulators of dendritic and T cells. Blood 2011; 118:5813–5823.
56. Bridgewood C, Fearnley GW, Berekmeri A, *et al.* IL-36γ is a strong inducer of IL-23 in psoriatic cells and activates angiogenesis. Front Immunol 2018; 9:200.
57. Sun Y, Mozaffarian A, Arnett HA, *et al.* 253: IL-36 induces inflammation and collagen deposition in the lung. Cytokine 2013; 63:303.
58. Mahil SK, Catapano M, Di Meglio P, *et al.* An analysis of IL-36 signature genes and individuals with IL1RL2 knockout mutations validates IL-36 as a psoriasis therapeutic target. Sci Transl Med 2017; 9:pii: eaan2514.
59. Teng X, Hu Z, Wei X, *et al.* IL-37 ameliorates the inflammatory process in psoriasis by suppressing proinflammatory cytokine production. J Immunol 2014; 192:1815–1823.
60. Boutet MA, Najm A, Bart G, *et al.* IL-38 overexpression induces anti-inflammatory effects in mice arthritis models and in human macrophages in vitro. Ann Rheum Dis 2017; 76:1304–1312.
61. Cavalli G, Koenders M, Kalabokis V, *et al.* Treating experimental arthritis with the innate immune inhibitor interleukin-37 reduces joint and systemic inflammation. Rheumatology (Oxford) 2016; 55:2220–2229.
62. van de Veerdonk FL, Stoeckman AK, Wu G, *et al.* IL-38 binds to the IL-36 receptor and has biological effects on immune cells similar to IL-36 receptor antagonist. Proc Natl Acad Sci U S A 2012; 109:3001–3005.
63. Nold-Petry CA, Lo CY, Rudloff I, *et al.* IL-37 requires the receptors IL-18Ralpha and IL-1R8 (SIGIRR) to carry out its multifaceted anti-inflammatory program upon innate signal transduction. Nat Immunol 2015; 16:354–365.
64. Rahman P, Sun S, Peddle L, *et al.* Association between the interleukin-1 family gene cluster and psoriatic arthritis. Arthritis Rheum 2006; 54:2321–2325.
65. Chou CT, Timms AE, Wei JC, *et al.* Replication of association of IL1 gene complex members with ankylosing spondylitis in Taiwanese Chinese. Ann Rheum Dis 2006; 65:1106–1109.
66. Guo ZS, Li C, Lin ZM, *et al.* Association of IL-1 gene complex members with ankylosing spondylitis in Chinese Han population. Int J Immunogenet 2010; 37:33–37.
67. Asquith M, Davin S, Stauffer P, *et al.* Intestinal metabolites are profoundly altered in the context of HLA-B27 expression and functionally modulate disease in a rat model of spondyloarthritis. Arthritis Rheumatol 2017; 69:1984–1995.
68. Breban M, Tap J, Leboime A, *et al.* Faecal microbiota study reveals specific dysbiosis in spondyloarthritis. Ann Rheum Dis 2017; 76:1614–1622.
69. Mielants H, Veys EM, Cuvelier C, *et al.* The evolution of spondyloarthropathies in relation to gut histology. II. Histological aspects. J Rheumatol 1995; 22:2273–2278.
70. Costello ME, Ciccia F, Willner D, *et al.* Brief report: intestinal dysbiosis in ankylosing spondylitis. Arthritis Rheumatol 2015; 67:686–691.
71. Yang L, Wang L, Wang X, *et al.* A possible role of intestinal microbiota in the pathogenesis of ankylosing spondylitis. Int J Mol Sci 2016; 17:pii: E2126.
72. Stebbings S, Munro K, Simon MA, *et al.* Comparison of the faecal microflora of patients with ankylosing spondylitis and controls using molecular methods of analysis. Rheumatology (Oxford) 2002; 41:1395–1401.
73. Rashid T, Ebringer A. Ankylosing spondylitis is linked to Klebsiella: the evidence. Clin Rheumatol 2007; 26:858–864.
74. Rashid T, Ebringer A, Tiwana H, Fielder M. Role of Klebsiella and collagens in Crohn's disease: a new prospect in the use of low-starch diet. Eur J Gastroenterol Hepatol 2009; 21:843–849.
75. Scher JU, Ubeda C, Artacho A, *et al.* Decreased bacterial diversity characterizes the altered gut microbiota in patients with psoriatic arthritis, resembling dysbiosis in inflammatory bowel disease. Arthritis Rheumatol 2015; 67:128–139.
76. Manasson J, Shen N, Garcia Ferrer HR, *et al.* Gut microbiota perturbations in reactive arthritis and postinfectious spondyloarthritis. Arthritis Rheumatol 2018; 70:242–254.
77. Dorofeyev AE, Vasilenko IV, Rassokhina OA. Joint extraintestinal manifestations in ulcerative colitis. Dig Dis 2009; 27:502–510.
78. Ranganathan V, Ciccia F, Zeng F, *et al.* Macrophage migration inhibitory factor induces inflammation and predicts spinal progression in ankylosing spondylitis. Arthritis Rheumatol 2017; 69:1796–1806.
79. de Koning A, de Bruin F, van den Berg R, *et al.* Low-dose CT detects more progression of bone formation in comparison to conventional radiography in patients with ankylosing spondylitis: results from the SIAS cohort. Ann Rheum Dis 2018; 77:293–299.
80. ■■ El-Zayadi AA, Jones EA, Churchman SM, *et al.* Interleukin-22 drives the proliferation, migration and osteogenic differentiation of mesenchymal stem cells: a novel cytokine that could contribute to new bone formation in spondyloarthropathies. Rheumatology (Oxford) 2017; 56:488–493.

This study shows the contribution of IL-22 in bone formation in spondyloarthropathies.

81. Ono T, Okamoto K, Nakashima T, *et al.* IL-17-producing γδ T cells enhance bone regeneration. Nat Commun 2016; 7:10928.
82. Croes M, Öner FC, van Neerven D, *et al.* Proinflammatory T cells and IL-17 stimulate osteoblast differentiation. Bone 2016; 84:262–270.
83. Watad A, Cuthbert RJ, Amital H, *et al.* Enthesitis: much more than focal insertion point inflammation. Curr Rheumatol Rep 2018; 20:41.
84. Cuthbert RJ, Fragkakis EM, Dunsmuir R, Li Z, *et al.* Brief Report: Group 3 innate lymphoid cells in human enthesis. Arthritis Rheumatol 2017; 69: 1816–1822.