Combating physical inactivity during the COVID-19 pandemic

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Physical inactivity is common during periods of self-isolation, but for patients with rheumatic diseases, there are crucial benefits to be gained from maintaining an active lifestyle throughout the COVID-19 pandemic. Patients should be provided with support to maintain physical activity and avoid prolonged periods of time spent sitting.

COVID-19, a disease caused by the SARS-CoV-2 virus, has been classified as a pandemic by the World Health Organization (WHO). In an effort to slow infection rates, particularly in groups predisposed to high risks of morbidity and mortality, extensive social distancing and isolation policies have been adopted worldwide.

Patients with rheumatic diseases commonly have a higher risk of serious infections than the general population owing to immunosuppression and disease activity, particularly in those with suboptimal control of disease activity[1]. Moreover, some patients are at increased risk of COVID-19 complications as they are older and have concomitant chronic diseases. Therefore, patients are recommended by national health advisory services to continue with their current treatment to prevent disease flares and practice self-isolation to prevent infection during the COVID-19 pandemic.

Physical inactivity (not meeting the physical activity guidelines) and sedentary behaviour (too much sitting) are highly prevalent in patients with rheumatic diseases; previous studies show that 38–72% are physically inactive, and sedentary time ranges between 8.3–14.0 hours/day, which is higher than for the general population (31% and ~7.5 hours/day, respectively)[2]. Importantly, both of these behaviours are associated with poor disease-related outcomes (such as high disease activity, pain and fatigue) and cardiometabolic risk factors (such as obesity and insulin resistance) in patients with rheumatoid arthritis[2]. Among patients with rheumatic diseases, cardiovascular diseases are the leading cause of morbidity and mortality and are likely to be exacerbated by physical inactivity and prolonged periods of sitting. Children and adolescents with rheumatic diseases are also commonly hypoactive compared with healthy peers owing to disease manifestations and symptoms and to overprotection by parents and health-care professionals[3]. The negative clinical effects of inactivity for paediatric patients include muscle atrophy and weakness, fatigue, obesity, insulin resistance, dyslipidaemia, reduced physical capacity and function and poor quality of life[3].

The COVID-19 pandemic has created an environment that promotes reduced amounts of habitual physical activity owing to self-isolation and quarantine requirements, reduced opportunities to remain physically active and fear of being infected. Sustained physical inactivity and sedentary behaviour are typically associated with poor physical and mental health and increased disease-specific and all-cause mortality risk[4]. Even brief periods of exposure to these behaviours can be deleterious; for example, a 2-week reduction in daily steps from ~10,000 to ~1,500 steps lead to impaired insulin sensitivity and lipid metabolism, increased visceral fat and decreased fat-free mass and cardiovascular fitness in healthy adults[5]. Interestingly, a bout of moderate-intensity exercise does not counteract the detrimental effects of 4 days of inactivity, suggesting that individuals can become 'resistant' to well-known exercise-induced metabolic adaptations[6].

In the past, bed rest was extensively used as part of the treatment for rheumatic diseases such as rheumatoid arthritis and myositis to avoid disease exacerbation and/or joint destruction. However, compelling evidence shows that disuse actually leads to joint destruction, muscle weakness and atrophy, causing reduced physical function[2,3]. Such evidence has led to the contemporary abrogation of the 'bed rest paradigm' for rheumatic diseases and has guided new approaches to clinical practice that emphasize physical activity as an important part of therapy to improve patients' symptoms[7]. Given that physical inactivity and sedentary behaviour will potentially increase as a function of social distancing measures during the COVID-19 pandemic, patients with rheumatic diseases who are already hypoactive might be at particular risk of worsened disease activity and symptoms, general comorbidities and poor quality of life (FIG. 1).

Exercise (structured physical activity) is advocated by EULAR as an integral part of standard care for patients with inflammatory arthritis and osteoarthritis[7]. Moreover, exercise improves disease symptoms, cardiovascular

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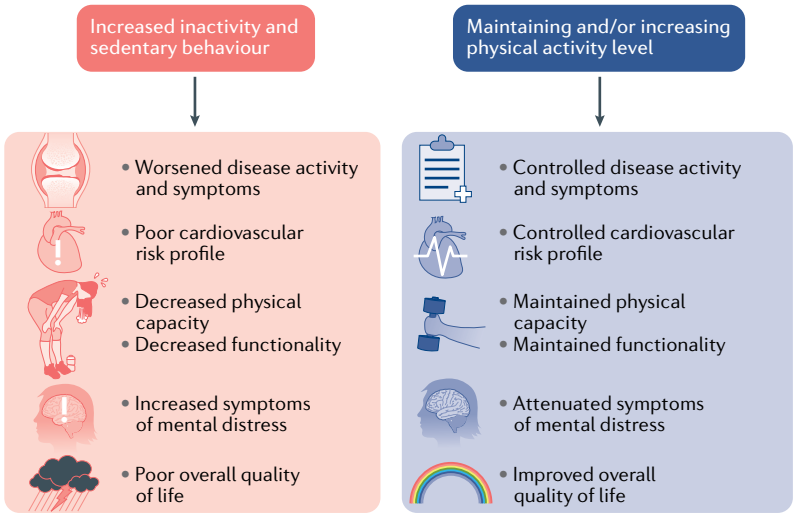

Fig. 1 | **The effects of physical inactivity during the COVID-19 pandemic on patients with rheumatic diseases.** Imposed physical inactivity and sedentary behaviour (sitting) can be disruptive to disease parameters, cardiovascular risk profile, physical capacity and function and mental health in patients with rheumatic diseases, resulting in poor quality of life, whereas maintaining more optimal physical activity levels can help to ameliorate these detrimental effects.

risk factors, physical capacity and function, mental health and quality of life for patients with other rheumatic diseases[8]. From a safety perspective, acute and long-term exercise did not trigger inflammation in a study involving patients with systemic lupus erythematosus[9]. Of relevance in the context of the COVID-19 pandemic, home-based exercise programmes are feasible and can be effective in promoting health benefits for patients with rheumatic diseases without causing any important adverse events[2,3,8] (FIG. 1).

Guided by this evidence, we recommend that clinicians and other health-care practitioners are proactive in prescribing physical activities to their patients during the COVID-19 pandemic. Several scientific and medical organizations are promoting messages to keep people active at home during the COVID-19 pandemic, including the WHO[10]. We highly recommend that associations within the field of rheumatology embrace this idea and help to spread the message that exercise is therapeutic to patients with rheumatic diseases.

What physical activity should be prescribed during self-isolation? In general, patients with rheumatic diseases can follow the current guidelines for the general population. If patients are physically inactive and have no previous experience with exercise programmes, they should engage in less intensive exercise regimes and progress slowly. If patients are already physically active, then they can either maintain their exercise routine, if viable (for example walking), or adapt their activities to be performed at home. A flexible clinical guide for physical activity promotion during the COVID-19 pandemic that can be downloaded as an electronic practitioner resource or shared with patients as a printed pamphlet, electronic document or wall poster is provided as Supplementary Fig. 1. Given patient and practitioner infection concerns about clinical contact, the file can also be adapted for use as a visual resource in telemedicine consultations.

For patients with particular physical limitations, uncontrolled disease activity and/or who are at high risk of injury when taking unsupervised exercise, home-based exercise programmes designed for the general population might not be ideal from a safety perspective. With these at-risk patients, strategies such as 'move more and sit less during the day' can be promoted as safe and accessible options to at least attenuate the deleterious effects of imposed inactivity during self-isolation. Importantly, simple strategies such as breaking up prolonged sedentary time (such as 2 minutes of walking for every 30 minutes of sitting) could improve some symptoms and cardiometabolic risk factors[2]. As children and adolescents with rheumatic diseases are commonly hypoactive, physical activity should be also recommended to paediatric patients with rheumatic disease[3].

Patients with rheumatic diseases are already at an increased risk of being hypoactive, which will probably be further aggravated by self-isolation measures imposed to tackle the spread of SARS-CoV-2. This issue raises pressing concerns, as substantial reductions in physical activity can be detrimental to disease outcomes, cardiovascular risk factors, physical capacity and mental health. In light of the systemic benefits of physical activity for patients with rheumatic diseases (FIG. 1), health practitioners are therefore encouraged to be proactive in promoting appropriate physical activity for these patients.

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Competing interests
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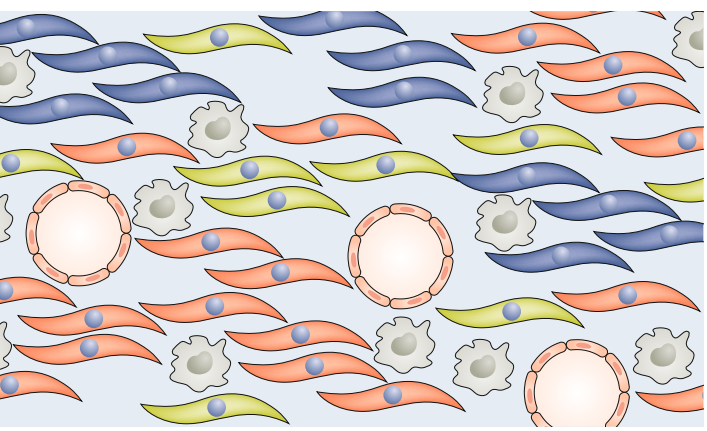
RHEUMATOID ARTHRITIS

Synovial fibroblast expansion in RA is driven by Notch signalling

> endothelium-derived Notch signalling contributes to the differentiation and expansion of synovial fibroblasts in RA

Research now published in *Nature* provides new insights into the heterogeneity of fibroblasts in the synovium in rheumatoid arthritis (RA), with direct relevance to RA pathology and the therapeutic targeting of these cells. The findings suggest that endothelium-derived Notch signalling contributes to the differentiation and expansion of synovial fibroblasts in RA, and that modulation of this signalling pathway could attenuate inflammation and joint damage.

Previous work by the researchers had shown that distinct subsets of fibroblasts in RA correlated with distinct functions, and that fibroblasts found in the synovial lining were predominantly responsible for driving joint injury whereas fibroblasts in the sublining layer were responsible for driving inflammation. Building on these findings, the new research suggests that lining and sublining fibroblasts are not separated into entirely distinct clusters but exist along a gradient that corresponds to the anatomical localization of the fibroblasts in the synovium, regulated in part by Notch signalling.



Credit: Springer Nature Limited

"Here we used single cell RNA-seq data to take an unbiased look at fibroblasts and examined their whole transcriptome," explains co-corresponding author Soumya Raychaudhuri. Trajectory transcriptional analyses demonstrated that expression of the synovial lining marker *PRG4* (encoding lubricin) and the sublining marker *THY1* (encoding CD90) gradually changes along the continuum of synovial fibroblast states.

Anatomically, expression of *THY1* was linked with proximity to the endothelium; this positional identity was lost after serial passages ex vivo, suggesting it was determined by the local microenvironment rather than cell-intrinsic factors.

Ligand–receptor analysis of synovial tissue and tissue organoid single-cell RNA-seq datasets suggested endothelium-derived Notch signalling as a potential pathway for driving the differentiation of *THY1*-expressing fibroblasts. In vitro, endothelial cells (which express Notch ligands) induced the expression of NOTCH3 and its ligand Jagged 1 in fibroblasts in a Notch-dependent manner, thereby inducing a fibroblast positional gradient. "Distance from the nearest blood vessel can now be appreciated to be key in understanding fibroblast heterogeneity in the RA synovium, with direct relevance to RA pathology," says co-corresponding author Michael Brenner.

Notably, the researchers found a higher proportion of NOTCH3-positive fibroblasts in synovial tissue from patients with RA than in tissue from patients with osteoarthritis, as well as upregulation of Notch target genes in RA synovial tissue.

The researchers then tested whether the genetic deletion of *Notch3* or antibody-mediated blockade of NOTCH3 signalling could attenuate arthritis in mice. "Both approaches abrogated inflammation and joint damage in a mouse model of inflammatory arthritis," recounts Brenner. In the K/BxN serum transfer model, arthritis activity and paw swelling was reduced in *Notch3*$^{-/-}$ mice as compared with wild-type mice. Furthermore, twice-weekly administration of an antibody against NOTCH3 attenuated arthritis severity and joint swelling in wild-type mice, compared with an isotype control antibody.

Raychaudhuri anticipates that researchers will increasingly be examining high-dimensional single-cell data from inflamed tissues, including synovial tissues; such studies could reveal novel therapeutic modalities, particularly in stromal cells, which have been relatively understudied. "In the future it may be possible to apply these same (or similar) technologies to patient samples to prioritize therapies that target specifically the most active or aberrant cell types," says Raychaudhuri.

"Defining fibroblast heterogeneity provides new insight into which cell states and the pathways they express are linked to disease pathology in RA and other diseases," adds Brenner. "This knowledge will better define selective targets for therapeutic intervention against fibroblasts."

Sarah Onuora

ORIGINAL ARTICLE Wei, K. et al. Notch signalling drives synovial fibroblast identity and arthritis pathology. *Nature* https://doi.org/10.1038/s41586-020-2222-z (2020)

IN BRIEF

RHEUMATOID ARTHRITIS

Early referral matters for RA outcomes

Among patients with a diagnosis of rheumatoid arthritis (RA) in the Leiden Early Arthritis Clinic and the French ESPOIR cohorts followed for 7–10 years, those who first met with a rheumatologist within 6 weeks of symptom onset were more likely to achieve sustained DMARD-free remission than those who were seen by a rheumatologist 7–12 weeks after symptom onset (HR 1.69; 95% CI 1.10–2.57) or more than 12 weeks after symptom onset (HR 1.67; 95% CI 1.08–2.58). Radiographic progression was more severe in those not seen within 12 weeks of symptom onset, but was similar in the other two groups.

ORIGINAL ARTICLE Niemantsverdriet, E. et al. Referring early arthritis patients within 6 weeks versus 12 weeks after symptom onset: an observational cohort study. *Lancet Rheumatol.* https://doi.org/10.1016/S2665-9913(20)30061-8 (2020)

SYSTEMIC LUPUS ERYTHEMATOSUS

Increased risk of infection-related death in SLE

In a retrospective study using the National Health Insurance Fund of Hungary database, the rate of death was higher in adults with systemic lupus erythematosus (SLE) than in matched individuals without SLE (standardized mortality ratio (SMR) 1.63; 95% CI 1.43–1.83) and even higher in the subgroup of patients with SLE who had received treatment within the first 6 months of diagnosis (SMR 2.09; 95% CI 1.80–2.39). Infection-related deaths were more common in the patients with SLE compared with the non-SLE group, attributable largely to an increased frequency of sepsis being the cause of death in the patients with SLE.

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SPONDYLOARTHRITIS

Anti-TNF response falls short in real-world cohort

In a study of participants in the British Society for Rheumatology Biologics Register for Ankylosing Spondylitis (BSRBR-AS), 51.3% of those with axial spondyloarthritis (axSpA) commencing treatment with TNF inhibitors reported a positive response, a lower proportion than that reported in clinical trials (61.7%). Compared with the real-world BSRBR-AS cohort, participants in the clinical trials were more likely to be male, HLA-B27 positive and younger (by approximately 6 years). Disease activity was similar in both groups but the BSRBR-AS participants reported poorer function prior to commencing treatment.

ORIGINAL ARTICLE Jones, G. T. et al. Real-world evidence of TNF inhibition in axial spondyloarthritis: can we generalise the results from clinical trials? *Ann. Rheum. Dis.* https://doi.org/10.1136/annrheumdis-2019-216841 (2020)

VASCULITIS

MMF comparable to cyclophosphamide in AAV

As a remission induction therapy in patients with antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis (AAV), the therapeutic efficacy of mycophenolate mofetil (MMF) is similar to that of cyclophosphamide, according to a meta-analysis of data from four randomized controlled trials. In the studies, which enrolled a total of 300 patients with AAV, MMF and cyclophosphamide led to similar rates of remission at 6 months, ANCA negativity at 6 months and long-term relapse. Rates of death among patients with AAV were similar with both treatments.

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RHEUMATOID ARTHRITIS

Disease onset goes with its gut in RA

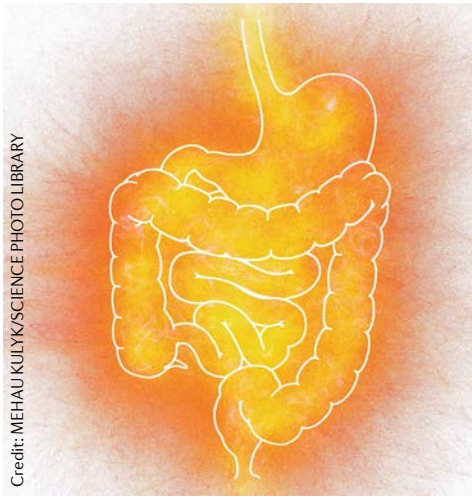
The mucosal origins hypothesis postulates that, in rheumatoid arthritis (RA), disease begins to develop at mucosal sites such as the gums, lungs and intestines and then transitions to involve synovial joints. Links between gut microbiota dysbiosis and RA, and between dietary intake of short-chain fatty acids (SCFAs) and autoimmune arthritis in mice, have provided support for this hypothesis, but a direct link between mucosal sites and the transition from systemic autoimmunity to arthritis has been missing.

A new study published in *Nature Communications* has revealed a role for intestinal barrier function, and specifically for zonulin, a precursor of haptoglobin 2 that controls epithelial tight junction permeability, in regulating the onset of joint disease in mice with collagen-induced arthritis (CIA) and potentially also in patients with RA.

Mice with CIA had increased intestinal permeability in the period between the induction of autoimmunity and the onset of clinical symptoms, which corresponded with a rise in serum zonulin. This reduction in intestinal barrier function was accompanied by an influx of effector T cells in the small intestine. Interestingly, arthritis only developed in mice that had this increased intestinal permeability.

Reducing intestinal permeability in the period before clinical arthritis, either by dietary supplementation with the SCFA butyrate, treatment with a selective intestinal cannabinoid receptor 1 agonist (cannabinoid receptor 1 regulates intestinal epithelial barrier function) or treatment with larazotide acetate (which blocks zonulin and is currently in phase III clinical trials for coeliac disease) delayed disease onset and reduced the severity of arthritis.

"The most significant finding is that improving the intestinal barrier function in mice with non-clinical CIA positively affects subsequent disease onset and severity," states corresponding author Mario Zaiss. "These results relate nicely to data published very recently by other groups showing mucosal inflammation in animal models of arthritis, and complete these interesting studies by providing a treatment opportunity."



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In a cohort of patients with RA-specific autoimmunity but no clinical symptoms (described as pre-RA), Zaiss and colleagues found evidence of intestinal barrier dysfunction and an increase in serum zonulin concentrations that correlated with the risk of developing RA. These results imply that zonulin could be studied further as a biomarker to predict disease onset in patients with pre-RA.

"Similar intestinal phenotypes could be found in individuals with pre-RA to those present in mice with CIA, and as improving the intestinal barrier function had clinical relevance on subsequent disease onset and severity in mice, we are currently planning the first studies to translate these findings to humans," says Zaiss.

Joanna Clarke

ORIGINAL ARTICLE Tajik, N. et al. Targeting zonulin and intestinal epithelial barrier function to prevent onset of arthritis. *Nat. Commun.* **11**, 1995 (2020)

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OSTEOARTHRITIS

TET1: an epigenetic controller of OA

Osteoarthritis (OA) is a heterogeneous disease mediated by multiple molecular pathways and governed by a complex interplay between various genetic, epigenetic and environmental factors. New findings published in *Science Translational Medicine* implicate the epigenetic regulator ten-eleven translocation 1 (TET1) as an important activator of multiple OA-associated pathways and as an attractive therapeutic target.

TET enzymes catalyse the initial step of DNA demethylation by converting 5-methylcytosine into 5-hydroxymethylcytosine (5hmC), an epigenetic process associated with gene activation. Previous evidence had shown that 5hmC accumulates on OA-related genes in osteoarthritic chondrocytes. To investigate this process further, the authors of the new study mapped changes in the 5hmC epigenome in mice following induction of OA by destabilization of the medial meniscus (DMM), with and without the expression of *Tet1*.

In wild-type mice, OA induction was accompanied by a genome-wide accumulation of 5hmC, predominantly in gene bodies or intergenic regions, and an upregulation in expression of hundreds of genes. Notably, almost half of the upregulated genes gained sites of 5hmC accumulation, including genes involved in WNT signalling, protein kinase A signalling and inositol metabolism.

The majority of 5hmC deposition was lost in mice lacking TET1. Importantly, loss of *Tet1* impeded the initiation and development of DMM-induced OA, including the deterioration of cartilage and osteophyte formation.

> “OA induction was accompanied by a genome-wide accumulation of 5hmC”

TET1 activated various pathways important in OA pathogenesis, including WNT signalling, metalloproteinases and STAT3 signalling. Indeed, shRNA-mediated knockdown of TET1 in chondrocytes from patients with OA decreased the expression of *MMP3* and *MMP13*.

To provide proof of principle that modulating TET1 activity is a promising therapeutic strategy, the researchers tested a small molecular inhibitor of TET1, 2-hydroxyglutarate (2-HG). Intra-articular injection of 2-HG after DMM surgery stalled OA progression in mice, and this inhibitor could replicate the effects of TET1 knockdown in osteoarthritic chondrocytes in vitro.

Jessica McHugh

ORIGINAL ARTICLE Smeriglio, P. et al. Inhibition of TET1 prevents the development of osteoarthritis and reveals the 5hmC landscape that orchestrates pathogenesis. *Sci. Transl Med.* **12**, eaax2332 (2020)
RELATED ARTICLE Rice, S. J. et al. Interplay between genetics and epigenetics in osteoarthritis. *Nat. Rev. Rheum.* **16**, 268–281 (2020)

RHEUMATOID ARTHRITIS

Targeting FLS signalling in RA

Fibroblast-like synoviocytes (FLS) can promote joint inflammation and destruction in rheumatoid arthritis (RA) through the production of pro-inflammatory mediators such as IL-15 and dickkopf-related protein 1 (DKK1). New findings published in *Arthritis & Rheumatology* highlight the involvement of a signalling axis downstream of discoidin domain receptor 2 (DDR2) in this process.

Previous studies had suggested that DDR2, a receptor tyrosine kinase (RTK), is expressed in FLS and contributes to cartilage and bone destruction in RA. In the new study, the researchers found that the expression of DDR2 correlated with the expression of IL-15 and DKK1 both in FLS from patients with RA (RA FLS) and in mice with collagen antibody-induced arthritis (CAIA).

Following collagen antibody treatment, *Ddr2*$^{-/-}$ mice had milder arthritis than wild-type mice and reduced expression of both IL-15 and DKK1. Restoring the expression of DDR2 in the joints of *Ddr2*$^{-/-}$ mice using a DDR2-expressing adenovirus increased the arthritis severity score as well as the expression of IL-15 and DKK1.

> “treatment with WRG-28 was associated with reduced clinical arthritis scores”

In vitro experiments in RA FLS identified the long non-coding RNA H19 as a downstream target of the DDR2 signalling cascade. H19 could then interact with and downregulate miR-103a, a microRNA previously shown to be downregulated in RA FLS. Notably, the predicted targets of this microRNA included *IL15* and *DKK1*, and, indeed, data from dual luciferase reporter assays suggested that miR-103a could directly target and repress the expression *IL15* and *DKK1*.

“DDR2 can be blocked by several FDA-approved RTK inhibitors, such as dasatinib and imatinib, and a recent study has shown that inhibition of DDR2 by dasatinib attenuates inflammation severity and bone destruction in mice with CAIA and RA FLS,” explains corresponding author Wei Zhang. Given the low specificity of dasatinib for DDR2, Zhang and colleagues explored the potential of a more recently developed small molecule inhibitor of DDR2, WRG-28.

In the CAIA model, treatment with WRG-28 was associated with reduced clinical arthritis scores, as well as reduced levels of inflammatory cell infiltration and destruction of cartilage and bone. In line with the proposed DDR2–H19–miR-103a signalling axis, WRG-28 treatment was also associated with decreased expression of H19, IL-15 and DKK1 and increased expression of miR-103 in the ankle joints of the mice.

Jessica McHugh

ORIGINAL ARTICLE Mu, N. et al. Blockade of discoidin domain receptor 2 as a strategy for reducing inflammation and joint destruction in rheumatoid arthritis via altered interleukin-15 and Dkk-1 signaling in fibroblast-like synoviocytes. *Arthritis Rheumatol.* https://doi.org/10.1002/art.41205 (2020)
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TARGETED THERAPY

Harnessing plant viruses to treat autoimmune diseases

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The induction of immune tolerance is a promising approach for treating autoimmune diseases. Several strategies have been trialled to induce immune tolerance, including the use of antigen-specific peptide vaccines, which have had some success but can trigger unwanted immune responses owing to the use of an adjuvant. The results of a new study suggest that plant viruses could be harnessed to deliver specific peptides without the need for an adjuvant, thereby improving the efficacy of peptide vaccines.

Nanoparticles created from plant virus coat protein subunits can be genetically engineered to express an antigen-specific peptide related to an autoimmune disease. These nanoparticles can then be grown in their natural plant hosts (a technique known as molecular farming) before being collected for use.

> “Administration of these nanoparticles to mice with collagen-induced arthritis reduced their symptoms”

“We started working 20 years ago on molecular farming as an enabling technology to solve the challenges posed by autoimmune diseases, in particular those related to tolerance induction,” says corresponding author Linda Avesani. “In our new paper, we demonstrated for the first time that bio-designed nanomaterials can be used to prevent autoimmune diabetes and to treat autoimmune arthritis.”

Avesani and colleagues created nanoparticles from tomato bushy stunt virus (TBSV) that expressed one of two peptides (pLIP1 or pFADK2), which had previously been identified in a peptide library screen as being immunodominant in patients with seronegative rheumatoid arthritis. Administration of these nanoparticles to mice with collagen-induced arthritis reduced their symptoms (and in the case of pLIP1, abolished them completely) compared with treatment with the same peptide and an adjuvant or with saline, and produced similar or better results to treatment with the glucocorticoid dexamethasone.

In addition, the researchers tested wild-type TBSV on its own, which was also able to reduce the symptoms of arthritis, albeit to a lesser degree than the peptide-engineered nanoparticles, suggesting that TBSV has innate immunomodulatory properties. “These results indicate that the virus structure acts both as a carrier (stabilizing the peptide) and as an adjuvant,” explains Avesani.

Similar results were also achieved in a model of autoimmune diabetes using a different plant virus. Avesani and colleagues hope to expand their use of this technique to other autoimmune diseases and to develop the nanoparticles for use in humans.

Joanna Clarke

ORIGINAL ARTICLE Zampieri, R. et al. Prevention and treatment of autoimmune diseases with plant virus nanoparticles. *Sci. Adv.* **6**, eaaz0295 (2020)

RHEUMATOID ARTHRITIS

Uncovering the pro-resolving gene network in RA

Credit: Yuichiro Chino/Moment

Resolution of inflammation is important for restoring tissue homeostasis, and failure to resolve can lead to chronic inflammatory diseases such as rheumatoid arthritis (RA). New findings shed light on this dynamic process, including the identification of three previously unknown pro-resolving factors, which could guide the development of new therapies and biomarkers for predicting disease remission.

“A variety of cellular processes contribute to anti-inflammatory responses,” explains Wan-Uk Kim, corresponding author on the new study. “Despite advancements in our understanding of inflammation resolution, global analyses have not been sufficiently explored to systematically discover the factors or pathways underlying resolution of chronic inflammatory diseases.”

To address this issue, Kim and colleagues used a systems biology approach to characterize the temporal changes in synovial gene expression profiles of mice with collagen-induced arthritis (CIA), including during the induction, peak and resolution phases of disease. Network analysis of differentially expressed genes across these phases identified three genes associated with spontaneous resolution of CIA: *Itgb1*, *Rps3* and *Ywhaz*.

All three genes encoded secretory proteins that could suppress the production of pro-inflammatory cytokines (such as TNF and IL-6) by a variety of effector cells in vitro, including by macrophages and fibroblast-like synoviocytes.

Notably, levels of YWHAZ were upregulated in the sera of mice during the resolution phase, suggesting that this pro-resolving factor could serve as a clinical biomarker. Indeed, in patients with RA, urinary levels of YWHAZ were increased following 4–6 months treatment with anti-rheumatic drugs, but only in those patients who had responded well to treatment and not in patients with a moderate or no response.

> “intra-articular injection of an *Ywhaz*-containing adenovirus suppressed progression of disease”

YWHAZ also showed potential as a therapeutic target. In mice with CIA, intra-articular injection of an *Ywhaz*-containing adenovirus suppressed progression of disease, including synovial inflammation, joint destruction and levels of pro-inflammatory cytokines.

Jessica McHugh

ORIGINAL ARTICLE Kong, J.-S. et al. Dynamic transcriptome analysis unveils key pro-resolving factors of chronic inflammatory arthritis. *J. Clin. Invest.* https://doi.org/10.1172/JCI126866 (2020)

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RHEUMATOID ARTHRITIS

Does methotrexate improve TNF inhibitor drug survival in elderly patients with RA?

Clément Lahaye and Martin Soubrier

Methotrexate is commonly used in combination with biologic DMARDs in the treatment of rheumatoid arthritis on the basis that the combined therapy has synergistic benefits. New data challenge this concept in the treatment of older adults and highlight the uncertainty of the mode of action of methotrexate in this population.

Refers to Bechman, K. et al. Is background methotrexate advantageous in extending TNF inhibitor drug survival in elderly patients with rheumatoid arthritis? An analysis of the British Society for Rheumatology Biologics Register. *Rheumatology* https://doi.org/10.1093/rheumatology/kez671 (2020).

For 40 years, methotrexate has been a cornerstone treatment for rheumatoid arthritis (RA), and this drug remains the most commonly used DMARD. However, multiple mechanisms contribute to the immunosuppressive anti-inflammatory effects of methotrexate in rheumatic diseases, and many secrets remain to be revealed[1]. The past 20 years have seen the gradual introduction of biologic DMARDs (bDMARDs), which have considerably expanded the therapeutic possibilities in RA, particularly for patients for whom methotrexate alone does not provide a good clinical response. Even in these patients, methotrexate is generally still continued owing to the improved efficacy, without necessarily a worsening toxicity, of combined bDMARD and methotrexate therapy compared with bDMARD monotherapy[2]. Indeed, EULAR recommends treatment with a combination of conventional synthetic DMARDs and bDMARDs[3]. However, does this concept hold true for all populations of patients? New findings from Bechman et al.[4] challenge this strategy for elderly (aged >75 years) patients with RA.

The synergistic effect of methotrexate with bDMARDs is partly attributed to the ability of methotrexate to prevent the formation of antibodies that target bDMARDs. Consequently, through its effects on anti-drug antibodies, methotrexate might prevent or slow down a loss of response to bDMARDs[5]. However, the immunogenicity-limiting effect of methotrexate has only been observed in very few studies, and immunogenicity itself is not equivalent among different bDMARDs[5]. Moreover, the immunogenicity of biologic therapies might be reduced in older individuals, owing to a gradual deterioration in the immune system's capacity to produce specific antibodies owing to immunosenescence. The effect of this aspect of immunosenescence on drug efficacy, and the possible implication of these effects in clinical practice, has, until now, not been explored in RA, owing to the insufficient proportions of older individuals included in previous studies.

> these results highlight important gaps in our knowledge that require further investigation

The treatment continuation rate (also known as drug survival) is sometimes used as an indicator of the long-term benefit-to-risk ratio of a drug, because a primary lack of efficacy or secondary loss of efficacy, adverse effects or administrative constraints of a drug can result in drug discontinuation. In the new study, Bechman et al.[4] investigated the drug continuation rate in patients with RA by analysing data from the British Society for Rheumatology Biologics Registry, a prospective observational study of 15,700 biologic-naive patients with RA. As expected, analysis of the overall cohort showed that TNF inhibitor monotherapy was associated with a higher rate of TNF inhibitor discontinuation than treatment with a TNF inhibitor combined with methotrexate (hazard ratio (HR) 1.12, 95% CI 1.06–1.18; $P < 0.001$). They hypothesized that the benefits, in terms of drug survival, of combination treatment compared with monotherapy that were observed over the entire cohort could be absent in elderly patients. To test their hypothesis, the authors focused on the 5% of patients in the cohort who were aged ≥75 years, a population with a higher frequency of comorbidities than

the population of patients aged <75 years. Overall, the older patients were more likely than the younger patients to discontinue treatment; the dropout rates per 100 patient years were 25.5 (95% CI 23.2–27.9) and 18.1 (95% CI 17.7–18.5), respectively. Notably, unlike with the younger patients, the older patients did not gain overall drug survival benefit with the combination therapy compared with TNF inhibitor monotherapy. Detailed analysis of the cause of drug discontinuation found that in the older individuals, the rate of treatment discontinuation caused by drug ineffectiveness was lower for patients on monotherapy than for patients on combination therapy (HR 0.66, 95% CI 0.43–0.99; $P = 0.04$), but that this benefit of TNF inhibitor monotherapy was compensated for by a much higher rate of treatment discontinuation rate caused by adverse events (HR 1.41, 95% CI 1.02–1.96; $P = 0.04$).

Bechman et al.[4] interpreted the good drug survival of monotherapy as an argument in favour of the notion that immunogenicity declines with age, owing to immunosenescence, which could obviate the need to continue treatment with methotrexate. However, various factors make the interpretation of these results difficult, and these results do not necessarily mean that monotherapy provided a better benefit-to-risk ratio. Indeed, the authors also rightly pointed out that the main cause of treatment discontinuation in the older individuals was the occurrence of adverse reactions. Unfortunately, stopping treatment because of an adverse event prevents any evaluation of risk of drug ineffectiveness in those patients, which could bias the results. The statistical methods used in that study might have only partially overcome this bias.

Another limitation to consider is potential bias caused by concurrent drug use. Corticosteroid therapy is a major, dose-dependent risk factor for adverse events, particularly infections, and has constituted a notable source of bias in numerous studies. In the new study, the exposure to corticosteroids was highest in the individuals aged ≥75 years (52% versus 39%; $P < 0.001$), including the individuals on TNF inhibitor monotherapy[5]. Although the difference in distribution of patients on corticosteroid therapy in the older and younger population was taken into account by the multivariate adjustment, this adjustment only considered the exposure of corticosteroids but not their dose. In addition, the corticosteroid-sparing effect of the two strategies could not be taken into account, because the analysis only considered glucocorticoid use at baseline (that is, at the time of biologic initiation).

Surprisingly, Bechman et al.[4] also did not specify the prescribed dose or route of administration of methotrexate and did not mention compliance issues. The methotrexate dose can influence both the risk of adverse events and the modulation of immunogenicity and, therefore, drug survival. In previous studies, the tolerated dose of methotrexate was lower in older individuals than in younger individuals, owing in particular to restrictions in drug elimination linked with renal impairment in older individuals[6]. Hence, differences in the route of methotrexate administration or inadequate doses in older patients could have contributed to the differences in drug survival between the two groups.

> “we need to develop specific treatment regimens that are adapted to the older population”

Most studies that have evaluated the effects of age on DMARD efficacy or tolerance have considered the 20 to 30% of patients over 65. In the study by Bechman et al.[4], the chosen threshold of 75 years was more ambitious; this threshold was justified by the desire to study a population that has a high chance of experiencing the effects of immunosenescence. In fact, the persisting benefit of combined treatment, in terms of drug survival, for patients aged 65–74 years seems to validate this threshold. However, it should be noted that immunosenescence is not a linear process linked with age; instead, this process is greatly influenced by associated pathologies, such as RA. In addition, a simple, specific marker for immunosenescence is currently lacking, as well as a standardized test for the detection of anti-drug antibodies. Hence, these results highlight important gaps in our knowledge that require further investigation.

The health status of the older population is very heterogeneous, and characteristics other than chronologic age can affect response to treatment. Bechman et al.[4] took comorbidities into account in their analyses, and the authors' intentions to account for the specific features of older patients is commendable. However, most assessments in older populations, including the one by Bechman et al.[4], unfortunately largely neglect the physical parameters of frailty (such as unintentional weight loss, self-reported exhaustion, grip strength muscle weakness, slow walking speed and low physical activity), which are well known prognosis factors associated with loss of autonomy and mortality as well as with immunosenescence[7–9].

Overall, these results have revived interest in the use of TNF inhibitors as monotherapy for elderly patients with RA, particularly those patients affected by polypharmacy and methotrexate toxicity. Several aspects of the study design limit immediate applicability of the results and hence require further investigation. These results should be confirmed in other cohorts, or in a randomized controlled trial, to overcome prescription bias. In addition, the potential efficacy of monotherapy in the older population should be explored for other biologic treatments (particularly abatacept, rituximab and tocilizumab). Indeed, to cope with the ageing population of patients with RA, we need to develop specific treatment regimens that are adapted to the older population. By questioning the dogma that bDMARDs plus methotrexate provide benefit to the older population, Bechman et al.[4] have forced us to move towards more specific strategies, which should hopefully lead to many more studies.

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IMMUNOLOGY

Learning from similarities between vaccine responses and SLE

Marie Wahren-Herlenius and Lars Rönnblom

Baseline immune variation predicts immune responses during vaccination, and the gene signatures capturing such immune variation seem to correlate with systemic lupus erythematosus (SLE) disease activity. Will the definition of these gene sets enable the development of the much needed concept of personalized medicine in SLE?

Refers to Kotliarov, Y. et al. Broad immune activation underlies shared set point signatures for vaccine responsiveness in healthy individuals and disease activity in patients with lupus. *Nat. Med.* **26**, 618–629 (2020).

Systemic lupus erythematosus (SLE) is a disease characterized by B cell activation with increased antibody synthesis and by ongoing type I interferon production, causing the prominent expression of type I interferon-regulated genes, known as the 'interferon signature'[1]. Both these features — the B cell activation and the type I interferon production — are also typical of most viral infections. Conversely, several clinical signs and symptoms that commonly occur during viral infections are also observed in patients with SLE. Thus, both patients with viral infections and those with SLE experience fever, fatigue, rash and pain in joints and muscles. Viral infections might also increase the risk of developing SLE[2]. Together, these observations suggest that similar immune processes are important in SLE and during viral infection. In a new study, Kotliarov et al.[3] investigated whether information that can be used to predict the immune response to an acute infection might also be of value in predicting the magnitude of disease flares in patients with SLE.

SLE is a heterogeneous disease that has both familial, monogenetic forms (including complement protein C1q deficiencies and *TREX1* mutations) and polygenetic forms that are associated with common single nucleotide polymorphisms, many of which have been mapped to type I interferon induction or response genes[4]. The chronic activation of the type I interferon system is present in many, but not all, patients with SLE and correlates with disease activity[5]. The ability to define patient subpopulations and understand the immune mechanisms that lead to disease flares in patients with SLE is important to avoid organ damage in these individuals and for the development of more precise therapies. In fact, this lack of targeted therapies is one of the most important unmet medical needs for patients with SLE.

In their new study, Kotliarov et al.[3] used a strategy influenced by their previous work on influenza vaccine responses to investigate similarities between vaccine responses in healthy individuals and disease flares in patients with SLE. On the basis of their previous identification of a baseline peripheral blood profile of $CD20^{+}CD38^{++}$ B cells that could be used to predict influenza vaccine responses in terms of specific antibody titres[6], they developed a blood-based transcriptional surrogate signature of ten genes that correlated with the amount of $CD20^{+}CD38^{++}$ B cells. These B cells are considered to be precursors of plasmablasts that have the capacity to secrete large amounts of antibodies. The predictive value of this ten-gene signature was investigated in several independent cohorts of individuals who had received influenza or yellow fever vaccination. In most, but not all, individuals, expression of the ten-gene signature at baseline (before vaccine administration) could predict whether that individual would have a high or low vaccine antibody response[3].

Kotliarov et al.[3] next assessed whether, at periods of no or low disease activity, expression of the same ten-gene signature in a clinically well-characterized longitudinal cohort of paediatric patients with SLE might be associated with the severity of subsequent disease flares. Previous efforts to define subgroups of paediatric patients with SLE by correlating disease activity with distinct combinations of transcriptomic signatures have revealed that a plasmablast signature is the most robust biomarker of disease activity[7]. Hypothesizing that the predictive capacity of a ten-gene signature would therefore be greatest in patients for whom disease activity correlated with a transcriptomic module enriched for plasma cells or plasmablasts, Kotliarov et al.[3] focused their analysis on this set of patients. Indeed, the ten-gene signature correlated with disease activity in this but not in other subgroups of patients. The authors also accounted for the effects of SLE treatment in the model. Overall, the results presented by Kotliarov et al.[3] suggest that the ten-gene signature evaluated during clinically quiescent periods can inform on the expected magnitude of disease flares in a defined subgroup of paediatric patients with SLE. Early onset SLE is generally more severe and has a higher degree of genetic contribution than adult onset SLE[8], so verification of the predictive value of the ten-gene signature in adult onset SLE will be important to understand its general clinical applicability.

Most cell types in the immune system can be involved in the SLE disease process, and identifying the most important cells responsible for triggering disease flares would be extremely helpful when selecting targeted therapies. However, linking the ten-gene signature back to a specific cell type proved difficult[3], despite the use of the powerful CITE-Seq method, which measures 82 cell surface immune markers in parallel with single-cell RNA sequencing of the same cells. A higher mean expression of the ten-gene signature was observed in plasmacytoid dendritic cells but this signature was not restricted to these cells and, following further bioinformatic analyses, Kotliarov et al.[3] concluded that the ten-gene signature captures the responsiveness to vaccination or prediction of SLE flares in multiple cell subsets in the peripheral blood.

Kotliarov et al.[3] also investigated the converse scenario of whether a baseline gene signature in SLE that was predictive of disease flare would also be predictive of antibody responses to vaccination in healthy individuals. Using a weighted gene co-expression network analysis to identify temporally stable transcripts across low disease activity time points in the paediatric patients with SLE, the researchers first defined a module that correlated with the disease-activity-associated change in the plasmablast score. Downstream analysis demonstrated that the module was indeed enriched for genes that had previously been associated with antibody responses in studies of influenza vaccination[3]. Overall, the data presented by Kotliarov et al.[3] emphasize that shared baseline gene signatures exist for influenza vaccination responses and for SLE disease activity.

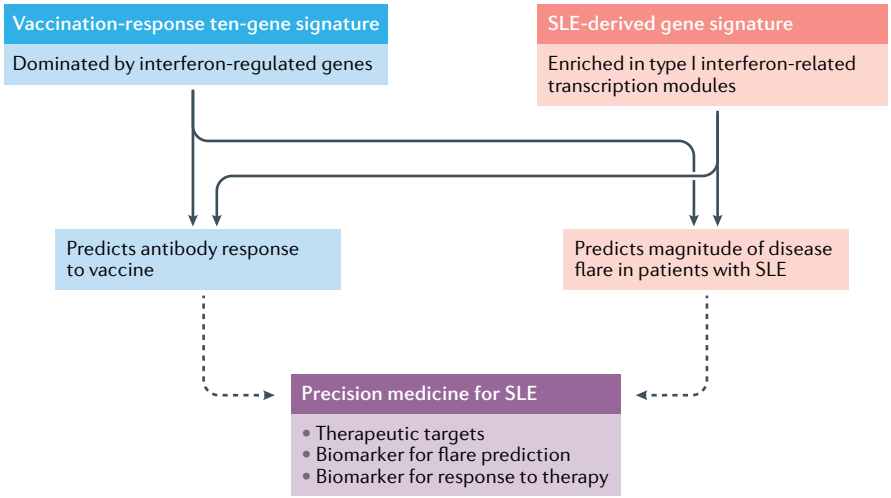

Fig. 1 | **A basal gene expression profile determines immune responses.** A ten-gene signature that predicts antibody responses to influenza vaccination also predicts flare magnitude in paediatric systemic lupus erythematosus (SLE), and conversely, a separate gene signature that predicts flare magnitude in paediatric SLE can predict antibody responses to influenza vaccination. Both gene signatures are dominated by interferon-regulated genes, but share only one gene in common. These gene signatures could be used to improve precision medicine for patients with SLE.

Perhaps not surprisingly, the SLE-derived module was enriched for type I interferon-related transcripts[3], and the type I interferon activation status of an individual is known to predict vaccine responses in patients with autoimmune diseases to both adjuvanted[9] and non-adjuvanted influenza vaccines[10]. Notably, all genes in the original ten-gene signature are interferon regulated, albeit with different expression profiles among different immune cell subsets when investigated using sites such as Interferome and The Human Protein Atlas. Several of the genes are of unknown or less extensively characterized function, leaving the question open as to whether these particular genes might be interesting therapeutic targets, or whether they will best serve as a biomarker signature for predicting flare or therapeutic responsiveness (FIG. 1). Regardless of the answers to these questions, the findings of the study by Kotliarov et al.[3] take us one step closer to the concept of personalized medicine for patients with SLE in all its forms.

The next goal will be to translate the ten-gene signature, or a refined version, into a clinically useful test that can identify patients with an increased risk for disease flares, irrespective of organ manifestations. Such a step will need the development of a reasonably priced assay and the validation of these results in longitudinal studies with large cohorts of patients. Achieving both these tasks would be a tremendous help to all clinicians faced with the challenge of caring for patients with SLE.

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SYSTEMIC LUPUS ERYTHEMATOSUS

Finding a unifying SLE expression signature in a sea of heterogeneity

Edward Wakeland and Prithvi Raj

The heterogeneity of systemic lupus erythematosus (SLE) confounds the diagnosis and treatment of this disease, and attempts at disease stratification are nascent. Researchers have identified a common set of biomarkers in patients with SLE that could identify new therapeutic targets and lead to new clinical assays to help address this issue.

Refers to Haynes, W. et al. Integrated, multicohort analysis reveals unified signature of systemic lupus erythematosus. *JCI Insight* **5**, e122312 (2020).

Systemic lupus erythematosus (SLE) is an extremely heterogeneous disease in which patients can have a spectrum of clinical manifestations coupled with a variety of dysregulations in immune system pathways. Patients with SLE can develop a diverse array of symptoms, including skin rashes, oral ulcers, glomerulonephritis, neurologic disorders, severe vasculitis and a distinct form of arthritis[1]. Hundreds of variations in genes and gene expression levels that affect the immune system have been associated with SLE, although only a handful of these features are consistently identified in most patients. The only known unifying characteristics of patients with SLE are a generalized loss of humoral immune tolerance and an aberrant activation of inflammatory effector mechanisms predominantly at the sites of immune complex deposition. However, data from a new study could help broaden our knowledge of unified features through the identification of a distinct set of biomarkers common to most patients with SLE[2].

Many studies in the past few years have focused on unravelling the extensive heterogeneity of SLE, and some progress has been made towards the stratification of patients with SLE into distinct subsets with specific disease characteristics[3–5]. Notably, a longitudinal analysis of a clinically well-characterized cohort of paediatric patients with SLE identified gene expression signatures that defined seven distinct subsets of patients with SLE[3]. Subgrouping patient cohorts in this fashion is a pre-requisite to the development of 'personalized' medicine strategies, which might ultimately improve disease management and therapy in SLE. However, the path forward to effective treatment of such a highly stratified

> “identification of a set of biomarkers common to all patients with SLE would be valuable for diagnosis”

patient population is complex. Thus, identification of a set of biomarkers common to all patients with SLE would be valuable for diagnosis and potentially could provide a unified interpretation of the dysregulated pathways that underlie the disease.

Haynes et al.[2] have discovered such a set of biomarkers in their new study published in *JCI Insight*. They identified a 93 gene expression MetaSignature and showed that this signature is a consistent feature of the expression profiles of patients with SLE that distinguishes them from healthy individuals and from patients with rheumatoid arthritis or several other related, rheumatic diseases or infectious inflammatory phenotypes. The SLE MetaSignature was identified by an integrated, multi-cohort meta-analysis of 7,471 transcriptomic profiles from 40 independent datasets of gene expression profiles. The technical and biological variability between the many studies combined in this meta-analysis is extensive, and the studies included data generated with several distinct technologies and assay platforms. The analysis was performed with MetaIntegrator, which is an impressive analytical program developed for analysing large datasets from heterogeneous sources. Although the effect size threshold used to define the 93 biomarkers in the SLE MetaSignature was relatively small (effect size ≥1.0 and false discovery ratio ≤5%), the result was statistically robust, and the signature was found to be uniquely expressed in SLE in multiple tissues (such as in the blood, skin and kidneys). Thus, the findings from Haynes et al.[2] provide a statistically robust identification of a core set of genes that are virtually universally dysregulated in patients with SLE.

The researchers further validated this signature in a prospective analysis by comparing cohorts of paediatric patients with SLE ($n = 43$), healthy individuals ($n = 12$) and patients with juvenile idiopathic arthritis (JIA; $n = 10$) using a 33 gene subset of the SLE MetaSignature genes. This prospective study statistically distinguished these three cohorts and validated the MetaSignature using an independent assay. However, the MetaScore (a quantitative value of the MetaSignature) varied considerably among the individuals in each cohort, leading to notable overlaps between the three groups. Thus, although the analysis statistically distinguished patients

> this study solidly establishes the potential of this signature for the development of clinical tests

with SLE from the other cohorts, some of the patients had Metascores that were indistinguishable from some healthy individuals or patients with JIA. Nonetheless, the ability of the MetaSignature to reproducibly distinguish patients with SLE from other patient groups and healthy individuals supports the feasibility of developing an assay of MetaSignature genes as a component of SLE diagnosis.

The identities of most of the genes included in the MetaSignature are not surprising, in that all but 14 of the 93 biomarkers were associated with either interferon (IFN) stimulation or expansions of the neutrophil or granulocyte lineages; both of these elements are known component phenotypes in SLE pathogenesis[6,7]. Interestingly, although the IFN-related genes are clearly dysregulated in multiple tissues, the neutrophil-associated component of the SLE MetaSignature seems to reflect an expansion of neutrophils in the samples from patients with SLE, rather than a dysregulated expression of these genes in neutrophils or immature granulocytes. Overall, this expression analysis supported an association of low-density granulocytes and NETosis with SLE but did not identify gene expression abnormalities in the neutrophils of patients with SLE.

The remaining 14 novel genes, which the authors refer to as the "underappreciated, non-IFN, non-neutrophil SLE MetaSignature genes", are an eclectic collection of genes associated with a variety of molecular pathways. Notably, several members of the metallothionein family are included, suggesting that oxidative stress or environmental stimuli that induce oxidative stress could be a consistent feature of SLE pathology. Alternatively, metallothionein genes are known to be upregulated by various cytokines[8], and thus their association with SLE might reflect the well-established increase in cytokine levels in patients with SLE[9]. The remaining 11 genes in the 'underappreciated' component of the MetaSignature genes included five genes of unknown function, four immune system genes (*KLRB1, GPR183, CD1C* and *ELANE*) and individual genes related to vitamin B_{12} metabolism (*TCN2*) and epidermal cellular integrity (*DSC1*). What role, if any, these genes have in SLE pathogenesis remains to be determined; however, their identification in this unified signature strongly supports the necessity of increased investigations into the functions of all of the 'underappreciated' SLE-associated genes. If future analyses identify a function for any of these genes in SLE pathogenesis, then they could become important new therapeutic targets.

The MetaSignature might be of value in medical practice not only for its use as a diagnostic tool, but also through its use as a quantitative measurement of disease severity. Five of the 40 studies analysed included SLEDAI scores and other clinical phenotypes. Haynes et al.[2] showed that the SLE MetaScore was positively correlated with the SLEDAI score and inversely correlated with levels of complement proteins C3 and C4. Although the statistical associations between MetaScore and these clinical measurements were quite high ($P = 2.47e-26$ and $P = 1.79e-20$), the correlation coefficients were low ($R = 0.25$ and $R = 0.3$), and the values for individual patients were quite scattered, despite the statistical strength. Thus, although the MetaScore might be of use as an empirical measurement of disease activity in clinical trials, its utility as a clinical measurement would be limited when evaluating individual patients.

In summary, this well-designed study provides important new data and identifies an expression signature that is specifically associated with SLE, based on a meta-analysis of over 7,400 transcription datasets produced by 40 different gene expression studies. Overall, this study solidly establishes the potential of this signature for the development of clinical tests that might improve SLE diagnosis and the assessment of disease severity in clinical trials. Thus, it seems that some uniformity can be found amidst the sea of heterogeneity that confounds most studies of SLE.

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CLINICAL TRIALS

Minimizing efficacy differences between phase II and III RCTs

Vibeke Strand

Results from phase II randomized controlled trials (RCTs) determine whether promising therapeutics will progress to phase III. According to detailed analyses in rheumatoid arthritis and psoriatic arthritis, phase II efficacy data systematically overestimate subsequent phase III results, raising the question of how this discrepancy might be addressed in future RCTs.

Refers to Kerschbaumer, A. et al. Efficacy outcomes in phase 2 and phase 3 randomized controlled trials in rheumatology. *Nat. Med.* https://doi.org/10.1038/s41591-020-0833-4 (2020).

The importance of phase II randomized controlled trials (RCTs) cannot be overestimated, as their results determine whether clinical development of a promising therapeutic proceeds to phase III, as well as determining the eligibility criteria and outcome measures that will be utilized. However, although efficacy data from phase II and phase III RCTs can be expected to differ for several reasons (BOX 1), whether these differences result in meaningful overestimations of efficacy of the therapeutic in phase III RCTs and how these discrepancies might be addressed remain uncertain. In a new publication, Kerschbaumer et al.[1] compared the results of multicentre, multinational phase II and phase III RCTs in rheumatoid arthritis (RA) and psoriatic arthritis (PsA) to assess whether and why phase II trials might systematically overestimate results reported in subsequent phase III RCTs.

Using mixed-model logistic regression analysis, including an exploration of potential determinants of efficacy overestimation, Kerschbaumer et al.[1] determined that in RA, phase II trials systematically overestimated subsequent phase III results. Data in PsA trials were similar to those in RA, but not statistically significant[1]. The primary end points across the trials were the ACR20, ACR50 and ACR70 response criteria (denoting ≥20%, ≥50% or ≥70% improvements in the ACR response criteria, respectively), which made analysis of paired trials conducted between 1998 and 2018 possible. Each RCT included in the analysis was evaluated in accordance with Grading of Recommendations, Assessment, Development and Evaluations (GRADE) scoring and Cochrane Collaborations guidelines, revealing a low risk of bias.

In their detailed analyses[1], no interactions were evident between study phase, year of study completion or publication, duration of placebo exposure, number of study sites or sample sizes, and insufficient details were included in publications to explore the effects of geographic recruitment differences. In particular, incorporating the mean baseline joint counts significantly improved the multivariate F values, indicating better statistical capacity to explain differences in response rates. After Bonferroni corrections for multiple testing, only inclusion criteria for the minimum required swollen joint count (SJC) and tender joint count (TJC), as well as the type of joint count (28-joint versus the 66/68-joint count, which requires evaluation of 66 joints for swelling and 68 joints for tenderness) were significant predictors of efficacy differences on the basis of *P* values for interaction terms between the respective determinant and study phase. Reviewing the supplementary figures from this study[1] illustrates the importance of the 66/68-joint count, both in terms of defined inclusion criteria and actual reported baseline SJC and TJC. These results led the authors to

> “utilizing the 66/68-joint count … would reduce phase II and III efficacy discrepancies”

conclude that utilizing the 66/68-joint count and requiring eight swollen and eight tender joints would reduce phase II and III efficacy discrepancies[1].

In both RA and PsA, the SJC and TJC are important components of disease activity criteria, and a higher joint count, especially the SJC, reflects a higher degree of inflammation[2,3]. Patients with more active disease are more likely to respond to effective investigational agents than patients with less active disease and generally report lower placebo responses[4]. In their analyses, Kerschbaumer et al.[1] demonstrated that the 66/68-joint count was preferable to the 28-joint count, as almost all RCTs that utilized the 28-joint count (which omits joints of the ankles and feet) demonstrated larger efficacy differences between phase II and III RCTs. Use of the 28-joint count has been controversial, and a 2018 workshop by Group for the Assessment of Psoriasis and PsA (GRAPPA) and Outcome Measures in Rheumatology (OMERACT) concluded that it was not appropriate for assessment of patients with PsA[5] (which, even as a pauciarticular arthritis, frequently affects the feet). Furthermore, requiring eight swollen and eight tender joints of the 66/68 joints assessed clearly reduced efficacy differences between phase II and phase III RCTs in RA and PsA[1].

So do the results of the study by Kerschbaumer et al.[1] mean we should no longer trust results from phase II RCTs? The authors analysed data from 39 trials that included 10,860 patients with RA (testing 17 agents across 28 regimens) and 12 trials that included 1,303 patients with PsA (testing 5 agents across 5 regimens). Inclusion of RCTs in PsA was limited by the lack of phase II trials, which are typically not conducted when an agent is investigated for a second clinical indication.

Box 1 | Why do efficacy results differ between phase II and III RCTs?

Efficacy results can differ between phase II and phase III randomized controlled trials (RCTs) for several reasons, some of which are listed below.

- Expectation bias after positive phase II data can change perceptions about the therapy and therefore who might be enrolled in phase III trials.
- Differences in enrolment criteria can occur for many reasons, including:
  - the requirement for increased sample sizes for large phase III RCTs
  - multinational enrolment in phase III RCTs
  - avoidance of overly restrictive eligibility criteria in phase III RCTs that could negatively affect labelling of the product at approval
- Regression to the mean: a phenomenon that arises if a random variable is extreme on its first measurement but closer to the population mean or average on its second measurement.

> we should continue to trust results from well-designed and adequately powered phase II RCTs

Ultimately, three products were not approved in RA: two owing to safety concerns (fostamatinib and sirukumab) and one to a lack of sufficient efficacy (tabalumab), indicating that only one product failed in a phase III trial after a successful phase II RCT. Of the products with successful phase II results that were not carried forward into phase III RCTs, reasons other than efficacy concerns led to discontinuation of clinical development with clazikizumab, mavrilimumab and ABT-122, which were judged insufficiently beneficial in the specific clinical indications to be competitive in today's market. Decernotinib was discontinued owing to an association with drug–drug interactions with statins, and the development of brodalumab in PsA was discontinued owing to suicidality. Olokizumab is still in clinical development[6], but the phase III programme had not yet been fully initiated when the study by Kerschbaumer et al.[1] was conducted. Thus, phase III trials were not conducted for six out of seven products for reasons other than efficacy as demonstrated in phase II RCTs, and only one failed to show efficacy in phase III following a positive phase II RCT.

Therefore, we should continue to trust results from well-designed and adequately powered phase II RCTs. Nonetheless, information that will enable differences in efficacy between phase II and III RCTs to be minimized should be welcomed, and Kerschbaumer et al.[1] are to be congratulated on their thorough analyses[1]. However, implementing their suggestion for a minimum of eight swollen and eight tender joints for enrolment could be a tall order, as many patients are treatment experienced yet have not achieved either low disease activity or remission[7–9]. Such patients are most common in North America and Western Europe and represent a persistent unmet need for new therapies. Adjusting enrolment criteria to instead require six swollen and eight tender joints of the 66/68 joints assessed might represent a compromise that will include patients with sufficiently active disease and still facilitate enrolment within a pragmatic time frame.

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Competing interests

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Atherosclerotic cardiovascular disease prevention in rheumatoid arthritis

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Abstract | Patients with rheumatoid arthritis (RA) are at high risk of developing cardiovascular disease (CVD). Inflammation has a pivotal role in the pathogenesis of CVD. RA is an inflammatory joint disease and, compared with the general population, patients with RA have approximately double the risk of atherosclerotic CVD, stroke, heart failure and atrial fibrillation. Although this high risk of CVD has been known for decades, patients with RA receive poorer primary and secondary CVD preventive care than other high-risk patients, and an unmet need exists for improved CVD preventive measures for patients with RA. This Review summarizes the evidence for atherosclerotic CVD in patients with RA and provides a contemporary analysis of what is known and what needs to be further clarified about recommendations for CVD prevention in patients with RA compared with the general population. The management of traditional CVD risk factors, including blood pressure, lipids, diabetes mellitus and lifestyle-related risk factors, as well as the effects of inflammation and the use of antirheumatic medication on CVD risk and risk management in patients with RA are discussed. The main aim is to provide a roadmap of atherosclerotic CVD risk management and prevention for patients with RA.

Diastolic dysfunction
A stiffening of the ventricles that restricts the ability of the heart to fill with blood between beats.

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Rheumatoid arthritis (RA) is a chronic systemic inflammatory disease of unknown aetiology that affects 0.2–1% of the adult population worldwide[1–4]. RA is characterized by inflammation in the joints that causes swelling, pain and stiffness and can often also involve the formation of autoantibodies such as rheumatoid factor (RF) or anti-citrullinated protein antibodies. The persistent inflammation leads to joint damage in all affected joints, as well as joint deformities, particularly in the hands and feet[5,6]. However, as a systemic autoimmune inflammatory disease, RA can affect any of the body systems in addition to the joints.

Cardiovascular disease (CVD) is a broad term that includes atherosclerosis, heart failure, cerebrovascular disease, peripheral vascular disease and several other cardiac abnormalities (including arrhythmias, valvular disease, pericarditis, myocarditis, cardiomyopathy and endocarditis). Inflammation, which is a central part of the immunological pathophysiology of RA, also has a role in the development of CVD, and patients with RA have an increased risk of most types of CVD[7]. Inflammation is also involved in the development of atherosclerosis, which results in ischaemic heart disease, myocardial injury leading to fibrosis causing diastolic dysfunction and heart failure with preserved ejection fraction[8].

Risk factors for CVD in the general population include modifiable lifestyle factors (such as smoking, BMI, degree of physical activity and a high-fat and/or high-sugar diet), as well as hypertension, hyperlipidaemia and diabetes mellitus. Increasing age and male sex are also associated with an increased risk of CVD. In addition to these traditional CVD risk factors, many other factors are associated with an increased risk of CVD, both in the general population and in patients with RA. Non-traditional risk factors, including characteristics of RA such as extensive erosive joint disease and extra-articular disease, might have a role in the development of CVD among patients with RA. Therapies used to treat RA might also affect the risk of CVD, either positively or negatively[7].

In this Review, we provide an overview of atherosclerotic CVD risk in patients with RA that includes CVD outcomes, CVD risk evaluation and CVD risk management, and we give a contemporary analysis of what is known and what needs to be further clarified. European guidelines for CVD prevention for the general population[9] are presented in greater detail than guidelines from the USA[10], but major differences between the sets of guidelines are discussed. At present, there are no RA-specific recommendations for CVD risk prediction from the USA; however, important points from the 2017

Key points

- Patients with rheumatoid arthritis (RA) have an increased risk of cardiovascular disease (CVD) compared with the general population.
- The improvement of CVD risk prevention in patients with RA is an unmet need.
- CVD risk calculators developed for use in the general population inaccurately predict CVD in patients with RA, but the addition of RA-specific risk factors does not improve CVD risk prediction
- The use of ultrasonography of the carotid arteries improves CVD risk classification in patients with RA by identifying atherosclerotic plaques.
- CVD risk prevention in patients with RA closely follows the recommendations for the general population; however, clinicians should be aware of some specific drug–drug interactions in this patient population.
- Inflammation and antirheumatic medication use in patients with RA does not affect the doses of statins or antihypertensive medications required for attainment of recommended lipid or blood pressure goals.

EULAR recommendations for CVD risk management in patients with RA[11] are presented.

CVD outcomes in RA

The chronic systemic inflammation that characterizes RA is also an important factor in atherosclerosis; therefore, it is not surprising that patients with RA have an increased risk of CVD. In general, CVD mortality is increased by ~50% in patients with RA compared with the general population[12]. In patients with RA, ~50% of deaths are attributable to CVD-related causes, and CVD events seem to occur at younger ages in patients with seropositive RA than in the general population[13]. Unlike the risk of a non-fatal CVD event, the risk of CVD mortality might not increase until 7–10 years after the onset of RA symptoms[14]. Reports from the past 5 years have shown that the global trend of reduced CVD mortality in the general population is also reflected in patients with RA, both in Europe and the USA[15,16]. Unfortunately, despite these improvements, the heightened risk of atherosclerotic CVD in patients with RA compared with the general population seems to persist[17].

Several types of CVD-related morbidity are increased in patients with RA, including myocardial infarction, stroke, atrial fibrillation and heart failure, which are briefly discussed below. However, in the rest of the Review we focus exclusively on the prevention of atherosclerotic CVD in RA.

Myocardial infarction

The onset of the increased risk of CVD in patients with RA seems to precede the onset of RA symptoms, as patients with RA had a threefold higher likelihood of hospitalization for myocardial infarction during the 2-year period before RA diagnosis than age-matched and sex-matched individuals without RA[18]. However, this increased risk of myocardial infarction before the onset of RA symptoms was not replicated in a study of two large Swedish cohorts[17]. Patients with RA are also less likely to report symptoms of angina and more likely to experience unrecognized (or 'silent') myocardial infarctions than the general population[18]. The increased risk of myocardial infarction after the onset of RA is undisputed, as many studies have consistently reported a 1.5-fold to 2.0-fold increased risk of myocardial infarction in patients with RA[19]. Furthermore, whereas short-term outcomes (within 6 months) after myocardial infarction were similar in individuals with and without RA, patients with RA who had experienced a myocardial infarction had a 1.5-fold increased risk of a long-term outcome of recurrent ischaemia and mortality compared with individuals without RA[20].

Stroke and atrial fibrillation

Large population-based studies have revealed that the risk of atrial fibrillation is substantially higher in patients with RA than in the general population with a pooled risk ratio of 1.29 (95% CI, 1.05–1.59)[21]. Furthermore, the risk of any type of stroke (haemorrhagic or ischaemic) is higher in patients with RA than in the general population with a risk ratio of 1.91 (95% CI, 1.73–2.12)[22–24]. Inflammation is associated with both atrial fibrillation and ischaemic stroke[15], and patients with RA have an increased risk of atherosclerotic CVD with a background of traditional risk factors and inflammation, in addition to the development of valvular disease, which are known risk factors for atrial fibrillation and ischaemic stroke. However, it is not clear whether the increased risk of atrial fibrillation and stroke is a result of their primary inflammatory state or of the overall risk of CVD in these patients. Short-term outcomes following stroke (such as in-hospital mortality, pneumonia and mechanical ventilator use) are not different for patients with or without RA[15]; however, the risk of recurrent stroke is 40% higher in patients with RA than in those without RA, particularly among smokers[25].

Heart failure

Patients with RA have a nearly twofold increased risk of developing heart failure compared with the general population[26,27]. These reports from the USA in the mid-2000s of increased heart failure risk in patients with RA were confirmed in large studies from 2017 and 2018 in Scandinavian cohorts[28,29]. This increased risk cannot be explained by a higher prevalence of ischaemic heart disease in this population, but is associated with RA disease activity[26,30]. Patients with RF-positive RA had a 2.5-fold increased risk of heart failure compared with individuals without RA, whereas patients with RF-negative RA did not have a statistically significant increased risk of heart failure[27]. A large study of two Swedish cohorts confirmed the lack of an increased risk of heart failure among RF-negative patients with RA and found only a 36% increased risk of heart failure among RF-positive patients with RA[28]. Furthermore, heart failure in patients with RA is characterized by a preserved ejection fraction, and patients with heart failure who have RA experience poorer outcomes than patients with heart failure who do not have RA[31]. Left ventricular diastolic dysfunction is also more common in patients with RA than in individuals without RA and is associated with RA disease activity[32].

CVD risk evaluation

The identification and management of risk factors is pivotal for the ultimate goal of medicine — the prevention of disease. The estimation of total CVD risk is instrumental

Heart failure with preserved ejection fraction
A form of heart failure in which the ejection fraction — the percentage of the volume of blood ejected from the left ventricle with each heartbeat divided by the volume of blood when the left ventricle is maximally filled — is normal, defined as greater than 50%.

Atrial fibrillation
An irregular, rapid heart rate that occurs when the atria beat out of rhythm with the ventricles, which can cause symptoms including heart palpitations, fatigue and shortness of breath.

for the detection of high-risk individuals who will benefit from CVD preventive efforts as, although RA is an independent risk factor for CVD, the presence of RA alone is not sufficient to warrant cardio-preventive medication[11,33]. In the following sections, we discuss CVD risk calculators, the use of imaging to assess CVD risk in patients with RA and the implementation of CVD evaluation strategies in the clinic.

CVD risk calculators

At present, more than 360 unique CVD prediction models have been developed[34]. The CVD risk factors included in the vast majority of risk calculators overlap, as most incorporate blood pressure, lipid levels and smoking status in addition to age and sex. The majority of CVD risk calculators focus on predicting atherosclerotic CVD, although some risk calculators use broader outcomes that include other forms of CVD. For example, the Systematic Coronary Risk Evaluation (SCORE) calculator[35] includes fatal myocardial infarction, and the most recent Framingham Risk Score (FRS) includes cerebrovascular and peripheral vascular disease events and heart failure[36].

The CVD outcomes and treatment thresholds used in various risk calculators[37] are presented in TABLE 1. Briefly, the SCORE calculator was derived and validated in 2003 from 12 European cohorts and is used in European countries to estimate the 10-year risk of CVD mortality[35]. A SCORE calculator for older people has also been derived and validated[38], which provides improved accuracy in CVD risk estimation for individuals aged 65 years and over, and might reduce excessive use of preventive drugs in this population. The FRS was derived and internally validated in a cohort in the USA for the estimation of the 10-year risk of CVD and has been updated several times, most recently in 2008 (REF.[36]). However, the FRS was replaced by the American College of Cardiology and American Heart Association Pooled Cohort Equation risk calculator (ACC/AHA PCE) in the 2013 ACC/AHA guidelines on CVD prevention[39]. In contrast to the previous FRS models, which used data from the predominantly white Framingham cohort, the ACC/AHA PCE used data from a cohort of individuals from different ethnic backgrounds. The Reynolds Risk Score was also developed in the USA from prospective cohorts of non-diabetic men and women and gained attention as a candidate CVD risk calculator for patients with RA as it incorporates high-sensitivity C-reactive protein (CRP) concentrations as a covariate[40,41]. In the UK, data from the QRESEARCH database covering numerous primary care practices contributed to the development of the first QRISK calculator[42,43] in 2007, which was later replaced by QRISK2 (REFS[42,43]) in 2008 and by QRISK3 (REF.[44]) in 2017. In the QRISK2 and QRISK3 calculators, RA is included as an independent risk factor and is designated a weight of 1.4.

Efforts have been made to develop RA-specific CVD risk calculators. Using data from ten different countries across different continents, A Transatlantic Cardiovascular Risk Consortium for RA (ATACC-RA) derived two candidate CVD risk algorithms[45]. Similarly, the Expanded Risk Score in RA (ERS-RA) was developed using data from the CORRONA cohort[46]. Comparable to the ATACC-RA models, the ERS-RA model included traditional CVD risk factors as well as RA-specific disease characteristics. The predictive ability of these RA-specific CVD risk calculators has been evaluated in a few RA cohorts using hard CVD end points; however, the results from these reports have been mostly underwhelming[45,47,48].

The CVD risk calculators developed for the general population, such as the FRS and the SCORE calculator, yield gross underestimations of CVD risk in most risk classes when used to assess CVD risk in patients with RA[49–51]. This underestimation might be caused by chronic high-grade inflammation being an independent

Table 1 | **Cardiovascular disease outcomes and treatment thresholds of various risk calculators**

Risk calculator	Target population	CVD outcome	Applicable age range (years)	Treatment threshold (%)
Framingham risk score (Adult Treatment Panel III)	USA	Coronary heart disease including myocardial infarction	30–74	10
Framingham risk score for general CVD	USA	CVD events (fatal and non-fatal) including acute coronary syndrome (myocardial infarction and unstable angina pectoris), chronic ischaemic heart disease (stable angina pectoris), coronary revascularization (percutaneous coronary intervention and coronary artery bypass graft surgery), coronary death, other cardiovascular death, cerebrovascular events (ischaemic cerebrovascular accident and transient ischaemic attack), peripheral vascular events (non-coronary revascularization procedures and peripheral artery disease) and heart failure	30–74	20
ACC/AHA pooled cohort equation	USA	Atherosclerotic CVD events (defined as first occurrence of non-fatal myocardial infarction, coronary heart disease death, or fatal or non-fatal stroke)	40–79	7.5
Reynolds Risk Score	USA	Myocardial infarction, ischaemic stroke, coronary revascularization and cardiovascular death	50+	10
QRISK2	UK	Coronary heart disease, stroke and transient ischaemic attack	35–74	10
SCORE	EU	Fatal CVD events	40–79	5

ACC/AHA, American College of Cardiology/American Heart Association; CVD, cardiovascular disease; EU, European Union; SCORE, Systematic Coronary Risk Evaluation. TABLE 1 is reprinted from REF.[37], Springer Nature.

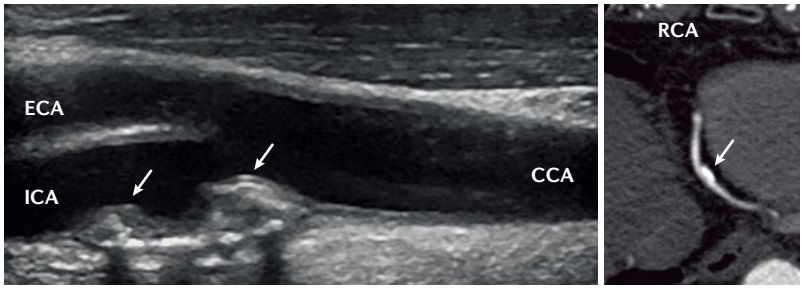

Fig. 1 | **Imaging modalities in cardiovascular disease risk evaluation.** The image on the left-hand side is an ultrasonograph of the carotid artery in the longitudinal view. On the right-hand side is a coronary CT angiogram image of the right coronary artery (RCA). The arrows point to the atherosclerotic plaques. CCA, common carotid artery; ECA, external carotid artery; ICA, internal carotid artery.

predictor of future CVD[52]. Hence, it has been advocated that multiplication factors should be used when estimating CVD risk in patients with RA using calculators developed for the general population. However, attempts to adapt CVD risk algorithms established for the general population with a multiplication factor, such as in QRISK, or adjustment of other risk factors has not yet yielded precise CVD risk estimates for patients with RA[51,53]. Furthermore, the Reynolds Risk Score (which incorporates high-sensitivity CRP concentrations) also failed to improve CVD risk prediction in a longitudinal cohort of patients with RA[49]. One validation study revealed that RA-specific CVD risk calculators, including the two ATACC-RA models and the ERS-RA calculator, were not superior in predicting CVD events to CVD risk algorithms that were developed for the general population[47]. However, two additional validation studies were subsequently published that reported different predictive ability of the ERS-RA model. One study[48] supported the conclusion that the ERS-RA calculator was not superior to other CVD risk algorithms, whereas the other study[54] reported a good discriminatory capability of the ERS-RA, but did not compare it to the estimates of other CVD risk calculators.

As discussed in detail elsewhere[45], the development of RA-specific CVD risk models presents several methodological challenges. One particular obstacle is the difficulty of estimating the cumulative burden of high-grade inflammation, which is associated with increased CVD risk. EULAR have acknowledged the limitations of the current CVD risk calculators and have stated that future research should aim to improve previously developed risk algorithms by adjusting them for application in RA[11]. Awaiting superior risk calculators, EULAR further recommended that, in the absence of national guidelines, CVD risk estimation should be performed using a 1.5 multiplication factor to the risk estimate by the SCORE calculator for patients with RA[11]. However, this multiplication factor needs to be validated for use in the USA. If a CVD risk calculator is used, the clinician needs to bear in mind the limitations of the calculator, especially those related to overprediction when used in typical clinical trial populations and underprediction when used in high-risk populations such as patients with RA. Owing to large geographical differences in CVD risk[4], clinicians are recommended to use a CVD risk algorithm that was developed for their specific population, such as the ACC/AHA PCE in the USA and the SCORE calculator in Europe.

Imaging in CVD risk assessment

Using non-invasive imaging techniques it is possible to detect the presence and extent of atherosclerotic disease. The identification of atherosclerosis can support the initiation of CVD preventive treatment.

Ultrasonography of carotid arteries. An association exists between the severity of atherosclerosis in one artery and the presence of atherosclerosis in other arteries[55]. A simple, non-invasive method to evaluate atherosclerotic burden is therefore ultrasonography of the carotid arteries, which can be used to measure the carotid intima–media thickness (CIMT) and to detect the presence of carotid plaques (FIG. 1). However, the quality of ultrasonography is operator dependent. Interestingly, a novel method of analysing the ultrasonography images has been proposed in which machine learning and deep learning are used to perform tissue characterization[56], which could simplify the ultrasonography procedure in the future. A CIMT >0.9 mm is considered to be pathologically enlarged; however, screening of CIMT by carotid ultrasonography is not currently recommended for use in CVD risk evaluation for the general population or for patients with RA[9,11]. The evidence for this recommendation was a meta-analysis, the authors of which concluded there was no value in adding CIMT measurement to the FRS in the prediction of CVD events[57]. The pathophysiological basis for the lack of added value of CIMT in CVD risk prediction might be that an increased CIMT could both be associated with atherosclerosis formation and with smooth muscle cell hypertrophy, mainly caused by hypertension[58].

Carotid plaques are considered to be a coronary heart disease risk equivalent in the current European Society of Cardiology/European Atherosclerosis Society (ESC/EAS) guidelines for the management of dyslipidaemias[9], meaning that CVD preventive therapy is indicated on similar terms for both conditions. An atherosclerotic plaque is defined by a CIMT ≥1.5 mm, or at least a doubling of the surrounding CIMT[59]. Owing to the high pre-test probability for the detection of carotid artery plaques by ultrasonography in patients with RA and the indication for statin treatment should a carotid plaque be present, EULAR recommends that screening for asymptomatic atherosclerotic plaques by carotid ultrasonography should be considered as part of CVD risk evaluation for all patients with RA[11]. In fact, carotid ultrasonography of 335 patients with inflammatory joint diseases contributed to their reclassification to a more appropriate CVD risk group in ~35% of patients with an estimated low to moderate risk and in ~60% of patients with an estimated high CVD risk[60]. Reportedly, most patients with inflammatory joint diseases who have carotid atherosclerotic plaques have coronary artery disease (CAD)[61], and the presence of carotid atherosclerotic plaques is strongly linked to future acute coronary syndrome and CVD mortality in patients

Coronary angiography
Radiography performed with contrast agent in the coronary arteries.

Coronary artery stenosis
Narrowing of the arteries that supply blood to the heart muscle.

with RA[62,63]. If a plaque is present, then lipid-lowering therapy is indicated with a low-density lipoprotein (LDL) cholesterol goal of <1.4 mmol/l[9]. Indeed, intensive lipid-lowering treatment with rosuvastatin induced carotid plaque regression in an intervention study that included 86 patients with RA[64].

Coronary CT angiography and calcium scoring. The association between chest pain and CAD is low in patients with inflammatory joint diseases[65]. Selective coronary angiography is considered the gold standard for assessment of CAD; however, non-invasive coronary CT angiography (CCTA) has become an established method to exclude clinically relevant CAD in patients with low to intermediate risk of CVD[66]. CCTA also reveals the localization and morphology of atherosclerotic plaques[67] (FIG. 1). In a study that involved 150 patients with RA and 150 age-matched and sex-matched individuals without systemic autoimmune diseases, the patients with RA had the highest prevalence of and most severe coronary artery plaques[68]. Those patients with RA who had the highest disease activity (as measured by the 28-joint disease activity score with CRP) had more non-calcified and mixed coronary plaques, indicating a higher risk of developing symptomatic CAD, than patients with less active disease[68].

The coronary artery calcium (CAC) score is used to indicate the total plaque burden in the coronary arteries and is measured by CCTA. A CAC score measured by the Agatston method of ≥300 is considered unfavourable[33]. The CCTA method has a high negative predictive value; hence, if the Agatston score is zero, a significant coronary artery stenosis is very unlikely and can, for all practical purposes, be excluded[69]. The Agatston score is an independent predictor of CAD[70] and could improve CVD risk evaluation if the result is taken into consideration in addition to conventional risk factors[71]. Interestingly, the results of the US Multi-Ethnic Study of Atherosclerosis showed that the CAC score improved the prediction of CVD more than carotid ultrasonography measurements[72]. Comparable results were found in another population in the USA, for which both the CAC score and the carotid plaque burden improved the specificity of CVD risk prediction and the net reclassification index, and the CAC score was superior to carotid plaque burden for CVD risk prediction[73].

Notably, CAC increase is accelerated by the use of statins; however, despite a statin-induced increase in CAC, CVD events did not occur more frequently in a study of individuals with suspected CAD[71]. Concerns also exist regarding costs and radiation exposure for this method. Furthermore, there is no clear recommendation for the use of CCTA and CAC scoring to decide an indication for statin treatment in European guidelines[9,33] or in the EULAR recommendations for CVD risk management in patients with RA[11]. However, the ESC suggests that the use of CCTA with CAC scoring using the Agatston method can be considered for individuals around the decisional thresholds at 5% and 10% estimated 10-year risk of future CVD in relation to an indication for lipid-lowering therapy[33]. The guidelines from the USA on blood cholesterol differ substantially from the European guidelines on this point and support the use of a CAC score ≥100 measured by the Agatston method as an indication for lipid-lowering therapy for those individuals with a borderline or intermediate risk for whom a decision is uncertain[10]. Furthermore, a CAC score can be used in the assessment of patients with inflammatory diseases if the patient or the clinician is uncertain about the need for statin therapy or if the patient has previously had adverse effects from this treatment[10]. Overall, the guidelines from the USA suggest that non-smoking individuals without CAC would have a very low risk of an atherosclerotic CVD event in the next decade, whereas a CAC score of greater than the 75th percentile for age and sex or ≥100 measured by the Agatston method would support the initiation of statin therapy[10].

CVD risk assessment in the clinic

Although the increased risk of CVD in patients with RA has been known for decades, this high-risk patient population has received limited attention from the medical community, and a clear need exists for the implementation of strategies for CVD risk factor gathering and recording for patients with RA[74,75]. Several reasons exist for the poor handling of CVD risk in patients with RA. One reason is that the awareness of the increased CVD risk in this patient population is low among health personnel[76]. Another important reason might be that, in most countries, CVD risk evaluation is performed by primary care physicians rather than by rheumatologists. Patients with RA do not see their primary care physicians as often as other individuals, possibly because they attend consultations at the rheumatology department[77]. This discrepancy is an important challenge to overcome. Interestingly, CVD risk factor recording and evaluation can and has been performed in rheumatology outpatient clinics (FIG. 2). Age, sex and smoking status are recorded routinely in rheumatology outpatient clinics; therefore, only blood pressure and lipid measurements need to be added to the recording in the clinic to enable a CVD risk evaluation to be performed using the SCORE calculator[9]. If the SCORE calculator were implemented as part of the electronic patient record system, the 10-year risk of CVD could be automatically calculated and would be available to the rheumatologist when the patient attended a consultation. This way of performing a CVD risk evaluation was feasible when implemented as a quality improvement project in seven rheumatology outpatient clinics in Norway over a 1.5-year period[78]. The project resulted in an increase in CVD risk prediction in patients with inflammatory joint diseases from 0% to 41% in these rheumatology outpatient clinics[78]. In this way, patients with RA could easily have their CVD risk evaluation carried out in rheumatology outpatient clinics without major relocation of resources. High-risk patients could then be referred to a primary care physician or a cardiologist for the initiation of CVD preventive measures (FIG. 2).

Overall, CVD risk evaluation in patients with RA should be performed in accordance with region-specific guidelines for the general population, as no RA-specific CVD risk algorithms are available to distinguish between patients with high and low CVD risk. However, owing to

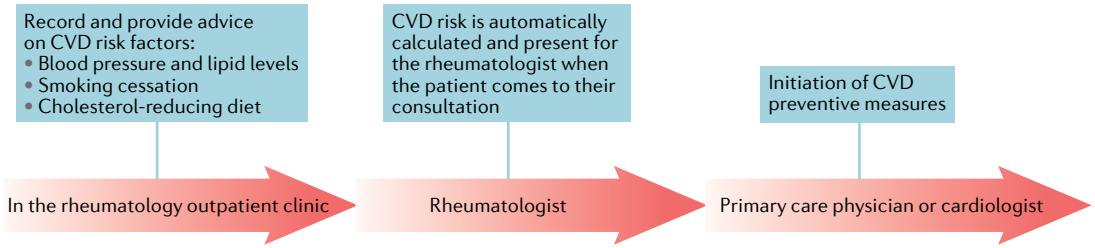

Fig. 2 | **Model for cardiovascular disease risk evaluation in a rheumatology clinic.** Cardiovascular disease (CVD) risk factor recording in rheumatology clinics can be accomplished in three steps. First, allied health-care providers add lipid levels (including total cholesterol, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol and triglycerides) to the routine rheumatology laboratory tests. Blood pressure measurements are taken at the time of the clinical joint examination, and all values are entered into the electronic patient journal. Next, the risk of experiencing a fatal CVD event in the coming 10 years is automatically estimated by the risk calculator (the Systematic Coronary Risk Evaluation algorithm). The rheumatologist evaluates the risk estimate, and if the patient has low or moderate CVD risk (<5%) then no further measures are taken, but a new CVD risk assessment should be performed after 5 years. Conversely, if the patient has a high or very high CVD risk (≥5%) then a referral note can be forwarded to the patient's primary care physician or cardiologist for the initiation of CVD preventive measures.

the high pre-test probability for the detection of carotid artery plaques by ultrasonography in patients with RA, the EULAR recommendations suggest screening for asymptomatic atherosclerosis using this method as part of CVD risk evaluation in all patients with RA[11]. The supplemental value of adding CIMT and/or carotid plaque measurements to CVD risk prediction models has not yet been proved for the general population or for patients with RA. However, according to the 2019 ESC/EAS guidelines for the management of dyslipidaemias[9], independently of their predicted risk of CVD, patients with carotid plaques identified by ultrasonography should be considered to be very high risk individuals for whom lipid-lowering treatment is recommended. Finally, it should be noted that outcome-based comparisons of CAC scores versus assessment of carotid or femoral plaque burden by ultrasonography for CVD risk reclassification in individuals at moderate or high risk is described as a gap in evidence in the current ESC/EAS guidelines[9]. Thus, more evidence is needed to conclude whether CCTA with a CAC score or the use of ultrasonography to detect carotid atherosclerosis is the most accurate method to reclassify individuals to the correct CVD risk group.

Cardiovascular risk management

Traditional CVD risk factors are predictive of future CVD risk among patients with RA, as in the general population[79]. For example, the authors of a systematic review found that hypertension and diabetes mellitus correlate with roughly a twofold increase in CVD risk in patients with RA, and patients with hypercholesterolaemia (although not properly defined in the study) have a 73% increased risk of CVD morbidity[80]. These risk factors are highly prevalent in patients with RA[81], a fact that might not only reflect a high background occurrence, but possibly also shared disease pathways and additional effects of RA disease activity, disability or adverse effects of RA treatment. The current EULAR recommendations advise that patients with RA should be screened every 5 years for CVD risk factors, with awareness that CVD risk factors are influenced by changes in RA disease activity and antirheumatic treatment[11]. Furthermore, the identification of CVD risk factors should lead to the initiation of CVD preventive medication regimens, as recommended for the general population[9]. If lifestyle interventions are insufficient, pharmaceutical agents (such as antihypertensives and lipid-lowering drugs) should be administered to achieve guideline-recommended treatment targets for blood pressure, lipid levels and glycated haemoglobin[11]. Individual CVD risk factors are discussed in the following sections.

Hypertension

The prevailing European definition of hypertension is a persistent systolic blood pressure/diastolic blood pressure (SBP/DBP) >140/90 mmHg[82], whereas guidelines from the USA state that all persons with an SBP/DBP >130/80 mmHg are hypertensive[83]. Globally, hypertension is widespread, and about a third of the adult general population is hypertensive[84].

Hypertension in RA. Although results from a 2011 meta-analysis[85] did not reveal any differences in the prevalence of hypertension between patients with RA and individuals who did not have RA, accumulating evidence indicates that hypertension is more common in patients with RA compared with the general population[86,87]. One study that included 400 patients with RA in secondary care in England revealed a high prevalence of hypertension (71%)[88]. Furthermore, hypertension in that population was underdiagnosed, especially in those aged <44 years, and undertreated in patients with RA who were over 65 years of age. Another study of 309 patients with RA showed that, of the patients who used antihypertensive medication, 47% had not reached their blood pressure goal[89]. By contrast, evidence from a California database indicated that patients with RA were more likely to have blood pressure measurements recorded and to receive antihypertensive medication than individuals who did not have RA[90]. Furthermore, only one-quarter of those who had an indication for antihypertensive treatment obtained the recommended blood pressure treatment goals[91].

Blood pressure goals. No evidence exists that the blood pressure goals for patients with RA should be different from those recommended for the general population. Treatment goals for SBP/DBP as recommended by the ESC and European Society of Hypertension (ESH)[82] are shown in TABLE 2. Specific blood pressure goals are provided for different age groups, with the lowest goals for those under the age of 65 years and less aggressive treatment goals recommended for those over the age of 65 years[82]. The Systolic Blood Pressure Intervention Trial (SPRINT)[92] has been of particular importance for the latest recommendations for antihypertensive treatment. SPRINT demonstrated the benefit of an SBP target of <120 mmHg (rather than the conventional target of <140 mmHg) in reducing CVD morbidity and mortality and all-cause mortality in patients with hypertension at high risk of CVD[92]. The only important disagreement between the European guidelines[82] and the hypertension guidelines from the USA[83] is that in the USA guidelines a reduction in blood pressure to <130/80 mmHg is recommended for all individuals, regardless of age[83].

Management of hypertension. EULAR advocates that patients with RA should receive antihypertensive drugs in accordance with national CVD preventive guidelines for the general population[11]. In this Review, we focus on the recommendations from the 2018 ESC/ESH guidelines for the management of arterial hypertension[82]. Hypertension management in patients with RA is summarized in FIG. 3. Various antihypertensive drugs might be suitable to lower blood pressure and, in general, a two-drug combination is advised[82]. The preferred combinations include a blocker of the renin–angiotensin system (RAS) (an angiotensin-converting enzyme inhibitor or an angiotensin receptor blocker) with either a calcium channel blocker or a diuretic, although other combinations could be used. Combinations that include beta blockers are particularly recommended for angina, post-myocardial infarction, heart failure or heart rate control. A step-up approach to triple therapy (compromising an RAS blocker, a calcium channel blocker and thiazide or a thiazide-like diuretic) is recommended for resistant hypertension[82]. However, in patients with resistant hypertension, the possibility of secondary hypertension should be considered. Monotherapy with a single blood pressure-lowering drug is suitable for low-risk patients with an SBP >140 mmHg but <150 mmHg and for frail elderly patients. After the initiation of antihypertensive treatment, the blood pressure lowering effect should be recorded within 2 months. Electrolytes and kidney function should be monitored if treatment with diuretics and/or RAS blockers is initiated.

Table 2 | **Blood pressure treatment targets in patients with rheumatoid arthritis**

Age group (years)	Additional comorbidities	SBP targets	DBP targets
18–65	Hypertension	130 mmHg or lower if tolerated; not <120 mmHg	70–79 mmHg
	Diabetes mellitus		
	CAD		
	Stroke or TIA		
	CKD	<140 to 130 mmHg if tolerated	
Over 65	All comorbidities	130–139 mmHg if tolerated	

Information in this Table is adapted from the 2018 European Society of Cardiology/European Society of Hypertension guidelines for the management of arterial hypertension[82]. CAD, coronary artery disease; CKD, chronic kidney disease; DBP, diastolic blood pressure; SBP, systolic blood pressure; TIA, transient ischaemic attack.

Lipids and lipoproteins

Lipoproteins as total cholesterol and LDL cholesterol are well known major modifiable risk factors involved in atherosclerotic CAD in the general population. The Cholesterol Treatment Trialists meta-analysis demonstrated that risk reductions for major adverse cardiovascular events with statin therapy have a consistent relationship with the absolute reduction in LDL cholesterol[93]. This section covers total cholesterol and LDL cholesterol in the context of CVD risk for patients with RA. High-density lipoprotein (HDL) cholesterol and triglycerides are also covered separately in later sections.

Total cholesterol and LDL cholesterol in RA. As with the general population, patients with RA also have an increased risk of CVD when lipid levels are either low or high[94]. Interpretation of the role of lipid abnormalities in patients with RA compared with individuals who do not have RA is somewhat distorted by the complex interactions between lipid levels and RA disease activity and the effects of these interactions on the future risk of CVD. Patients with RA have lower total cholesterol and LDL cholesterol[24] than individuals who do not have RA, which has been be associated with inflammation in these patients[30,95,96] (FIG. 4). Low LDL cholesterol levels complicate CVD risk evaluation when using risk calculators and could cause underestimation of risk because cholesterol is a major variable in risk algorithms. Interpretation of the total burden of lipid abnormalities on CVD risk among patients with RA is also hampered by the inconsistent (or lacking) definition of lipid abnormalities in published studies. Hyperlipidaemia has been defined as having a total cholesterol level >6.2 mmol/l (>240 mg/dl)[97]. Despite wide-ranging estimates, lipid abnormalities are probably quite commonly present in patients participating in studies; roughly one third of patients with RA have hypercholesterolaemia[81,87].

Indications for lipid-lowering treatment and treatment targets. Despite the fact that patients with RA have a high risk of CVD, they seem to receive inadequate CVD screening and CVD risk management (both for primary and secondary prevention)[91], including lipid-lowering treatment. For all practical purposes, statins are the undisputed first choice for lipid-lowering therapy. Other lipid-lowering medications that can be considered are ezetimibe and proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors. Individual lipid-lowering therapies are discussed in detail in the following sections.

To decide if there is an indication for lipid-lowering treatment, the total burden of CVD risk factors should be evaluated using a CVD risk calculator, such as the SCORE calculator, which is recommended by the ESC and EAS for CVD risk evaluation in the general population in Europe[9]. EULAR recommendations for the management of CVD risk advocate that the SCORE calculator

or a country-specific algorithm should be used for CVD risk estimation in patients with RA[11]. The current CVD risk classes and LDL cholesterol treatment targets for the general population are also recommended for patients with RA, and are summarized in TABLE 3.

Briefly, when an estimated risk of a fatal CVD event in the next 10 years is <5%, the patient is categorized as low to moderate risk and there is no indication for CVD preventive measures. The cut-off value for initiation of CVD preventive treatment with statins is a CVD

Antihypertensive treatment

Two-drug combination
- Hypertensive patients with RA should be treated equally to hypertensive patients without RA
- Initiation of antihypertensive treatment with a two-drug combination is advised
- A RAS blocker in combination with either a CCB or a diuretic is the first drug of choice
- Beta blockers are only indicated for heart rate control, angina, post-myocardial infarction or heart failure

Adverse effects

Kidney failure: NSAID use results in a vasoconstriction of the afferent renal arteriole; RAS blockers cause efferent renal arteriolar vasodilatation; and diuretics can cause hypovolaemia

Lipid-lowering therapy

Statins
- Statin initiation in patients with RA should be carried out in the same way as for the general population
- To initiate statin therapy in patients older than 75 years, consider quality of life and whether life expectancy is >5 years
- Statins can be considered in the elderly as part of symptom relief treatment for intractable angina pectoris

Adverse effects
- Myalgia: change to another statin or start with the lowest dose of a statin taken 2 to 3 times a week and increase by one tablet per week every 3–4 weeks until maximum tolerable dose is acquired
- Liver: if the increase in liver enzymes is >3 times the ULN, discontinue statin treatment; if the increase in liver enzymes is <3 times the ULN, reduce the statin dose or stop or reduce methotrexate

Drug–drug interactions
- If a patient is taking tocilizumab, choose fluvastatin, pravastatin or rosuvastatin
- Drugs that are metabolized by CYP3A4 potentially interact with statins, so care should be taken when using: calcium antagonists (e.g. amlodipine, diltiazem and verapamil); anti-infective medications (e.g. erythromycin, clarithromycin, HIV proteases and antifungal agents); and some other medications (e.g. amiodarone, gemfibrozil and cyclosporine)
- Although not a drug, grapefruit juice is also not recommended when using statins

Statins and ezetimibe
- No difference in efficacy and safety in patients with RA from what is reported for the general population
- If a patient with RA is intolerant to statins, monotherapy with ezetimibe can be considered

PCSK9 inhibitors

Drug interactions or other adverse effects could theoretically occur when a patient uses a bDMARD and a PCSK9 inhibitor, but this effect has not been evaluated

Management of hypertriglyceridaemia
- The recommendations for patients with RA do not differ from those for the general population
- If a fibrate is combined with a statin the fibrate should be taken in the morning and the statin in the evening to reduce the risk of myopathy
- Fibrates, niacin and omega-3 fatty acids can be used when triglycerides are >10 mmol/l (900 mg/dl) to prevent pancreatitis

Management of low HDL cholesterol
- Low HDL cholesterol levels (<1.0 mmol/l for men; <1.2 mmol/l for women) are common in metabolic syndrome
- Low HDL cholesterol levels can be raised by: a change in diet to contain more free fatty acids; exercise; moderate alcohol intake; smoking cessation; and weight loss

Management of high lipoprotein(a)

Reduce other CVD risk factors, including LDL cholesterol, thereby reducing overall CVD risk

Glycaemic control in T2DM
- The first drug of choice for initiation of antiglycaemic treatment in T2DM is metformin
- Patients with T2DM and atherosclerotic CVD or who are at high risk of CVD should be offered treatment with an SGLT2 inhibitor or a GLP1-RA

Fig. 3 | **Cardiovascular disease risk management in patients with rheumatoid arthritis.** Cardiovascular disease (CVD) risk management in patients with rheumatoid arthritis (RA) differs little from strategies recommended for the general population. The guidelines summarized in this overview are synthesized from the 2019 European Society of Cardiology (ESC)/European Atherosclerosis Society guidelines for the management of dyslipidaemias[9], the 2018 ESC/European Society of Hypertension guidelines for the management of arterial hypertension[82] and the 2019 ESC guidelines on diabetes, pre-diabetes and cardiovascular diseases developed in collaboration with the European Association for the Study of Diabetes[143]. bDMARD, biologic DMARD; CCB, calcium channel blocker; CYP3A4, cytochrome P450 3A4; GLP1-RA, glucagon-like peptide 1 receptor agonist; HDL, high-density lipoprotein; LDL, low-density lipoprotein; PCSK9, proprotein convertase subtilisin/kexin type 9; RAS, renin–angiotensin system; SGLT2, sodium–glucose cotransporter 2; TDM2, type 2 diabetes mellitus; ULN, upper limit of normal.

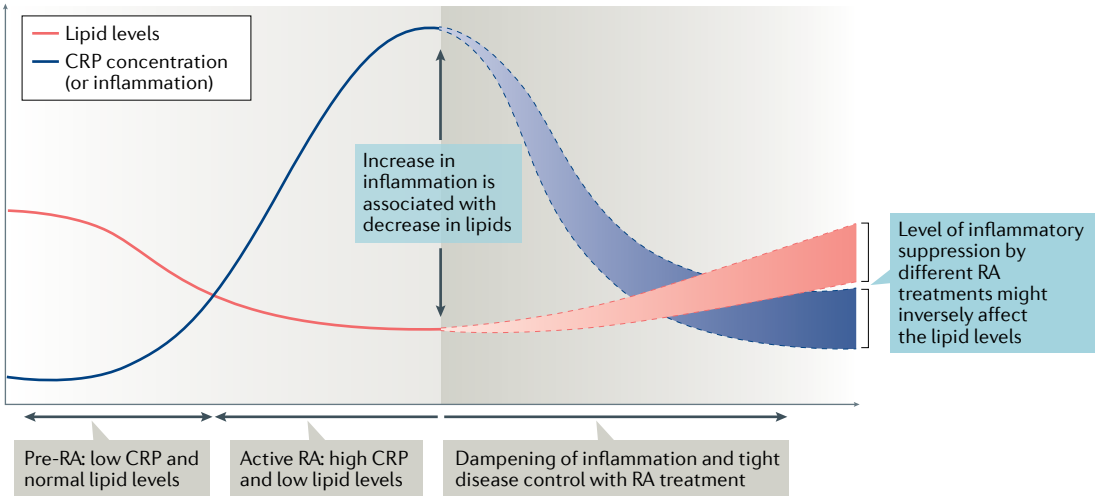

Fig. 4 | **The interaction between lipids and inflammation in rheumatoid arthritis.** In the pre-rheumatoid arthritis (RA) phase, before disease manifestation, inflammation is low. As disease activity increases, inflammation (here shown as C-reactive protein (CRP)) increases and lipid levels decrease. Antirheumatic medication dampens inflammation (reduces CRP concentrations), which is accompanied by an inverse increase in lipid levels. FIGURE 4 is adapted with permission from REF.[96], Oxford University Press.

risk ≥5%. Classification to the high CVD risk group is appropriate if the patient has diabetes mellitus (either type 1 or type 2), a total cholesterol level >1.8 mmol/l and/or a calculated CVD risk by SCORE ≥5% and <10%. The recommended LDL cholesterol goal for patients in the high CVD risk group is <1.8 mmol/l (<70 mg/dl)[9]. Patients with established CVD and/or a calculated CVD risk by SCORE ≥10% and an LDL cholesterol level >1.8 mmol/l (>70 mg/dl) should be classified to the very high CVD risk group. For patients in the very high CVD risk group, an LDL cholesterol goal of <1.4 mmol/l (<55 mg/dl) is recommended[9].

Guidelines for lipid-lowering treatment and treatment targets. In the 2018 guideline on the management of blood cholesterol from the USA[10], the focus is not on specific LDL cholesterol targets, as in the ESC/EAS guidelines[9]. Instead, statin treatment is recommended to aim at a certain percentage reduction of LDL cholesterol and should be initiated at low, moderate or high intensity depending on the estimated risk of CVD[10]. For primary prevention, the ACC/AHA PCE atherosclerotic CVD risk calculator is recommended for estimating the risk of a non-fatal or fatal CVD event in the next 10 years[39]. If the estimated risk is between 5% and 7.5%, moderate-intensity statin therapy can be considered. For an estimated CVD risk ≥7.5% but <20%, moderate-intensity statin therapy to reduce LDL cholesterol by 30–49% is advocated, and for patients with an estimated risk ≥20%, high-intensity statin treatment aimed at an LDL cholesterol reduction of ≥50% is recommended[10]. Regarding lipid-lowering therapy after a CVD event, the guidelines from the USA distinguish between patients who are not at very high risk and those at very high risk. For patients not at very high risk following a CVD event, high-intensity statin treatment with a goal of ≥50% reduction in LDL cholesterol is recommended. If a patient has experienced a CVD event and has one or more CVD risk factors (age ≥65 years, heterozygous familial hypercholesterolaemia, history of prior coronary artery bypass surgery or percutaneous coronary intervention, diabetes mellitus, hypertension, chronic kidney disease, current smoking, LDL cholesterol level ≥2.6 mmol/l (≥100 mg/dl) despite maximally tolerated statin therapy and ezetimibe, or history of congestive heart failure), then maximal statin therapy (80 mg atorvastatin or 40 mg rosuvastatin) is recommended. If the LDL cholesterol level is still >1.8 mmol/l (>70 mg/dl), then adding ezetimibe is considered reasonable before initiation of a PCSK9 inhibitor[10].

The guidelines from the USA on the management of blood cholesterol present a specific section with recommendations for adults with chronic inflammatory disorders[10]. The guidelines state that, in adults aged 40 to 75 years with an LDL cholesterol value of 1.7–4.8 mmol/l who have a 10-year risk of atherosclerotic CVD ≥7.5%, a chronic inflammatory disorder is a risk-enhancing factor that favours moderate-intensity or high-intensity statin therapy. Furthermore, a fasting lipid profile and assessment of atherosclerotic CVD risk factors are advocated as guides as to who might benefit from statin therapy and for monitoring or adjusting lipid-lowering therapy before, and 4 weeks and 12 weeks after, initiation of DMARD treatment. The guidelines from the USA underline that high RA disease activity is associated with low lipid levels, which could lead to the underestimation of future CVD risk. Therefore, they recommended that lipid levels are rechecked 2–4 months after the inflammatory disease has been brought under control[10]. Lifestyle changes should be the focus for the first 3–6 months of intervention, including smoking cessation, and the CVD risk estimate should then be reassessed. If the predicted risk of CVD in the coming 10 years is still >5%, then moderate-intensity statin treatment can be initiated[10].

Table 3 | **Cardiovascular disease risk classes, lipoprotein targets and interventions for patients with rheumatoid arthritis**

CVD risk class[a]	Description	Target levels	Intervention
LDL cholesterol			
Low	CVD risk <1%	LDL cholesterol <3.0 mmol/l (<116 mg/dl)	Consider adding a lipid-lowering drug (a statin or ezetimibe) if LDL cholesterol is 3.0 to <4.9 mmol/l; add a statin if LDL cholesterol >4.9 mmol/l
Moderate	CVD risk ≥1% to <5% and LDL cholesterol 2.6 to <3 mmol/l (100 to <115 mg/dl)	LDL cholesterol <2.6 mmol/l (<100 mg/dl)	Consider adding a lipid-lowering drug (statins, statins and ezetimibe or ezetimibe monotherapy) if LDL cholesterol 2.6 to <4.9 mmol/l; add a statin if LDL cholesterol ≥4.9 mmol/l
High	CVD risk ≥5% and <10% and LDL cholesterol 1.8 to <2.6 mmol/l (70 to <100 mg/dl) and/or diabetes mellitus and/or a total cholesterol >8.1 mmol/l	LDL cholesterol <1.8 mmol/l (<70 mg/dl) or ≥50% reduction of baseline LDL	Statins; statins and ezetimibe; ezetimibe monotherapy; a statin and a PCSK9 inhibitor; PCSK9 inhibitor monotherapy
Very high	CVD risk >10% and LDL cholesterol >1.8 mmol/l (70 mg/dl) and/or established CVD	LDL cholesterol <1.4 mmol/l (<55 mg/dl) or ≥50% reduction of baseline LDL cholesterol	Statins; statins and ezetimibe; ezetimibe monotherapy; a statin and a PCSK9 inhibitor; PCSK9 inhibitor monotherapy
HDL cholesterol			
Increased risk	HDL cholesterol <1.0 mmol/l (<40 mg/dl) in men and <1.2 mmol/l (<45 mg/dl) in women	No target HDL cholesterol level, but recommended HDL cholesterol >1.0 mmol/l (>40 mg/dl) in men and >1.2 mmol/l (>45 mg/dl) in women	Exercise; diet; weight loss; moderate alcohol intake
Triglycerides			
Normal	Triglyceride level <1.7 mmol/l (<150 mg/dl)	No target level, but a triglyceride level <1.7 mmol/l (<150 mg/dl) indicates lower risk	Exercise; avoiding sugar and refined carbohydrates; optimization of glucose control in diabetes mellitus; withdrawal of oestrogen therapy; weight loss; choose healthier dietary fats; moderate alcohol intake
Borderline high	Triglyceride level 1.8 to 2.2 mmol/l (150 to 199 mg/dl)		
High	Triglyceride level 2.3 to 5.6 mmol (200 to 499 mg/dl)		
Very high	Triglyceride level ≥5.7 mmol/l (≥500 mg/dl)		Fibrates, niacin and omega-3 fatty acids can be used when triglyceride levels are >10 mmol/l to prevent pancreatitis

Information in this Table adapted from the 2019 European Society of Cardiology/European Atherosclerosis Society guidelines for the management of dyslipidaemias: lipid modification to reduce cardiovascular risk[9]. CVD, cardiovascular disease; HDL, high-density lipoprotein; LDL, low-density lipoprotein; PCSK9, proprotein convertase subtilisin/kexin type 9. [a]CVD risk classes described using the Systematic Coronary Risk Evaluation calculator.

Overall, the ESC/EAS guidelines for the management of dyslipidaemia[9] are more aggressive than the blood cholesterol guidelines from the USA[10], as they aim for LDL cholesterol goals of <1.4 mmol/l (<55 mg/dl) in individuals at very high CVD risk and have LDL cholesterol goals for those with lesser levels of risk. By contrast, the guidelines from the USA do not advocate fixed targets or goals for LDL cholesterol[10]. In patients with very high CVD risk, the guidelines from the USA use an LDL cholesterol threshold >1.8 mmol/l (>70 mg/dl) for decision-making regarding intensive lipid-lowering treatment, and in patients with high CVD risk who should receive primary prevention the guidelines recommend a goal of 50% lowering of LDL cholesterol with a maximally tolerated statin and, if needed, ezetimibe.

Statins. Given that patients with RA have a high absolute risk of CVD, a background of systemic inflammation and low lipid levels, it is questionable whether the lipid-lowering effects of statins are the same in patients with RA as in the general population. Aside from the Trial of Atorvastatin for the Primary Prevention of Cardiovascular Events in Patients with RA (TRACE-RA)[98], randomized controlled trials (RCTs) with hard CVD end points that investigate the use of lipid-lowering medication and/or antihypertensive treatment in patients with RA are lacking and will probably never be performed. In TRACE-RA, the study ended after the inclusion of 3,002 patients with RA owing to unexpectedly low CVD event rates. Despite not reaching statistical significance, possibly because of a short median follow-up time of 2.5 years, a 32% reduction in CVD events was reported in those who received statin treatment, which is comparable to the results of other large RCTs of statins[99,100]. Furthermore, after adjusting for baseline confounding variables, stratification by centre, compliance and non-study statin treatment, the hazard ratio for future CVD events was 0.54 (95% CI, 0.30–0.98; $P = 0.045$). In this respect, TRACE-RA could be considered to have positive results concerning statin protection for major adverse cardiovascular events in patients with RA in a primary preventive setting. A 2018 study from Taiwan using nationwide registry data that included 49,227 patients with RA revealed lower event rates of first-onset acute coronary syndrome in patients with RA who received statin therapy than in those who did not[101].

With regards to secondary CVD prevention with statins in patients with RA, promising results have

been reported from the post hoc analyses of two large statin trials with hard CVD end points. In the TNT and IDEAL trials, high-dose statin treatment with 80 mg atorvastatin and conventional statin treatment with either 40 mg simvastatin or 10 mg atorvastatin had comparable lipid-lowering effects and risk reduction for CVD in patients with coronary heart disease who did and did not have inflammatory joint diseases[102]. Currently, there are no indications in the literature that obtainment of lipid goals lower than those recommended for the general population would improve CVD outcomes in patients with RA.

Despite a lack of RCT data on lipid-lowering therapy for patients with RA, promising real-world data on statin use in these patients are emerging. A report from a preventive cardio-rheuma clinic demonstrated that out of 435 referred patients with inflammatory joint diseases, 64% had an indication for lipid-lowering therapy, and 90% reached their lipid goals within three consultations, highlighting that CVD risk factor management with statins is feasible in this patient population[103].

When choosing the starting dose of statin, the LDL cholesterol level should be taken into consideration and the dose should be adjusted until lipid goals are achieved. Several types of statins were used in the preventive cardio-rheuma clinic and the same lipid-lowering effect and adverse effects described for the general population were seen in the patients with RA[103]. Systemic inflammation (measured by CRP or erythrocyte sedimentation rate) or the use of any antirheumatic medication did not influence the dose of statin needed to obtain recommended LDL cholesterol goals[104]. Overall, the CVD risk classes, recommended LDL cholesterol targets and interventions for CVD prevention in patients with RA might be the same as those for the general population.

Adverse effects of statins. Statins are generally well tolerated by both the general population[105] and by patients with RA[98,102]. Specific points to consider regarding the use of statins in patients with RA are presented in FIG. 3. Notably, only a small proportion of adverse effects reported by individuals taking statins are related to the statins themselves, as the adverse effects of statins are reported equally in the placebo and statin arms of RCTs[106]. In addition, the perceived adverse effects of statins might also be caused by the nocebo effect[107]. The risk of myopathy is low (1 in 1,000 individuals), but myopathy can on rare occasions lead to rhabdomyolysis and renal failure[9,108]. If a patient experiences myalgia with no increase in creatine kinase (occurring in 5–10% of treated patients), statin treatment can be continued if the muscle pain is tolerable for the patient[9,108]. An increase of creatine kinase <5 times the upper limit of normal in two blood samples is considered acceptable[9,108].

An increase in liver enzymes is dependent on statin dose and occurs in 0.5–2.0% of treated patients[9]. Measurement of liver enzymes after statin therapy is especially important in patients with RA who are taking methotrexate because this drug might affect liver function. A small increase in type 2 diabetes mellitus (T2DM) has been reported among those in the general population using statins[109], but this was mainly seen in individuals with an increased risk of developing T2DM before statin initiation. No data are available on the risk of T2DM development in patients with RA who use statins. Furthermore, the benefit of using statins on CVD risk surpasses the risk of developing T2DM in patients at high risk of future CVD[9,109].

Statin use in elderly individuals. Older people often have comorbidities and take multiple medications, which is also common among patients with RA. Thus, special care should be taken regarding drug–drug interactions in these populations. Additionally, owing to commonly altered pharmacokinetics in elderly individuals, starting on a low-dose statin and titrating up to the maximum tolerated dose in relation to the recommended LDL cholesterol goal might be beneficial[9]. In a meta-analyses of 28 statin RCTs with hard CVD outcomes that included 186,854 individuals from the general population, 8% were older than 75 years of age, and statin treatment resulted in a substantial reduction in CVD events irrespective of age[105]. However, less direct evidence of benefit was seen in patients older than 75 years of age if they did not already have atherosclerotic disease[105]. Currently, there are no studies in elderly patients with RA on the effects of statins on CVD risk reduction. Therefore, we suggest that when considering starting statin treatment in an elderly individual their life expectancy should be 5 years or more, and the recommendations for elderly individuals in the general population should be followed[9].

In general, treatment with statins is recommended for older people with atherosclerotic CVD in a similar way to that for younger patients. Treatment with statins is recommended for primary prevention, according to the level of risk, in people aged ≤75 years and initiation of statin treatment for primary prevention can be considered in this population if patients are classed as high risk (or above) for CVD. The statin should be started at a low dose if there is substantial renal impairment and/or the potential for drug–drug interactions and then titrated upwards to achieve recommended LDL cholesterol treatment goals[9].

Drug–drug interactions. Evidence is scarce on drug–drug interactions involving antirheumatic medications and lipid-lowering medications. In patients with active RA, IL-6 might suppress cytochrome P450 (CYP) activity, and the bioavailability of certain drugs might be increased. Tocilizumab, an IL-6 inhibitor, might reverse the suppression of CYP3A4 activity, thereby increasing the clearance of statins metabolized by this cytochrome[110]. For example, in a study investigating the drug–drug interaction between simvastatin and tocilizumab in patients with RA, at 1 week and 5 weeks after tocilizumab infusion the pharmacokinetics of simvastatin were reduced compared with the pharmacokinetics before tocilizumab infusion[110]. Simvastatin, lovastatin and, to a lesser extent, atorvastatin are metabolized by the hepatic isoenzyme CYP3A4 and often have drug–drug interactions, so any new drug for RA should be checked for interactions. Statins that are not metabolized by CYP3A4 are often preferred for use in patients

on multi-drug regimens for this reason. Care should be taken when treating patients with RA (as with the general population) with drugs that are metabolized by CYP3A4 and that potentially interact with statins, including calcium antagonists (such as amlodipine, diltiazem and verapamil), and anti-infective medications (such as erythromycin, clarithromycin, HIV proteases and antifungal agents) and some other medications (such as amiodarone, gemfibrozil and cyclosporine). Although not a drug, grapefruit juice can substantially increase the blood concentrations of statins metabolized by CYP3A4 (such as simvastatin and atorvastatin), and thus consumption of grapefruit juice is not recommended when using these drugs.

Ezetimibe. Ezetimibe is a drug that selectively inhibits intestinal cholesterol absorption through inhibition of NPC1-like intracellular cholesterol transporter 1 (REF.[111]). A trial in 2002 demonstrated an additional reduction in LDL cholesterol levels of 12–19% when ezetimibe was taken in addition to a statin by patients with primary hypercholesterolaemia[112]. In the IMPROVE-IT trial, the combination of statins with ezetimibe effectively reduced the risk of CVD in the general population[113]. Although no information exists on the effect of this combination of lipid-lowering medications on CVD in patients with RA, there are no indications that the effect in these patients should be different from that observed in the general population.

PCSK9 inhibitors. Monoclonal antibodies against PCSK9 are available for high-risk individuals with atherosclerotic CVD or familial hypercholesterolaemia who are already taking the maximally tolerated statin therapy but who require greater LDL cholesterol reduction. In combination with a statin, a PCSK9 inhibitor reduces LDL cholesterol by 40–60% and reduces CVD events by 15–20%[114]. Two PCSK9 inhibitors are commercially available, alirocumab and evolocumab. Both the FOURIER trial[114] and ODYSSEY OUTCOMES trial[115] confirmed that the risk of major adverse cardiovascular events can be reduced by a further lowering of LDL cholesterol with PCSK9 inhibitors beyond that achieved with statin therapy alone. However, no solid data exist to prove that the incremental benefit of reducing LDL cholesterol from 50 mmol/l to 10 mmol/l is worth the substantial cost of treating a patient with a PCSK9 inhibitor[108]. Both the safety of the drug and the safety of low LDL cholesterol levels has been proven[108]. Effects on cognitive function have been a particular concern in relation to very low LDL cholesterol levels, a topic that has been addressed in a subgroup of patients in the FOURIER trial who were examined for psychomotor speed, memory and executive function and for whom no difference was found between those taking placebo or evolocumab[116].

Patients with RA often use biologic DMARDs (bDMARDs) that are monoclonal antibodies. Theoretically, drug interactions or other adverse effects can occur when a patient uses two monoclonal antibodies, although the particular combination of PCSK9 inhibitors and bDMARDs has not been evaluated.

Triglycerides

Hypertriglyceridaemia. Inflammation, especially chronic inflammation, can cause increased triglyceride levels. Among other reasons for high triglyceride levels are genetic factors, hypothyroidism, metabolic syndrome, poorly controlled diabetes mellitus and alcohol intake[9]. High triglyceride levels can also be an adverse effect of certain drugs, including diuretics, oestrogen, progestin, retinoids, glucocorticoids, beta blockers and some HIV medications. Triglyceride levels above 1.7 mmol/l (150 mg/dl) (TABLE 3) might be an independent risk factor for coronary heart disease[117], but the mechanism by which triglyceride lowering affects CVD risk in the general population has not yet been fully elucidated. However, in patients without RA receiving statin therapy after a coronary heart disease event, a triglyceride level <1.7 mmol/l was associated with a reduced risk of recurrent coronary events independent of the level of LDL cholesterol[118]. No similar data exist in patients with RA.

No specific goal is recommended for triglycerides, but a level <1.7 mmol/l (<150 mg/dl) indicates a lower risk of CVD and higher values indicate the need to control other CVD risk factors.

Management of hypertriglyceridaemia. Ways of reducing hypertriglyceridaemia include those related to a healthy lifestyle, such as daily exercise, avoiding sugar and refined carbohydrates, losing weight, consuming unsaturated fatty acids and limiting alcohol consumption[9]. Statins reduce triglyceride levels by 10–20% from baseline values[119]. Furthermore, intake of omega-3 fatty acids (2–4 g daily) is highly effective and can reduce triglycerides by 45%[120]. Nicotinic acid (niacin) also lowers triglycerides but does not reduce CVD morbidity or mortality in the general population[121]. No evidence suggests that these properties would differ in patients with RA.

Fibric acid derivatives (fibrates) are a class of medication that lower blood triglyceride levels by reducing the production of very-low-density lipoprotein (the triglyceride-carrying particle that circulates in the blood) by the liver and by speeding up the removal of triglycerides from the blood[122]. Fibrates are modestly effective in increasing blood HDL cholesterol; however, they are not effective in lowering LDL cholesterol[122]. The fibrate gemfibrozil interferes with the breakdown of certain statins, such as simvastatin and lovastatin, resulting in increased statin concentrations in the blood and hence a higher likelihood of muscle toxicity from the statin[123]. Gemfibrozil should therefore not be added to statin treatment. Caution should be taken when combining fibrates and statins, especially if kidney or liver disease is present. No data exist on fibrate use in patients with RA, and the recommendations for use in the general population should be followed[9,10].

HDL cholesterol

HDL cholesterol levels and CVD risk. HDL cholesterol is involved in reverse cholesterol transport, and increasing levels of HDL cholesterol have an inverse relationship with CVD events in population studies and clinical trials[124]. Infusion of the major protein of HDL cholesterol

has proven beneficial effects on atherosclerotic plaques[125], but most therapeutic approaches have focused on raising HDL cholesterol through cholesterol ester transfer protein (CETP) modulation[126]. The results of large clinical trials of the effect of CETP inhibitors on CVD events have been disappointing[127]. Additionally, Mendelian randomization studies related to CETP should be interpreted with caution owing to limitations of these studies[128,129]. Thus, there is no current evidence from RCTs or genetic evidence that raising HDL cholesterol will reduce the risk of atherosclerotic CVD in the general population or in patients with RA.

Management of low HDL cholesterol. Low levels of HDL cholesterol (defined as <1.0 mmol/l in men and <1.2 mmol/l in women) are common in metabolic syndrome. A non-pharmaceutical approach for raising low HDL cholesterol levels involves dietary changes, an increase in exercise, moderate alcohol intake, smoking cessation and weight loss[33]. These are general lifestyle recommendations that improve the risk of CVD in both the general population and in patients with RA[33]. Whether this improvement in CVD risk is a result of increased HDL cholesterol levels is unknown.

Lipoprotein(a)

High levels of lipoprotein(a) are associated with an increased risk of CVD[130–132] and are frequently found in individuals and families with premature CVD despite other risk factors. Lipoprotein(a) seems to be a weaker CVD risk factor than LDL cholesterol[133,134], and there is no evidence that reducing lipoprotein(a) with either niacin or CETP inhibitors reduces CVD risk. In contrast to statins, PCSK9 inhibitors reduce lipoprotein(a) levels by 30–40%, and data from a 2019 study imply that PCSK9 inhibitors might reduce the risk of CVD partly through this reduction in lipoprotein(a)[135]. However, the mechanism for lipoprotein(a) reduction by PCSK9 inhibition is unclear, and these results need to be verified. Currently, there are no recommendations for lowering elevated lipoprotein(a), either in the general population or in patients with RA. The present approach to managing high lipoprotein(a) levels in patients with RA is to reduce other CVD risk factors, including LDL cholesterol, thereby reducing the overall CVD risk.

Type 2 diabetes mellitus

Worldwide, the prevalence of diabetes mellitus is ~9% of the adult population, of which ~90% is T2DM[136]. The diagnosis of T2DM requires the presence of any one or a combination of: a fasting glucose concentration ≥7 mmol/l (≥126 mg/dl); a random plasma glucose concentration ≥11.1 mmol/l (≥200 mg/dl) in association with the presence of hyperglycaemia or hyperglycaemic crisis; a glucose concentration ≥11.1 mmol/l (≥200 mg/dl) 2 h after a standardized oral glucose tolerance test; or the presence of a measured glycated haemoglobin ≥6.5% (≥48 mmol/mol)[137].

CVD risk in patients with RA and T2DM. Although patients with RA might be at increased risk of developing insulin resistance and T2DM owing to physical inactivity, glucocorticoid treatment and a high degree of disease activity, reports on the prevalence of T2DM in patients with RA compared with those who do not have RA are conflicting[77,86,138–140]. However, available data point towards an increased prevalence of T2DM among patients with RA[86]. Interpretation of data is limited by the high rate of underdiagnosis of T2DM; potentially half of individuals with T2DM are undiagnosed[141]. In addition, if an individual has both RA and T2DM, the risk of CVD is increased 2.6-fold compared with someone who does not have RA and T2DM[142].

Management of CVD risk and T2DM in patients with RA and T2DM. No evidence exists that the treatment of T2DM in patients with RA should differ from that recommended for the general population (FIG. 3; TABLE 4). Briefly, if tolerated and in the absence of contraindications, metformin has been the principle agent to treat T2DM in patients with RA, as for the general population[143]. However, patients with T2DM who have established CVD or a high risk of future CVD should be considered for treatment with a sodium–glucose cotransporter 2 inhibitor (such as empagliflozin) or a glucagon-like peptide 1 receptor agonist at an early stage, regardless of whether they are already using metformin, to reduce CVD morbidity and mortality[143]. Typically, patients with T2DM have multiple risk factors for CVD and thus require particularly attentive screening and management of CVD risk factors via lifestyle intervention and CVD preventive medication[143]. In principle,

Table 4 | **Recommended glycaemic and lipid targets in patients with rheumatoid arthritis and diabetes mellitus**

Patient population	Recommendation	Treatment targets
Most patients (adjusted according to duration of diabetes mellitus, age and comorbidities)	Glycaemic control	Glycated haemoglobin <7.0% (<53 mmol/mol)
Very high CVD risk	Lipid-lowering therapy	LDL cholesterol <1.4 mmol/l (<55 mg/dl) and LDL cholesterol lowering >50%
High CVD risk	Lipid-lowering therapy	LDL cholesterol <1.8 mmol/l (<70 mg/dl) and LDL cholesterol lowering >50%
Moderate CVD risk	Lipid-lowering therapy	LDL cholesterol <2.6 mmol/l (<100 mg/dl)

Information in this Table adapted from the 2019 European Society of Cardiology/European Atherosclerosis Society guidelines for the management of dyslipidaemias: lipid modification to reduce cardiovascular risk[9] and the 2019 European Society of Cardiology guidelines on diabetes, pre-diabetes, and cardiovascular diseases developed in collaboration with the European Association for the Study of Diabetes[143]. CVD, cardiovascular disease; LDL, low-density lipoprotein.

Box 1 | **Advice on managing lifestyle-related cardiovascular disease risk factors**

Tobacco smoking
- Patients with rheumatoid arthritis (RA) who smoke tobacco should receive smoking cessation advice.
- Smoking cessation programmes should be implemented in rheumatology clinics.

Body weight and composition
Increased lean mass and reduced body fat percentage could have beneficial effects on cardiovascular disease risk in patients with RA.

Diet
- Heart-friendly diet recommendations such as Mediterranean-style diets and Dietary Approaches to Stop Hypertension should be employed when delivering dietary advice to patients with RA.
- Brief advice such as 'ask, advice and assist' can be effective for changing to more heart-friendly diets.

Exercise and physical activity
The WHO recommends 30 min of moderate-to-intense activity five times per week, which is safe and advisable for patients with RA.

lipid-lowering and antihypertensive treatments for patients with RA and T2DM do not differ from those for patients with T2DM without RA[9] (FIG. 3; TABLE 4). Although high LDL cholesterol levels have been associated with a reduced risk of developing T2DM, and low levels of LDL cholesterol with an increased risk of T2DM[144], no solid evidence exists of increased insulin intolerance after statin treatment. Recommended glycaemic and lipid targets for patients with RA who have T2DM are summarized in TABLE 4.

Inflammation

Atherosclerosis is known to have features of both local and low-grade systemic inflammation[145]; however, it is currently unclear whether this is simply an epiphenomenon, or if inflammation has a causal role in atherosclerosis. The inflammatory biomarker high-sensitivity CRP can be used to independently predict future vascular events[146]. Statin therapy reduces high-sensitivity CRP concentrations in healthy individuals[147], patients with stable coronary disease[148] and in those with acute coronary syndrome[149]. The magnitude of the benefit associated with statin therapy correlates in part with the achieved high-sensitivity CRP concentrations. CRP values and the erythrocyte sedimentation rate have also been linked with increased CVD morbidity in patients with RA, even after adjusting for traditional CVD risk factors[150]. The inflammatory processes in the rheumatoid synovium and atherosclerotic plaques are remarkably similar[96], suggesting that the intensity of vascular inflammation is an important factor in the development of accelerated atherosclerosis in patients with RA. Although the exact mechanisms by which rheumatic inflammation and atherosclerosis influence each other remains to be determined, growing evidence supports the notion that pro-inflammatory cytokines such as TNF and IL-6 disrupt endothelial haemostasis, leading to vascular dysfunction — an early step in atherogenesis[150].

In the JUPITER trial, which included healthy individuals without hyperlipidaemia but who had increased high-sensitivity CRP values, treatment with rosuvastatin significantly reduced the incidence of major cardiovascular events ($P < 0.00001$)[109].The inclusion criteria for this trial was an LDL cholesterol level <3.4 mmol/l (<139 mg/dl) and a high-sensitivity CRP value >2 mg/l. Patients with RA commonly have lower lipid levels than individuals without RA, but their CRP is often twofold to tenfold higher than that of the general population[96]. To further investigate the role of inflammation in atherosclerotic disease, and the pro-inflammatory cytokine IL-1β in particular, the Canakinumab Anti-inflammatory Thrombosis Outcomes Study (CANTOS) tested a selective monoclonal antibody against IL-1β in individuals with a history of myocardial infarction and found a reduction in major adverse cardiovascular events without an effect on LDL cholesterol[151]. Three doses of canakinumab were tested (50 mg, 150 mg and 300 mg); however, only the 150 mg dose showed a protective effect against future CVD. The fact that the highest and lowest doses of canakinumab did not protect against future CVD events leaves uncertainty as to the cardio-protective effects of anti-inflammatory medications in individuals without RA. Similarly, the Cardiovascular Inflammation Reduction Trial (CIRT), another large placebo-controlled trial, reported that use of low-dose methotrexate did not reduce the CVD event rate compared with placebo in patients with a history of myocardial infarction or multi-vessel coronary disease[152]. Notably, the inclusion criteria for participating in CIRT and CANTOS were different; CANTOS required a CRP >2 whereas CIRT did not have specific CRP requirements. However, this difference might not have affected the general results regarding the effects of these anti-inflammatory medications on atherosclerosis. Thus, whether medications such as monoclonal antibodies targeting cytokines or conventional synthetic DMARDs in patients with RA will affect the development of atherosclerosis or CVD outcomes needs further investigation.

Lifestyle-related CVD risk factors

The WHO estimate that around three-quarters of CVD events can be prevented by improving lifestyle-related CVD risk factors[153]. Although the relative effects of lifestyle factors on CVD outcome in patients with RA are unknown, substantial harmful effects have been suggested. In this section, modifiable lifestyle-related CVD risk factors are discussed, along with recommendations for their management (BOX 1).

Tobacco smoking. Smoking is a well-established risk factor for developing RA, and a high proportion of patients with RA are smokers[154]. Patients with RA who smoke have an increased risk of CVD events and all-cause mortality compared with patients with RA who do not smoke[155]. In fact, smoking and hypertension are the traditional CVD risk factors with the highest population-attributable risk for CVD in patients with RA[79]. Moreover, smoking might lead to high RA disease activity and blunt the effects of common antirheumatic medications[156]. The opposite, that smoking cessation might be associated with low disease activity, has been demonstrated in a register study of 3,311 patients with RA[157]. This topic is currently under further investigation in an ongoing RCT[158].

Evidence for beneficial effects of smoking cessation in patients with RA is also emerging[159]. In fact, smoking cessation in patients with RA is a predictor of reduced CVD event rates compared with current smoking (HR, 0.70; 95% CI, 0.51–0.95; $P = 0.022$)[157].

Although rheumatologists report that they are keen to address smoking as a CVD risk factor, programmes to promote smoking cessation are largely lacking in rheumatology clinics[160]. The UK National Health Service has developed a 30 second algorithm to deliver advice on smoking cessation to patients with RA[161]. Intensive smoking cessation counselling and nicotine-replacement or non-nicotine medications can also be used. When delivering advice on smoking cessation to patients with RA, clinicians should be aware of potential RA-specific factors, such as unawareness of the association between RA and smoking, and smoking being used as a distraction from pain or as a coping mechanism.

Weight and body composition. Around 60% of patients with RA are either overweight or obese (defined as having a BMI ≥25 kg/m^2 and ≥30 kg/m^2, respectively), which is comparable to the general population[162,163]. However, although being overweight or obese is a risk factor for CVD events in the general population, studies have reported both cardio-protective[164] and harmful[165] effects in patients with RA. One hypothesis is that the BMI is a poor proxy for the nutritional and metabolic state of patients with RA, as reduced lean mass in these patients might mask increased body fat percentages. This state, known as rheumatoid cachexia, might affect up to 20% of patients with RA and has a negative effect on CVD risk factors[166]. No evidence exists that modifying body composition in patients with RA will reduce CVD morbidity and mortality; however, it seems logical that, as for the general population, an increase in lean mass and a reduction in fat percentage would incur beneficial effects on CVD risk in patients with RA.

Diet. Aside from suggestions of a cardio-protective effect of Mediterranean-style diets[167], evidence concerning the effect of diet on CVD risk in patients with RA is scarce. Accordingly, dietary recommendations for the general population should be followed for patients with RA[11]. The Mediterranean-style and Dietary Approaches to Stop Hypertension dietary patterns are two examples that include an increased intake of fruits, vegetables, whole grains, low-fat dairy products, lean meat, legumes, nuts, seeds, seafood and vegetable oils and a reduced intake of dietary cholesterol, sugars, refined grains, sodium, alcohol, saturated fat and *trans*-fatty acids[168]. A 2018 randomized controlled pilot study in patients with inflammatory joint disease found that 4 min of standardized advice on a cholesterol-reducing diet accompanied by a brochure provided by a cardiologist had the same effect on changes in diet as 60 min of individually tailored dietary advice provided by a dietician[169]. Larger studies are warranted to verify these results. Although optimal dietary advice would be individually tailored from a dietician, in a clinical setting with limited resources brief advice might be equally effective in helping a patient to change to a cholesterol-reducing diet, and could be given by any health personnel and easily implemented in any type of outpatient clinic.

Exercise and physical activity. Patients with RA are often physically inactive and have low cardiorespiratory fitness[170], which might be caused by RA-related barriers including pain, fatigue, stiffness and reduced mobility, or a perception that physical activity could lead to higher disease activity and joint damage[171]. In fact, physical activity is not only safe for patients with RA, it might also have beneficial effects on disease activity and symptoms[172]. Increased inactive or sedentary time is associated with traditional CVD risk factors and an increased 10-year risk of CVD in patients with RA[173]. Although some beneficial effects of physical activity on CVD risk factors and CVD surrogate biomarkers have been reported in patients with RA[174], an effect on hard CVD end points has not yet been investigated.

Compared with pharmaceutical therapies, exercise is a relatively underutilized treatment for CVD risk in patients with RA. An exercise programme has been devised that is specifically tailored for patients with RA[175], and it seems wise to recommend the global recommendations on physical activity for health published by the WHO and the European recommendations for CVD prevention, which advocate 30 min of moderate-to-intense activity five times per week[33]. If symptoms of previously unrecognised angina pectoris or peripheral artery disease should occur during exercise, proper medical evaluation must be sought before physical activity is resumed in patients with RA, as for the general population.

Antirheumatic treatment and CVD risk

Medications used for the treatment of rheumatic diseases might affect CVD risk. Although NSAIDs have long been associated with an increased risk of CVD in the general population, a meta-analysis demonstrated no evidence that their use was associated with increased CVD risk in patients with RA[176]. Low doses of glucocorticoids (<7.5 mg/day) might be beneficial owing to their anti-inflammatory effects, whereas high doses might be detrimental in relation to CVD risk in patients with RA[177]. A 2019 study on the adverse effects of glucocorticoids in patients with RA confirmed long-standing findings of a 33% increased risk for T2DM, a 30% increased risk of thrombotic stroke or myocardial infarction and a 30% increased risk of death in patients taking these drugs[178]. The risk of adverse events was higher with increasing cumulative and average daily glucocorticoid doses.

The use of DMARDs (particularly methotrexate and hydroxychloroquine, but also biologic agents) is thought to decrease CVD risk[179,180], potentially owing to their suppression of systemic inflammation; however, these findings might be confounded by indication. The safety of TNF inhibitors and other biologic agents has to some extent been investigated. The adverse effects of TNF inhibitors on lipid profiles and fasting glucose concentrations are a particular concern[181]. However, a systematic literature review suggested a decreased risk of CVD in patients with RA being treated with TNF inhibitors[182].

The increase in lipid levels in these patients might actually reflect a normalization of their lipid profile towards what it was before the onset of RA[96] (FIG. 4). Patients who respond well to TNF inhibitors seem to benefit the most with regard to reductions in CVD risk[183]. However, a lack of information as to whether the antirheumatic medication consisted of monotherapy with a TNF inhibitor or combination therapy of a TNF inhibitor with methotrexate is a recurrent problem in the studies that have so far evaluated the influence of bDMARDs on CVD events. The concurrent use of methotrexate is therefore a potential confounder. To date, the studies published on this topic have had short follow-up times, whereas the progression of atherosclerosis is a protracted process. Furthermore, few studies are adjusted for confounding by indication, which is a major concern because patients with the most severe and long-standing rheumatic disease are the most likely to be treated with bDMARDs. A 2019 meta-analysis showed that treatment with the IL-6 inhibitor tocilizumab might be associated with a reduced risk of CVD events compared with treatment with TNF inhibitors, and that use of conventional synthetic DMARDs might be associated with an increased risk of CVD events and stroke[184]. Another meta-analysis, this time on the effect of TNF inhibitors on heart failure in patients with RA, concluded that more good-quality studies are needed before the effects of biologic agents on heart failure in RA can be determined[185]. Although conflicting results have been published, it seems that the evidence collected so far suggests an overall beneficial effect of bDMARDs on the risk of CVD in patients with RA[186].

The efficacy of targeted synthetic DMARDs (such as Janus kinase (JAK) inhibitors) in reducing joint inflammation has been proven in RCTs[187]. However, in the phase II and III RCTs, a signal of increased thromboembolic events emerged that requires further investigation[188]. The advantage of JAK inhibitors over bDMARDs is that they are orally administered and have a short half-life. Two JAK inhibitors have been approved by the FDA for the treatment of RA: tofacitinib and baricitinib. The efficacy and safety of tofacitinib has been studied in several double-blinded phase III RCTs in patients with RA. CVD-related adverse effects are reportedly related to the increases in LDL cholesterol and HDL cholesterol levels[189]. Preliminary results from the Safety Study of Tofacitinib Versus TNF Inhibitor in Subjects with RA study have shown that patients with RA treated with high-dose tofacitinib (10 mg twice daily) have an increased risk of pulmonary embolism and death[190]. Baricitinib has been studied in three clinical trials in patients with RA, two compared with placebo and one with the TNF inhibitor adalimumab. Similar to tofacitinib, an increase in LDL cholesterol and HDL cholesterol levels occurred with baricitinib[191]. However, initiation of statin therapy in patients with RA who were treated with baricitinib substantially reduced the LDL cholesterol levels[192]. A 2019 meta-analysis on the risk of CVD events after treatment with JAK inhibitors concluded that the short-term risk of CVD was not increased by the use of these drugs, but that more data are needed to confirm their CVD safety, especially around the increased prevalence of thromboembolic events that occurred with high-dose tofacitinib and baricitinib treatment[193].

Conclusions

The increased risk of CVD in patients with RA has been known for decades, but implementing measures to reduce this risk into clinical practice is challenging. Awareness of the high CVD risk in patients with RA is low among the patients themselves and among health personnel. In addition, the recording of CVD risk factors and risk evaluation is inadequate in patients with RA. Furthermore, patients with RA are undertreated with CVD preventive medication, both in primary prevention and after they have had a CVD event. The overarching goal should be that patients with RA have a structured system for CVD prevention equal to that of other high-risk patient populations. This Review synthesizes the evidence on CVD prevention from several guidelines and recommendations for the general population and describes how to manage CVD risk in patients with RA to provide a useful resource for primary care physicians, rheumatologists, cardiologists, internists and other health personnel.

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Global epidemiology of gout: prevalence, incidence, treatment patterns and risk factors

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Abstract | Gout is the most common inflammatory arthritis and occurs when hyperuricaemia, sustained elevation of serum urate levels resulting in supersaturation of body tissues with urate, leads to the formation and deposition of monosodium urate crystals in and around the joints. Recent reports of the prevalence and incidence of gout vary widely according to the population studied and methods employed but range from a prevalence of <1% to 6.8% and an incidence of 0.58–2.89 per 1,000 person-years. Gout is more prevalent in men than in women, with increasing age, and in some ethnic groups. Despite rising prevalence and incidence, suboptimal management of gout continues in many countries. Typically, only a third to half of patients with gout receive urate-lowering therapy, which is a definitive, curative treatment, and fewer than a half of patients adhere to treatment. Many gout risk factors exist, including obesity, dietary factors and comorbid conditions. As well as a firmly established increased risk of cardiovascular disease and chronic kidney disease in those with gout, novel associations of gout with other comorbidities have been reported, including erectile dysfunction, atrial fibrillation, obstructive sleep apnoea, osteoporosis and venous thromboembolism. Discrete patterns of comorbidity clustering in individuals with gout have been described. Increasing prevalence and incidence of obesity and comorbidities are likely to contribute substantially to the rising burden of gout.

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Gout is the most common form of inflammatory arthritis globally. The risk of gout increases with age, and it is thus more common in ageing populations. Gout results from sustained elevation of serum urate levels (hyperuricaemia) that leads to the deposition of monosodium urate crystals in joints, tendons and other tissues, which triggers recurrent episodes of pronounced acute inflammation, known as gout flares. Despite gout being one of only a few 'curable' rheumatic diseases (through the use of pharmacological urate-lowering therapies (ULTs)), management of the disease is inadequate in many parts of the world, owing to low uptake of ULT and patient adherence to these medications. Gout is often accompanied by various comorbidities, including cardiovascular disease (CVD), chronic kidney disease (CKD), obesity and other conditions. In this Review, we summarize the current understanding of the epidemiology of gout, in terms of trends in prevalence and incidence, treatment and comorbidities, focusing on data published since the publication of a similar review in 2015 (REF.[1]).

Gout prevalence

As gout is the most common inflammatory arthritis globally, understanding trends in gout prevalence is of great importance to facilitate adequate health-care resource planning, not least because gout can be 'cured' using accessible and inexpensive treatments. It is difficult to estimate the global occurrence of gout accurately owing to a lack of data for many countries and the highly variable prevalence estimates across different geographical regions and populations that are obtained using different disease definitions (FIG. 1; TABLE 1). A gout diagnosis should ideally be based on classification criteria or the demonstration of monosodium urate crystals in aspirated joint fluid or tophi, but most studies rely on self-reported diagnosis or identification of diagnostic codes or prescription of gout-specific medication in medical registries, which are prone to recall or misclassification bias. Underascertainment is possible due to long intercritical periods between flares, individuals not consulting for flares that they self-manage, or when prevalence and/or incidence are measured in

Key points

- Gout is a common chronic crystal deposition disorder that affects between <1% and 6.8% of the population depending upon the population studied.
- Both prevalence and incidence of gout seem to be rising across the globe.
- Management of gout continues to be poor, with fewer than one half of patients receiving definitive 'curative' urate-lowering therapy.
- Adherence to urate-lowering therapy is often poor and rates of non-persistence are high.
- Obesity and comorbidities are important risk factors for gout and are important drivers of its rising prevalence and incidence.

secondary or tertiary care registries rather than primary care where most patients with gout are managed. A 2015 meta-analysis of 71 studies of gout prevalence published between 1962 and 2012 found a pooled global prevalence of 0.6% (95% CI 0.4–0.7%), although there was marked statistical heterogeneity among the included estimates[2].

Gout prevalence by geographical region

Oceania. The highest prevalence of gout in the world has been reported in Oceanic countries, particularly in specific ethnic groups, such as Taiwanese Aboriginals and Maori, for which a prevalence exceeding 10% has been reported[1]. In the past decade, four studies from Australia in which gout diagnosis was based on medical records or self-reports have been published, showing prevalence ranging from 1.5% in adults >20 years of age in a large electronic general practice database for the period 2008–2013 to 6.8% for self-reported medically diagnosed gout in the entire population in 2015 (REFS[3–6]) (TABLE 1).

North America. The reported prevalence of gout in the USA is fairly high, with 3–4% of adults affected in 2007–2008 (REF.[7]). Consistent with this estimate, in the 2015–2016 National Health and Nutrition Examination Survey (NHANES), a stratified, multistage sample representative of the US adult population, the prevalence of self-reported, health professional-diagnosed gout was 3.9% (REF.[8]). In an analysis of a health-care database with full coverage of the 4.5 million inhabitants of British Columbia in Canada, the overall prevalence of gout, defined as the presence of one or more International Classification of Diseases (ICD) codes for gout, was 3.8% in 2012 (REF.[9]), which is a substantial increase from the estimated gout prevalence of 3% in the adult population in 2003 (REF.[10]).

Europe. Gout is common in Europe, with studies in France, Germany, Greece, Italy, the Netherlands, Spain and the UK finding a gout prevalence ranging from 1% to 4% for the period 2003–2014 (REF.[1]), which has been confirmed in subsequent studies from other European countries[11]. Studies of Scandinavian populations, based predominantly on medical records and/or diagnostic codes, found gout prevalence ranging from 0.02% to 1.8%, although with considerable heterogeneity in terms of duration of data collection, data sources and age of included subjects[12–16].

Asia. The prevalence of gout varies markedly among Asian countries, and new data from China[17] and South Korea suggest that the prevalence is increasing. A 2017 meta-analysis of 30 studies published from 2000 to 2016 found a pooled prevalence of gout in the adult population in China of 1.1%, with prevalence increasing slightly from 1.0% in 2000–2005 to 1.3% in 2010–2016 (REF.[18]). In South Korea, a study analysing data from a national health claims database for specialized care found that gout prevalence increased from 0.35% in 2007 to 0.76% in 2015 in the entire population, and predicted a further increase to 1.66% by 2025 (REF.[19]). By contrast, a study in the United Arab Emirates including 3,985 randomly chosen adult primary care patients in 2009 found a gout prevalence of 0.1%[20].

Other regions. Data are scarce concerning the prevalence of gout in Africa, whereas the prevalence of gout in Central and South American countries has previously been reported to be low[1]. In Community Oriented Program for Control of Rheumatic Diseases (COPCORD) studies, the estimated prevalence of gout was 0.1% in a semi-urban community of 2,454 individuals ≥15 years of age in Nigeria in 2015–2016 (REF.[21]) and 0.4% in Ecuador in 2014 (REF.[22]).

In summary, the prevalence of gout varies considerably across the globe, with the highest prevalence in Oceanic countries, particularly in indigenous and South Pacific island populations, and the lowest prevalence in the developing world. In addition to previously reported increasing prevalence of gout in Europe and the USA, there is evidence of increasing prevalence in Australia (self-reported), Canada, China and South Korea.

Gout prevalence by demographic factors

The prevalence of gout is influenced by demographic factors, such as ethnicity, age and sex. Ethnicity-related differences in diet, comorbidity patterns and genetics can increase susceptibility to gout. In the NHANES study in the USA for the period 2015–2016, the prevalence of gout in the non-Hispanic black population was 4.8%, compared with 4.0% in the non-Hispanic white population and 2.0% in the Hispanic population[8]. In a study in Sweden, the risk of incident gout was greater for men born in Iraq (adjusted hazard ratio (aHR) 1.82, 95% CI 1.54–2.16) or Russia (aHR 1.69, 95% CI 1.26–2.27) than for men born in Sweden. In women, the relative risk (RR) was highest for individuals born in Africa (RR 2.23, 95% CI 1.50–3.31), Hungary (RR 1.98, 95% CI 1.45–2.71) or Iraq (RR 1.76, 95% CI 1.13–2.74)[23]. Surprisingly, the risk of gout was lower for men born in Greece and Spain, in which the prevalence of gout is high, than for men born in Sweden[24,25].

The prevalence of gout increases with advancing age (FIG. 2), a pattern that is seen over the entire lifespan in men and especially after the menopause in women, possibly owing to the uricosuric effects of oestrogen. Consequently, the demographic profile of the population being studied affects the prevalence of gout. Furthermore, the rise in life expectancy across the world is contributing to the increasing prevalence of gout worldwide. In Canada, the prevalence of gout among men and women 70–79 years of age in 2012 was 11.8%, compared with 5.1% in those 50–59 years of age and <1% in those <30 years of age[9]. In Australia, the prevalence of gout in men in 2013 ranged from 0.2% in

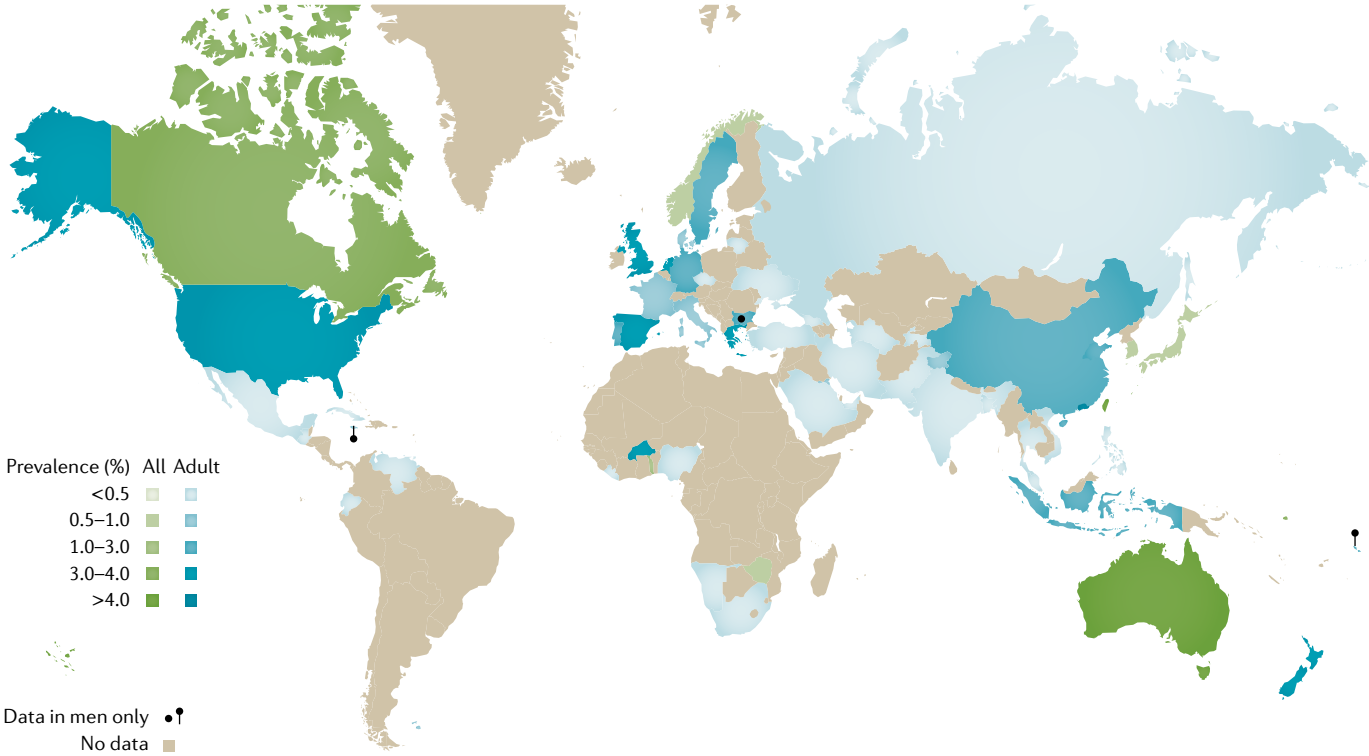


Fig. 1 | **Estimated prevalence of gout worldwide.** The figure shows the prevalence in different parts of the world, as indicated in the key. Prevalence is highest in developed countries, whereas data are lacking for some parts of the developing world, particularly Africa and South America.

those 25–29 years of age to 11.05% in those ≥85 years of age[3], whereas in women, the prevalence was very low premenopausally, increasing to 4.64% in those ≥85 years of age. The prevalence of gout in South Korea in 2015 was 1.36% in men and 0.16% in women[19]. Furthermore, men also develop gout earlier in life than women, with a Danish study of adult incident gout cases between 1995 and 2015 finding a mean age of onset of 65.3 years in men and 71.4 years in women[16].

Gout incidence

Fewer studies have addressed the incidence of gout than the prevalence of the disease (TABLE 2). Increased incidence of gout has been reported previously in the USA and the UK over several decades up to 2012 (REF.[1]), whereas the incidence of gout decreased in Taiwan in the period 2005–2010 (REF.[1]). Studies published in 2015 and thereafter showed substantial increases in gout incidence over recent decades in the USA, Canada, Denmark, Sweden and South Korea (FIG. 3), and confirmed greater incidence in men than in women and also increasing incidence in the later decades of life. Recent studies in North America and Scandinavia found a 1.5–2-fold increase in gout incidence over the past two to three decades[9,13,16,26–28], whereas gout incidence in South Korea increased by 25% between 2009 and 2015 (REF.[19]). Thus, it is clear that the incidence of gout has increased in many countries over recent decades and that ageing populations in these countries may further exacerbate these observed increases in incidence. Furthermore, as many of these studies used registry data, changes in reimbursement systems over time may have affected the frequency of diagnosis.

Urate-lowering therapy

ULT involves various strategies to reduce urate levels, typically pharmacological agents that reduce purine breakdown (that is, xanthine oxidase inhibitors (XOIs)) or increase urinary excretion of uric acid (that is, uricosuric agents). Allopurinol is the oldest XOI currently in use; originally described in 1956, it was demonstrated to have urate-lowering effects in 1964. Febuxostat entered the market in 2008 and is a potent XOI that is more selective than allopurinol and has a less complicated dosing regimen, albeit with a higher price and concerns regarding cardiovascular safety[29]. Marketed uricosuric agents that increase uric acid excretion, mainly through the urine, include probenecid, sulfinpyrazone and benzbromarone, as well as the more recently approved lesinurad[30,31]. Drawbacks of all uricosuric agents include multiple-dose regimens, unsuitability for use in patients with severe CKD and increased risk of kidney stone formation, although they can be combined with XOIs when hyperuricaemia is refractory to monotherapy[29].

ULT prescription

Successive surveys have demonstrated widespread suboptimal treatment of gout, with less than half of patients ever receiving ULT[32], a pattern that seems to be continuing. For example, the proportion of patients with gout in Australia who received ULT decreased from 57% in 2008–2013 to 36% in 2015 (REFS[3,6]), whereas only 42%

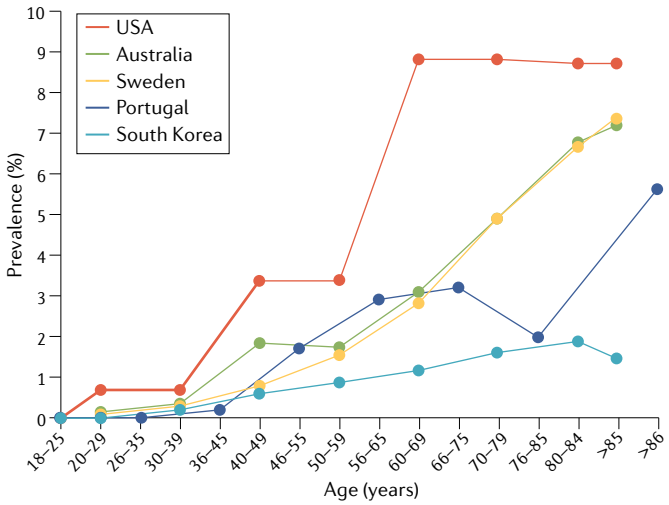
of patients with gout in western Sweden received ULT in 2012 (REF.[13]). The proportion of patients receiving ULT did not change between 2000 and 2012 in British Columbia, Canada (22%), or between 2007 and 2014 in the USA (33%; based on NHANES data)[8,9]. Data from a national health claims database suggest that a greater proportion of patients with gout in South Korea receive ULT, although there seemed to be a slight but steady decline between 2007 (86.1%) and 2015 (81.6%)[33]. In a study in the UK, whereas 44% of patients with gout fulfilled the criteria for ULT at diagnosis, only 17% received treatment, although this improved somewhat at 1 year after diagnosis (61% and 30%, respectively) and at 5 years after diagnosis (87% and 41%, respectively)[34]. Similarly, in Sweden 30% of patients with incident gout received ULT 1 year after diagnosis[35].

Overall, allopurinol is by far the most commonly prescribed ULT, with febuxostat and uricosuric drugs used

Table 1 | **Prevalence of gout worldwide**

Location	Setting	Gout diagnosis or definition	Data collection period	Age (years)	Prevalence	Ref
Australia	General practice point-of-care electronic records over a 5-year period	Receiving allopurinol or colchicine; gout diagnosis or tophi; podagra in medical records	2008–2013	>20	1.54% (1.27% age-standardized)	3
Australia	Medical records from more than 550 practices with 'active' patients (that is, three or more visits every 2 years)	Gout diagnosis in medical records	2013–2016	≥18	1.6%	4
Australia	Longitudinal study of 4,056 adults ≥18 years of age randomly selected from the electronic White Pages	Self-reported gout or use of gout-specific medication	2008–2010	>25	5.2% (8.5% in males; 2.1% in females)	5
Australia	Face-to-face interviews with representative population sample	Self-reported medically-diagnosed gout	2015	All ages	6.8% (11.3% in males; 2.4% in females)	6
Canada	Province-wide medical records database for British Columbia	Physician-diagnosed gout	2000–2012	All ages	3.8%	9
China	Two randomly selected population samples in the Shantou region (COPCORD)	ACR 1977 criteria	2012	≥16	1.08%	17
China	Meta-analysis	ACR 1977 criteria	2000–2016	All ages	1.1% (1.7% in males, 0.5% in females)	18
Denmark	Nationwide specialist-care registry	Physician-diagnosed gout	1995–2015	≥18	0.68%	16
Ecuador	COPCORD	ACR 1977 criteria	2014	≥18	0.4%	22
Nigeria	COPCORD; all inhabitants of Katon Rikkos ($n = 2,454$)	ACR 1977 criteria	2015–2016	≥15	0.1%	21
Norway	Questionnaire to 10,000 people not living in institutions; nationally administered reimbursement for primary care; or specialist care	Self-reported or physician-diagnosed gout	2012	All ages	0.54% in males and 0.39% in females (questionnaire); 0.7% in males and 0.22% in females (reimbursement records); and 0.09% in males and 0.02% in females (specialist care)	15
Portugal	10,661 randomly selected adult participants	ACR 1977 criteria	2011–2013	≥18	1.3% (2.6% in males, 0.1% in females)	11
South Korea	National health claims database for specialized care	Physician-diagnosed gout	2007–2015	All ages	0.76%	19
Sweden	Health-care database with complete coverage of all inhabitants of Stockholm	Physician-diagnosed gout	2013–2014	All ages	0.55% (0.8% in males; 0.3% in females)	14
Sweden	Health-care database with complete coverage of all inhabitants of Skane	Physician-diagnosed gout	1998–2013	≥18	1.69% (2.44% in males; 0.96% in females)	12
Sweden	Health-care database with complete coverage of all inhabitants of Western Sweden Health Care Region	Physician-diagnosed gout	2000–2012	≥20	1.8% (2.5% in males; 1.1% in females)	13
United Arab Emirates	Individuals who attended 1 of 13 primary health-care clinics; questionnaire and positive answers were followed up by rheumatologist examination	ACR 1977 criteria	2009	18–85	0.1%, (0.3% in males; 0.0% in females)	20
USA	Third National Health and Nutrition Examination Survey (NHANES-III)	Self-reported gout	2015–2016	Adults	3.9% (5.2% in males; 2.7% in females)	8

ACR, American College of Rheumatology; COPCORD, Community Oriented Program for Control of Rheumatic Diseases.



	Prevalence (%) in indicated age range (years)							
Location	20–29	30–39	40–49	50–59	60–69	70–79	80–84	>85
USA	0.7	0.7	3.4	3.4	8.8	8.8	8.7	8.7
Australia	0.08	0.33	1.84	1.68	3.03	4.9	6.72	7.19
Sweden	0.06	0.27	0.8	1.54	2.83	4.89	6.61	7.38
South Korea	0.03	0.2	0.59	0.85	1.15	1.59	1.9	1.49

	Prevalence (%) in indicated age range (years)							
Location	18–25	26–35	36–45	46–55	56–65	66–75	76–85	>86
Portugal	0	0	0.2	1.7	2.9	3.2	2	5.6

Fig. 2 | **Age-specific prevalence of gout in five countries/territories.** The graph shows estimated prevalence by age group for Australia[3], the USA[8], Portugal[11], Sweden[13] and South Korea[19].

as alternatives in patients who do not tolerate allopurinol. In a study examining 2013–2014 NHANES data[8], allopurinol accounted for 95% of self-reported prescribed ULTs. In South Korea, 96% of ULT prescriptions in 2011 were for allopurinol, which decreased to 82% in 2015, whereas prescriptions for febuxostat increased from 6% in 2012 to 14% in 2015 and prescriptions for benzbromarone remained stable (3–4%)[33].

Patient adherence to ULT

Adherence to medication is a measure of the extent to which patients take their medication as prescribed and includes assessment of the actual intake of the medication by the patient expressed as a percentage (medication taken divided by the prescribed dose) and assessment of non-persistence, that is gaps in taking the therapy (for example, gaps of at least 30 or 60 days) or the complete discontinuation of the medication[36]. In a systematic review of 24 studies published up to August 2016 (REF.[36]), the pooled overall adherence (defined as taking >80% of the prescribed ULT) was 46% (95% CI 41–51%), with similar results obtained in studies performed in the USA (45%, 95% CI 40–51%) and in other countries (48%, 95% CI 37–59%) (TABLE 3). Slightly higher levels of adherence were found in studies that were not based on prescription or claims data, probably owing to different methodologies and patient selection criteria, or to possible recall bias in studies based on self-reported data. Importantly, adherence did not change over time. In studies published since 2016, low adherence persists in the UK (39%)[37], Ireland (46%)[38] and Singapore (44%)[39], and is even lower in China (22%)[40] (TABLE 3). In the only study that examined trends in adherence over time, a gradual improvement in adherence was found, with 25% higher rates of adherence in the UK in 2010–2014 compared with 1987–1999 (REF.[37]).

A systematic review revealed that the pooled, overall non-persistence with ULT up to 2016 ranged from 54% to 87%, although the observation periods in the included studies were rather short (4–12 months)[36]. The variability of the outcomes is mainly attributable to differences in the permissible gap length and in the observation period between studies. A similarly high non-persistence (75%) over a 2-year follow-up period was found in a study in Sweden[35]. In the aforementioned UK study for the period 1987–2014, non-persistence increased from 57.8% (95% CI 57.3–58.2%) at 1 year to 80.8% (95% CI 80.4–81.2%) at 5 years after the initiation of ULT. However, non-persistence decreased, with patients who started ULT during the period 2010–2014 showing 7% better persistence than those who started during the period 1987–1999 (REF.[37]). Both the systematic review[36] and subsequent studies in Europe[35,38], Asia[40] and the USA[8] consistently showed that increasing age, male sex and having more comorbidities (such as CKD, hypertension, obesity and diabetes) increase both adherence to and persistence with ULT.

The ability to evaluate recent trends in ULT adherence has been limited in many studies by their collection of data over fairly short periods, typically the preceding 3–10 years. Taking this into account, there seems to be a modest trend towards more extensive use of ULT and better adherence to prescribed ULT regimens, although both adherence to and persistence with ULT remain unsatisfactorily low[36].

Improving ULT uptake and adherence

New and better ways to deliver ULT are needed so that adherence can be improved. Obstacles to effective ULT from the patient perspective include a lack of knowledge about the chronicity of gout and the availability of effective treatments; and beliefs that the disease is self-inflicted[39,41]. The main provider-related barriers to ULT prescription uncovered in the UK relate to health professionals' lack of knowledge of gout and management guidelines[41]. Several of these factors are modifiable, as demonstrated in randomized trials in which information provided to participants and predetermined treatment goals have been components of successful interventions[42,43]. Future trials will hopefully evaluate treatment strategies that entail the provision of information about the disease to patients and predetermined treatment goals by different health-care providers, as different treatment strategies may be more or less suitable for implementation in different health-care systems and countries.

Gout risk factors and comorbidities

Considerable advances have been made in recent years in understanding risk factors for gout, in particular the importance of obesity, lifestyle factors, comorbidities and

genetics. Most factors increase gout risk by predisposing to hyperuricaemia. The importance of hyperuricaemia as a risk factor for the development of gout has been confirmed in recent studies. In the Malmö Preventative Study, the absolute risk of gout in hyperuricaemic individuals (serum urate >405 µmol/l (6.75 mg/dl)) over more than 25 years of follow-up was 13.3% in men and 17.7% in women, compared with 2.7% and 1.9%, respectively, in those with serum urate <360 µmol/l (6 mg/dl)[28]. In a meta-analysis of data from four cohort studies (including 18,889 participants who were gout-free at baseline), the 15-year cumulative incidence of gout ranged from 1.1% (95% CI 0.9–1.4%) in individuals with serum urate <6 mg/dl to 49% (95% CI 31–67%) in those with serum urate ≥10 mg/dl. Compared with individuals with a baseline serum urate of <6 mg/dl, the adjusted hazard ratio in those with a baseline serum urate of 6.0–6.9 mg/dl was 2.7 (95% CI 2.0–3.6) and increased in a concentration-dependent manner in those with a baseline serum urate of ≥10 mg/dl to 64 (95% CI 43–96)[44]. Thus, although hyperuricaemia greatly increases the risk of developing gout, most hyperuricaemic individuals do not develop gout, even over a lengthy follow-up period. As a result, hyperuricaemia alone is insufficient to enable a diagnosis of gout without the presence of typical clinical features or evidence from joint aspiration.

Obesity and dietary factors

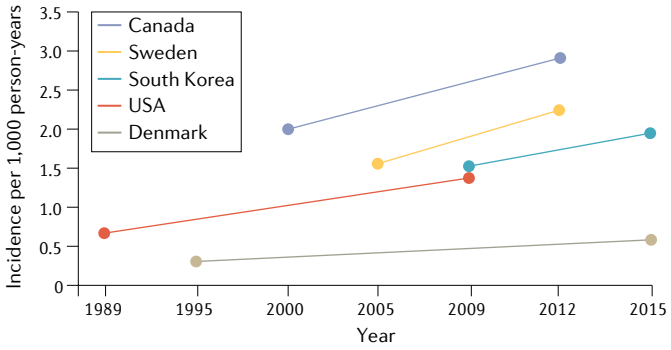
Obesity is an important risk factor for gout and is thought to be a major contributor to the rising prevalence and incidence of gout. In a 2018 meta-analysis, obesity (BMI ≥30 kg/m^2) was associated with a >2-fold increased risk of developing gout (adjusted RR (aRR) 2.24, 95% CI 1.76–2.86) compared with those with a BMI of <30 kg/m^2 (REF.[45]). In a Mendelian randomization study, higher BMI but not waist-to-hip ratio was causally associated with gout, independent of other risk factors[46]. There has been interest in the effect of weight loss achieved through bariatric surgery on gout incidence. In the non-randomized Swedish Obese Subjects intervention study, the incidence of gout was 40% lower after bariatric surgery over a follow-up of up to 26 years (aHR 0.60, 95% CI 0.48–0.75)[47].

Although the role of dietary factors in the pathogenesis of gout has been suspected for centuries, supporting epidemiological evidence has only emerged over the past 15 years. Consumption of red meat, seafood and shellfish, fructose, sugar-sweetened soft drinks and alcoholic drinks (particularly beer) increase the risk of incident gout, whereas low-fat dairy products, vitamin C and coffee are protective[1]. The prospective Singapore Chinese Health Study confirmed a slightly increased risk of gout associated with intake of total protein (aHR 1.27, 95% CI 1.12–1.44), poultry (aHR 1.27, 95% CI 1.11–1.45), and fish and shellfish (aHR 1.16, 95% CI 1.02–1.32), but not red meat (aHR 1.08, 95% CI 0.94–1.24)[48]. Meta-analyses of prospective cohort studies found an increased risk of incident gout associated with fructose and sugar-sweetened soft drink consumption (aRR 1.62 (95% CI 1.28–2.03) and 2.08 (95% CI 1.40–3.08), respectively, comparing the highest with the lowest quantile)[49,50]. These studies investigated associations between individual food types and

Table 2 | **Incidence of gout worldwide**

Location	Setting	Gout definition or diagnosis	Data collection period	Age (years)	Incidence	Ref
Canada	Medical records of >95% of Ontario residents	Diagnosis by two physicians or by one physician and gout-specific medication use	2008–2014	≥66	1,699 cases per 100,000 population; 6-year gout incidence	27
Canada	Province-wide medical records database for British Columbia	Physician-diagnosed gout	2012	All ages	2.9 cases per 1,000 person-years	9
Denmark	National specialist care registry	Physician-diagnosed gout	2015	≥18	57.5 cases 100,000 person-years	16
South Korea	National health claims database for specialized care	Physician-diagnosed gout	2015	All ages	1.94 cases per 1,000 person-years	19
Sweden	Health-care database with complete coverage of all inhabitants of Skane	Physician-diagnosed gout	2013	≥18	23.8 cases per 10,000 person-years	12
Sweden	Health-care database with complete coverage of all inhabitants of Western Sweden Health Care Region	Physician-diagnosed gout	2012	≥18	190 cases per 100,000 person-years	13
Sweden	Cohort of 33,000 adults in Malmö, Skane, followed for 28 years	Physician-diagnosed gout	1974–1992	Adults	3.8% (4.5% in males; 2.4% in females)	28
USA	Olmstead County, Rochester, MN	ACR 1977 criteria; Rome criteria; New York criteria	1989–1992 and 2009–2010	≥18	1989–1992: 66.6 cases per 100,000 persons; 2009–2010: 136.7 cases per 100,000 persons	26

ACR, American College of Rheumatology.



Location	Incidence per 1,000 person-years in indicated year						
	1989	1995	2000	2005	2009	2012	2015
Canada			2			2.9	
Sweden				1.55		2.24	
South Korea					1.52		1.94
USA	0.67				1.37		
Denmark		0.32					0.58

Fig. 3 | **Trends in gout incidence in five countries/territories.** The graph shows the estimated incidence of gout (per 1,000 person-years) in British Columbia, Canada, between 2000 and 2012 (REF.[9]), western Sweden between 2005 and 2012 (REF.[13]), Denmark between 1995 and 2015 (REF.[16]), South Korea between 2009 and 2015 (REF.[19]) and Olmsted County, Minnesota, USA, between 1989 and 2009 (REF.[26]).

gout risk, whereas recent studies have also examined the effect of overall dietary patterns on gout risk. The Dietary Approaches to Stop Hypertension (DASH) diet comprises fruit, vegetables, low-fat dairy products and reduced saturated and total fat content. Over 26 years of follow-up, men in the Health Professionals Follow-up Study who followed the DASH diet were at lower risk of incident gout (highest versus lowest quintile; aRR 0.68, 95% CI 0.57–0.80)[51]. Furthermore, in two prospective cohorts from Tzu Chi, Taiwan, a vegetarian diet was associated with a lower risk of gout (aHR 0.40 (95% CI 0.17–0.97) and 0.61 (95% CI 0.41–0.88))[52].

The relative contributions of dietary and genetic factors to the aetiology of hyperuricaemia and gout have been the subject of recent debate. In a meta-analysis of 16,760 individuals of European ancestry in five cohort studies in the USA, dietary patterns explained ≤0.3% of the variance in serum urate levels whereas common, genome-wide single-nucleotide variants explained 23.9% of the variance[53], suggesting that diet has less impact on hyperuricaemia risk than genetics. However, some researchers have questioned the suitability of the proportion of variance explained as a measure of effect and have instead contrasted the population-attributable risks associated with being overweight or obese (44%), non-adherence to a DASH-style diet (9%) and alcohol use (8%), with the fairly small variance in serum urate levels explained by these factors (8.9%, 0.1% and 0.5%, respectively)[54,55]. Thus, the relative contributions of dietary and genetic factors to the risk of hyperuricaemia and gout and the clinical effectiveness of specific dietary interventions versus weight loss are worthy of further study, although it seems that the population impact of being overweight or obese might be greater than that of specific dietary patterns.

Comorbidities

Associations between gout, hyperuricaemia and comorbidities are complex, with some diseases predisposing to hyperuricaemia and/or gout and others arising as a consequence of gout[1]. Recent research has confirmed earlier observations about traditional cardiovascular risk factors (such as hypertension and hyperlipidaemia), CVD and CKD. A large study of data from the UK Clinical Practice Research Datalink (CPRD) investigated temporal relationships between the occurrence of comorbidities before and after a first diagnosis of gout[56]. This study confirmed hypertension, hyperlipidaemia and renal disease as risk factors for gout, as well as the well-recognized association of gout with subsequent CVD and renal disease. A meta-analysis of cohort studies confirmed that hypertension and diuretic therapy predispose to gout[45]. Cohort studies from the UK, USA and Canada have also confirmed the bidirectional association between gout and CKD, with CKD predisposing to gout that in turn increases the risk of CKD progression[57–60]. In the Western Sweden Health Care Region register, most comorbidities that were present at first gout diagnosis were more prevalent in women than in men[61].

Novel gout-associated comorbidities

Several novel comorbid associations of gout have been described in recent years. Besides the established association between gout and CVD, evidence has emerged that gout increases the risk of other vascular disorders. Studies analysing CPRD data found that individuals with gout in the UK have >50% greater risk of being diagnosed with peripheral vascular disease in the decade before and after a gout diagnosis, and the risk is greater in women (aHR 1.89, 95% CI 1.50–2.38) than in men (aHR 1.18, 95% CI 1.01–1.38)[56,62]. Population-based cohort studies in Taiwan and the UK have shown that gout is associated with an increased risk of erectile dysfunction (21% in Taiwan and 31% in the UK)[63–65]. The higher incidence in the UK was possibly due to underascertainment in Taiwan, as erectile dysfunction is not covered by the Taiwan National Health Insurance programme. In Taiwan, risk of erectile dysfunction was greater in individuals with gout and comorbidities than in those with gout who do not have comorbidities, whereas the UK study stratified analyses by diabetes and CKD, and found no increased risk of erectile dysfunction associated with gout in these groups. Thus, both these studies suggest that comorbidities have a stronger influence than gout on erectile dysfunction risk.

Cohort studies in the USA, Taiwan and the UK have found that gout is associated with an increased risk of developing atrial fibrillation, possibly owing to hyperuricaemia and chronic inflammation[66–69]. In a study analysing US health-care claims data, the risk of atrial fibrillation was higher in individuals with gout and in older age groups (aHR 1.21 (95% CI 1.11–1.33) in individuals ≥40 years of age and 1.92 (95% CI 1.88–1.96) in those ≥65 years of age)[66,69]. A similar risk of atrial fibrillation in people with gout (aHR 1.38, 95% CI 1.27–1.48) was found in a study analysing the Taiwan National Health Insurance Research Database (NHIRD)[67].

However, using CPRD data, the risk of atrial fibrillation in individuals with gout was lower (aHR 1.09, 95% CI 1.03–1.16) in the UK, after adjustment for confounding lifestyle factors, comorbidities and medications[68].

An association between gout and obstructive sleep apnoea (OSA) has also been demonstrated. Hypoxia enhances nucleotide turnover, increasing the generation of purines that are metabolized to uric acid, which can lead to hyperuricaemia[70,71]. Two cohort studies that used UK primary care data found that OSA is an independent risk factor for gout. After adjusting for comorbidities, BMI and alcohol consumption[72,73], risk of incident gout was 50% higher among those with OSA than in those without OSA over a follow-up period of 1 year in The Health Improvement Network (THIN)[72] and 42% higher over a follow-up period of up to 10 years in the UK CPRD[73]. The risk of gout was highest 1–2 years after OSA diagnosis and was greater in individuals with normal BMI than in those who were overweight or obese, suggesting that OSA increases the risk of gout independent of BMI. By contrast, a cohort study analysed US Medicare claims from adults ≥65 years of age to investigate the reciprocal relationship between gout and subsequent OSA and to test the hypothesis that comorbidities, chronic inflammation and oxidative stress associated with gout could predispose to OSA[74]. The risk of incident OSA in individuals with gout was twice that in those without gout (aHR 2.07, 95% CI 2.00–2.15), after adjustment for demographics, comorbidities and medications but not obesity.

Several studies have examined the associations between gout and osteoporosis and fracture, with differing findings. A population cross-sectional study in China found an increased risk of low-trauma fracture in women with gout (OR 2.00, 95% CI 1.12–3.56) but not in men (OR 1.30, 95% CI 0.58–2.88)[75]. Gout was associated with an increased risk of fracture (aHR 1.17, 95% CI 1.14–1.21) in the Taiwan Longitudinal Health Insurance Database (LHID)[76] and of fractures of the wrist (aRR 1.12, 95% CI 0.92–1.36) and hip (aRR 1.38, 95% CI 1.14–1.68) in women in the US Nurses' Health Study[77]. Comprehensive adjustment was performed in these studies for lifestyle characteristics, comorbidities and medications, but not for corticosteroid use. In a separate cohort study of data from the Taiwan LHID, gout was associated with a slightly increased risk of osteoporosis (aHR 1.20, 95% CI 1.06–1.35) after adjustment for corticosteroid use[78]. By contrast, no association was found between gout and non-vertebral fracture (aHR 0.98, 95% CI 0.85–1.12) or hip fracture (aHR 0.83, 95% CI 0.65–1.02) in the USA or fragility fracture (aHR 0.97, 95% CI 0.92–1.02) in the UK[79,80] after adjustment for corticosteroid use. These detailed associations between gout and osteoporosis, fracture at different sites and predisposing factors (such as corticosteroid use) require further study.

Several cohort studies have shown an increased risk of venous thromboembolism (VTE) in individuals with gout, including two studies undertaken using data from the Taiwan LHID. In one study, the risk of both deep vein thrombosis (DVT) (aHR 1.66, 95% CI 1.37–2.01) and pulmonary embolism (aHR 1.53, 95% CI 1.01–2.29) was increased in individuals with gout compared with age-matched and sex-matched individuals without gout, after adjustment for age, sex and comorbidities[81]. Similarly, the other study found that the risk of DVT was

Table 3 | **Adherence to urate-lowering therapy in individuals with gout**

Study	Data collection period	Location	Adherence method[a]	Adherence to ULT (%)	Comment
Scheepers et al. (2018)[36]	1997–2015	Worldwide	MPR and/or PDC	46.0 (95% CI 40.8–51.2)	Studies published between 2004 and August 2016
		USA	MPR and/or PDC	45.2 (95% CI 39.8–50.6)	Eight studies
		Other countries	MPR and/or PDC	47.5 (95% CI 36.5–58.6)	Five studies
		Various	MPR	44.1 (95% CI 37.6–50.7)	Six MPR-based studies
			PDC	33.5 (95% CI 24.4–43.2)	Four PDC-based studies
McGowan et al. (2018)[38]	2008–2012	Ireland	MPR	6 months after initiation: 45.8 (95% CI 45.2–46.3); 12 months after initiation: 22.6 (95% CI 22.2–23.0)	34,634 patients prescribed anti-gout therapies
Sheng et al. (2017)[40]	2014	Singapore	Self-reported[b]	21.9 (95% CI 17.5–26.3)	341 male outpatients with gout at a university clinic
Chua et al. (2018)[39]	2014–2015	China	MMAS-8[c]	44.4 (95% CI 35.0–53.8)	108 patients with gout from rheumatology tertiary care
Scheepers et al. (2018)[37]	1987–2014	UK	MPR	47.6 (95% CI 47.2–48.1)	48,280 patients with gout starting allopurinol in the UK; hazard ratios for non-adherence (PDC <0.80) for calendar year starting allopurinol (reference period 1987–1999): 0.91 (95% CI 0.84–0.98) for 2000–2005, 0.86 (95% CI 0.80–0.94) for 2006–2009, 0.75 (95% CI 0.69–0.82) for 2010–2014
			PDC	38.4 (95% CI 38.0–38.8)	

[a]The medication possession ratio (MPR) is the sum of the days supply of medication for a specific period divided by the number of days of observation, with an MPR of ≥0.8 defined as adherent. The proportion of days covered (PDC) is more conservative than the MPR, as possible overlaps between two prescriptions are truncated. A PDC of ≥0.8 is defined as adherent. [b]Self-reported adherence of >80%. [c]The eight-item Morisky Medication Adherence Scale (MMAS-8) scores adherence on a scale of 0–8, with 8 indicating high adherence.

higher in patients with gout requiring ULT (HR 1.38, 95% CI 1.18-1.62) than in individuals without gout (matched for propensity score for the likelihood of gout)[82]. In the Canadian health-care database Population BC, individuals with incident gout were at increased risk of VTE (aHR 1.22, 95% CI 1.13–1.32), DVT (aHR 1.28, 95% CI 1.17–1.41) and pulmonary embolism (aHR 1.16, 95% CI 1.05–1.29) compared with those without gout, after adjustment for comorbidities, medication and health-care use[83]. Furthermore, incident gout was associated with incident VTE (aHR 1.25, 95% CI 1.15–1.35, adjusted for sociodemographic factors, comorbidities, medications and lifestyle factors) in the UK CPRD[84]. In the Atherosclerosis Risk in Communities Study in the USA, hyperuricaemia was associated with an increased risk of VTE (≥95th percentile serum urate versus ≤25th percentile; aHR 1.90, 95% CI 1.30–2.78)[85]. However, although self-reported gout was associated with an increased risk of VTE after adjustment for age, sex and ethnicity (aHR 1.49, 95% CI 1.07–2.08), this risk was attenuated after full multivariate adjustment (aHR 1.32, 95% CI 0.94–1.84).

Multiple comorbidities

Most studies have investigated epidemiological associations between gout and single comorbidities, whereas multiple comorbidities commonly coexist. However, little is known about the associations between these multiple comorbidities. Metabolic syndrome is a constellation of interrelated conditions, including obesity, dyslipidaemia, hypertension and insulin resistance, and is associated with increased risk of atherosclerosis. The prevalence of metabolic syndrome in individuals with gout in a Korean university hospital[86] and the US NHANES-III (REF.[87]) was 51% and 63%, respectively, and the NHANES-III patients with gout had three times the odds of having metabolic syndrome than age-matched and sex-matched controls without gout. In the Taiwan NHIRD, individuals with gout were more likely to have two or more comorbidities than individuals without gout, and the degree of risk increased with an increasing number of comorbidities (OR 3.58 (95% CI 2.94–4.36), 7.03 (95% CI 5.20–9.51) and 10.08 (95% CI 4.85–20.93) for the risk associated with two, three and four comorbidities, respectively)[88]. Discrete patterns of comorbidity clustering in individuals with gout have been described in cross-sectional studies in France and the UK[89,90]. The French study identified five clusters of comorbidities, whereas the UK study found only four, but both included a cluster with isolated gout and few comorbidities and one with more severe gout and prevalent renal disease (FIG. 4). In the UK study, the isolated gout cluster had the youngest age of gout onset, suggesting that genetic factors may be important in this cluster[90]. Insight into the longitudinal relationships between groups of comorbidities was gained from a latent transition analysis of data from the Taiwan LHID[91]. Three latent comorbidity classes were identified in men with incident gout: hypertension with high prevalence of gout-related comorbidities (class 1), hypercholesterolaemia with moderately prevalent comorbidities (class 2), and low prevalence of comorbidities (class 3). Over time, most patients did not change class, although a small proportion moved from class 2 and class 3 to class 1.

Comorbidities and trends in gout epidemiology

The rising prevalence and incidence of gout over recent decades is probably related to similar trends in the occurrence of comorbidities and obesity. The Global Burden of Disease (GBD) study found that diabetes, ischaemic heart disease (IHD) and alcohol use disorders are among the leading 30 causes of years lived with disability (YLD)

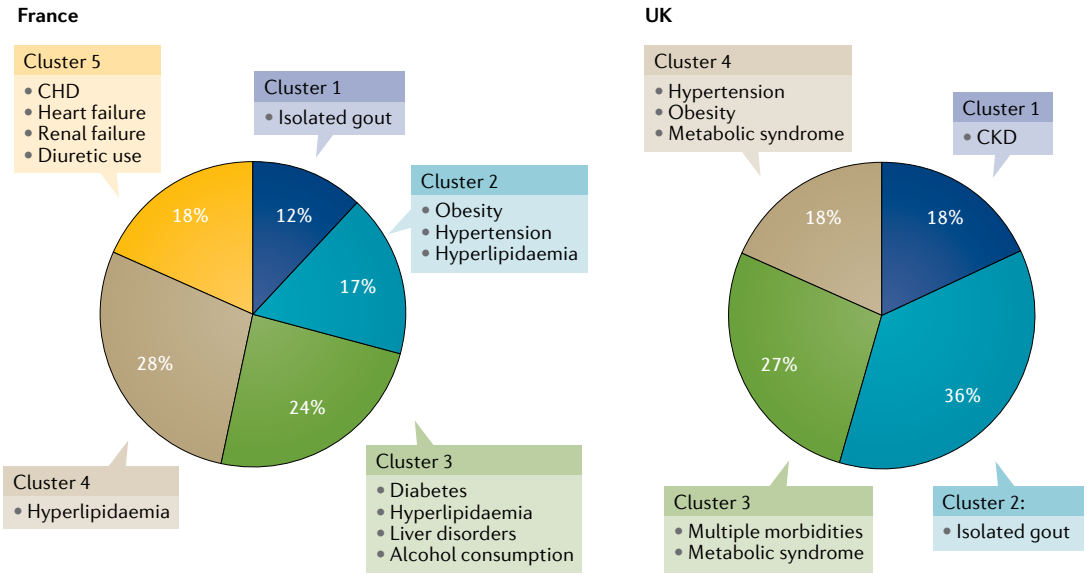


Fig. 4 | **Patterns of comorbidity clustering in individuals with gout.** Comorbidity clustering is depicted for individuals with gout in studies in France[89] and the UK[90]. Percentages are the proportion of the whole cohort in each cluster. The comorbidities listed are those that are most prevalent in that cluster. Metabolic syndrome was defined as the presence of a BMI >30 kg/m^2 and at least two of the following conditions: hyperlipidaemia, currently taking lipid-lowering agents, diabetes mellitus or hypertension[90]. CHD, coronary heart disease; CKD, chronic kidney disease.

globally in 2016 (REF.[92]). Among comorbidities commonly associated with gout, significant increases in YLD were observed for IHD, diabetes and CKD between 2006 and 2016, and the change in all-age and age-standardized YLD rates differed by ≥10% for diabetes and IHD, highlighting their importance and contribution to YLD in the elderly in the context of an ageing population. An earlier analysis of GBD study data found that the global prevalence of being overweight or obese (BMI ≥25 kg/m^2) increased between 1980 and 2013 from 29% to 37% in men and from 30% to 38% in women[93]. A small study in the USA found higher gout incidence in 2009–2010 than in 1989–1992 and found that obesity, diabetes, renal disease and hyperlipidaemia were more prevalent at gout diagnosis in the 2009–2010 cohort[26].

During the past 15–20 years, hospital admissions for gout have increased by 50–100% in the UK, USA and Sweden[94–97]. Gout now results in more hospitalizations than rheumatoid arthritis in these countries, and hospitalizations between 2007 and 2012 in Nebraska, USA, increased more for gout than for other rheumatological conditions[94]. Comorbidities contribute substantially to gout-related admissions. Of 54,215 gout admissions in Australia and New Zealand between 2009 and 2014, gout was the main reason for admission in 19,790 admissions but was a complication in 34,435 admissions for a different principal reason, most commonly CVD, infection, stroke, arrhythmia and diabetes mellitus[98].

Conclusions

Although direct comparisons between studies can only be made with caution owing to differing case definitions, sampling methods and data sources, studies in the past 5 years continue to show increases in gout incidence and prevalence across the globe. Despite this worrying increase, uptake of, adherence with and persistence with definitive, curative ULT continue to be poor. In recent years, associations between novel comorbid diseases and gout have been identified, and understanding the inter-relationships between different comorbidities in individuals with gout has improved, emphasizing the complex associations between gout and comorbidities. The rising prevalence and incidence of comorbidities and obesity is thought to be a substantial contributor to similar trends in the occurrence of gout, and addressing these will form an important pillar of endeavours to reduce gout. Thus, gout is a growing challenge worldwide and international collaborative efforts are required to address and improve widespread suboptimal management.

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Kawasaki disease: pathophysiology and insights from mouse models

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Abstract | Kawasaki disease is an acute febrile illness and systemic vasculitis of unknown aetiology that predominantly afflicts young children, causes coronary artery aneurysms and can result in long-term cardiovascular sequelae. Kawasaki disease is the leading cause of acquired heart disease among children in the USA. Coronary artery aneurysms develop in some untreated children with Kawasaki disease, leading to ischaemic heart disease and myocardial infarction. Although intravenous immunoglobulin (IVIG) treatment reduces the risk of development of coronary artery aneurysms, some children have IVIG-resistant Kawasaki disease and are at increased risk of developing coronary artery damage. In addition, the lack of specific diagnostic tests and biomarkers for Kawasaki disease make early diagnosis and treatment challenging. The use of experimental mouse models of Kawasaki disease vasculitis has considerably improved our understanding of the pathology of the disease and helped characterize the cellular and molecular immune mechanisms contributing to cardiovascular complications, in turn leading to the development of innovative therapeutic approaches. Here, we outline the pathophysiology of Kawasaki disease and summarize and discuss the progress gained from experimental mouse models and their potential therapeutic translation to human disease.

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Kawasaki disease is a systemic vasculitis that affects infants and young children[1–3]. Kawasaki disease is now the leading cause of acquired heart disease among children in North America, Europe and Japan[4,5]. The cardiovascular sequelae resulting from childhood Kawasaki disease are increasingly recognized to extend into adulthood, and the disease is no longer considered self-limiting[6–9]. The triggering agents for Kawasaki disease remain unidentified; however, results from our laboratory[10,11] and others[12,13] are consistent with the interpretation that a conventional antigen is probably responsible. Coronary arteritis and predominantly coronary artery aneurysms (CAAs) occur in up to 30% of untreated children, although this rate is reduced to 5–7% in children treated with high-dose intravenous immunoglobulin (IVIG)[3,14,15]. IVIG treatment leads to CAA regression in 60–75% of patients with Kawasaki disease[16,17]. However, the exact mechanisms by which IVIG reduces the rate of cardiovascular complications are unknown[18]. Up to 15–20% of patients with Kawasaki disease do not respond to IVIG treatment, and these individuals have an increased rate of CAA development[3,15,19–21].

Kawasaki disease is associated with infiltration of the coronary artery wall by a broad variety of innate and adaptive immune cells. Immunohistochemical analysis of human post-mortem tissues shows accumulation in the arterial wall of monocytes, macrophages and neutrophils[22,23], and the presence of activated $CD8^+$ T cells[24] as well as IgA^+ plasma cells[25,26]. The release of pro-inflammatory cytokines, such as TNF and IL-1β, by infiltrating immune cells promotes vascular endothelial cell damage and the development of CAAs[27,28].

However, understanding of Kawasaki disease pathophysiology is limited by the low availability of human tissues of the disease, failure to identify specific aetiological agents triggering the disease, and incomplete understanding of the molecular and cellular mechanisms leading to cardiovascular sequelae. Therefore, experimental animal models mimicking the human features of Kawasaki disease and their translational utility have been invaluable to investigation of this disease. In this Review, we discuss advances from human and mouse studies that have contributed to an improved understanding of Kawasaki disease pathophysiology and the cellular and molecular circuitries involved in disease development. We also outline how evidence obtained from experimental mouse models of Kawasaki disease vasculitis has paved the way for the development of new efficient therapeutics to treat human Kawasaki disease.

Aetiological agents

The causative agents initiating the disease have still not been identified >50 years after the first description of Kawasaki disease. However, the trigger is suspected to be of viral origin and to enter the body through the

Key points

- Kawasaki disease is a childhood systemic vasculitis leading to the development of coronary artery aneurysms; it is the leading cause of acquired heart disease in children in developed countries.
- The cause of Kawasaki disease is unknown, although it is suspected to be triggered by an unidentified infectious pathogen in genetically predisposed children.
- Kawasaki disease might not be a normal immune response to an unusual environmental stimulus, but rather a genetically determined unusual and uncontrolled immune response to a common stimulus.
- Although the aetiological agent in humans is unknown, mouse models of Kawasaki disease vasculitis demonstrate similar pathological features and have substantially accelerated discoveries in the field.
- Genetic and transcriptomic analysis of blood samples from patients with Kawasaki disease and experimental evidence generated using mouse models have demonstrated the critical role of IL-1β in the pathogenesis of this disease and the therapeutic potential of targeting this pathway (currently under investigation in clinical trials).

mucosal surfaces in the lung[29] (FIG. 1). This hypothesis is supported by the seasonality of Kawasaki disease outbreaks, which is similar to that of other respiratory infections. In Japan, two seasonal peaks have been observed, one in winter and another in summer, whereas in the USA, the incidence peaks are observed during spring and winter[30]. Development of Kawasaki disease is age specific, with children from 6 months to 5 years of age at greatest risk[3,30,31], which suggests a protective maternal passive immunity against the causative agent from birth to 6 months of age and the importance of immune system maturation in children ≥6 years of age[29].

The clinical features of Kawasaki disease, such as high fever, skin rash and peeling, conjunctivitis and intense release of pro-inflammatory cytokines, are reminiscent of other infectious diseases such as staphylococcal and streptococcal toxic shock syndromes[32]. Some studies have shown that, compared with healthy control individuals, patients with Kawasaki disease have a skewed Vβ T cell repertoire and increased frequencies of circulating Vβ2$^+$ and Vβ8.1$^+$ T cells, leading to the early suggestion that a superantigen toxin might have a role in triggering Kawasaki disease[33–35]. However, similar results were not reproduced in later studies[36,37], leading to the more generalized hypothesis that the development of Kawasaki disease might be triggered by multiple conventional antigens.

Several early studies showed reduced prevalence of antibodies to the Epstein–Barr virus (EBV) capsid antigen in Japanese children with Kawasaki disease compared with age and sex-matched control patients[38–40], suggesting the involvement of an abnormal immune response to EBV in disease development. However, this difference in EBV antibody seropositivity could not be reproduced in other studies[41–43]. A human coronavirus was detected more frequently in respiratory secretions of patients with Kawasaki disease than in control individuals[44], although, again, other studies could not replicate this finding[45,46], indicating that the original association might have been coincidental. The possibility that a retrovirus is the triggering agent for Kawasaki disease has also been proposed, owing to detection of retrovirus-specific reverse transcriptase activity in the co-culture supernatant of peripheral blood mononuclear cells (PBMCs) from patients with Kawasaki disease but not controls[47,48]. However, this result could not be replicated in later studies[49–51]. A peptide recognized by antibodies produced during the acute phase of Kawasaki disease has been identified in 2020 (REF.[52]). Although the protein epitopes seem similar to hepaciviruses[53], further studies are required to determine the specific gene sequence from which this peptide emerges.

Altogether, the absence of consistent and reproducible studies pinpointing a specific aetiological agent suggests that Kawasaki disease is caused not by one but by multiple infectious agents. Acute Kawasaki disease is associated with infiltration of IgA$^+$ plasma cells in the respiratory tract, implying that the upper airways act as a portal of entry[25,26]. One suggestion is that the triggering agent might be an environmental toxin or antigen transported by wind currents[54]; however, this possibility cannot be rigorously assessed until precise identification of the aetiological agents is achieved[29].

SNPs influencing susceptibility

Although Kawasaki disease has been observed around the world and in multiple ethnic groups, geographical differences exist in incidence. The highest incidence is in Asian countries such as Korea and Japan, where it has increased over the past decades and is now 10–20 times more prevalent than in North America and Europe[30]. This increased susceptibility in Asian children, as well as in children with Asian ancestry living in North America[31], indicates that genetic components predispose to disease susceptibility. In Japan, siblings of children with Kawasaki disease are at increased risk of developing the disease[55]. Single nucleotide polymorphisms (SNPs) in multiple genes have been associated with increased susceptibility to Kawasaki disease (FIG. 1); however, mechanisms linking those SNPs with Kawasaki disease progression are not yet well understood and require more investigation.

Calcium signalling pathway. Inositol 1,4,5-trisphosphate 3-kinase C (ITPKC), a kinase that phosphorylates inositol 1,4,5-triphosphate (IP_3), is involved in many signalling processes in a wide array of cells. In T cells, IP_3 is released after T cell receptor stimulation, thus increasing levels of intracellular Ca^{2+} through IP_3 receptors expressed on the endoplasmic reticulum and leading to nuclear translocation of nuclear factor of activated T cells (NFAT), IL-2 production and T cell activation[56]. By blocking the interaction of IP_3 with its receptor, ITPKC negatively regulates T cell activation. A functional SNP in *ITPKC* has been associated with increased risk of coronary artery lesions in Taiwanese[57], Japanese and American patients with Kawasaki disease[58]. Mechanistically, this ITPKC polymorphism might directly contribute to T cell hyperactivity, and more importantly, it might promote NLRP3 inflammasome activation and increase production of IL-1β and IL-18 (REF.[59]). ORAI1 is a membrane-bound Ca^{2+} channel protein encoded by *ORAI1* that is involved in the Ca^{2+}–calcineurin–NFAT signalling pathway. Although no significant association between *ORAI1* polymorphisms and Kawasaki disease

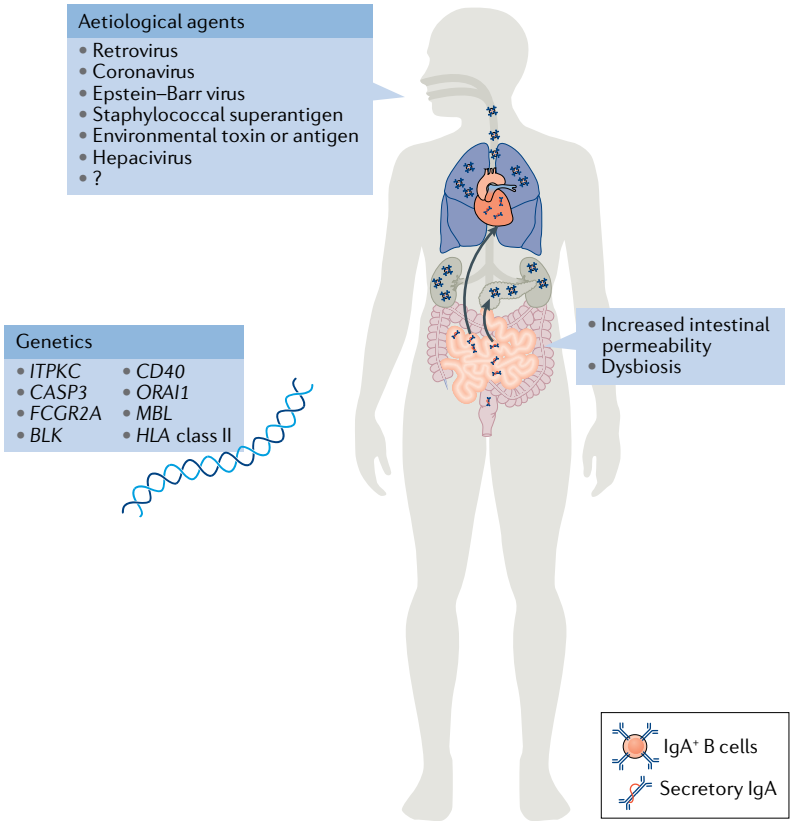

Fig. 1 | **Environmental and genetic factors implicated in the development of Kawasaki disease.** Different aetiological agents, from viruses to environmental toxins, have been proposed as triggering agents for Kawasaki disease; however, none has been corroborated, and the aetiological agent remains unidentified. Increased numbers of IgA^+ plasma cells have been detected in the pancreas, the kidneys, the coronary artery wall and the respiratory tract of patients with Kawasaki disease. Patients with Kawasaki disease have increased concentrations of secretory IgA in their serum, indicative of defective intestinal barrier function and increased intestinal permeability. Changes in the gut microbiota composition (dysbiosis) have also been suggested to have a role in the development of Kawasaki disease. Single nucleotide polymorphisms in the genes listed have been associated with susceptibility to Kawasaki disease and disease severity. The current understanding is that Kawasaki disease is triggered in genetically predisposed children by a ubiquitous environmental stimulus that typically would not result in an uncontrolled immune response and development of vasculitis.

susceptibility or IVIG treatment response was initially reported in the Taiwanese population[60], an SNP in exon 2 of *ORAI1* is associated with Kawasaki disease susceptibility in the Japanese population[61], and interestingly this SNP is 20 times more frequent in the general Japanese population than in the general European population[61]. Another SNP in *SLC8A1*, which encodes the Na^+–Ca^{2+} exchanger, is also associated with susceptibility to Kawasaki disease and aneurysm formation[62], further highlighting the critical role of calcium signalling pathways in development of Kawasaki disease. Crucially, the Ca^{2+}–NFAT signalling pathway is also key to intracellular Ca^{2+} regulation and therefore to NLRP3 inflammasome activation and IL-1β production[63,64].

CD40 ligand. CD40 ligand (CD40L) is a protein expressed by a large array of cells including activated T cells, B cells, monocytes and platelets. CD40L receptor, CD40, is expressed by antigen-presenting cells as well as endothelial cells[65]. CD40 engagement is associated with cell survival, activation, proliferation and cytokine production[65]. Compared with control patients with other febrile illnesses, patients with Kawasaki disease have increased CD40L expression on $CD4^+$ T cells and platelets, which correlates with increased development of coronary artery lesions and is reduced by IVIG treatment[66]. An SNP in *CD40L* has been reported in Japanese patients with Kawasaki disease and is more frequent in male patients with coronary artery lesions than in female patients[67]. This polymorphism was not observed in a cohort of Taiwanese patients[68]; however, another SNP in the *CD40* gene has been reported in an independent cohort of Taiwanese patients and is associated with increased susceptibility to Kawasaki disease and development of coronary artery lesions[69]. These results indicate a role of the CD40–CD40L pathway in the development and severity of Kawasaki disease and highlight this pathway as a potential therapeutic target.

Mannose-binding lectin. Mannose-binding lectin (MBL), a pattern recognition molecule of the innate immune system, binds the surface of pathogenic organisms and activates the complement pathway[70]. A polymorphism in *MBL2* was found to be an age-related risk factor for development of coronary artery lesions in a Dutch cohort of patients[71,72]. Another study in a cohort of Japanese patients with Kawasaki disease showed that codon 54 variants in *MBL2* are significantly associated with susceptibility to Kawasaki disease[73]. Interestingly, in the *Candida albicans* water-soluble fraction (CAWS) mouse model of Kawasaki disease vasculitis, MBL-A and MBL-C deposition are observed in the aortic root, suggesting involvement of the MBL-dependent lectin pathway in this experimental model[74]. However, further studies are required to understand the pathogenic roles of those two proteins as well as their potential as therapeutic targets.

Fcγ receptors. Polymorphisms in genes encoding the receptors for the Fc portion of immunoglobulins, Fcγ receptors (FcγRs), have been associated with the development of autoimmune and infectious diseases[75–77]. As Kawasaki disease is considered an infectious disorder, several studies have investigated the potential association of FcγR SNPs with Kawasaki disease susceptibility and the development of coronary artery lesions. In a cohort of Dutch patients, no difference in FcγR SNP distribution was observed between healthy individuals and patients with Kawasaki disease, and no association was noted between SNPs in FcγR genes and Kawasaki disease susceptibility[78]. However, a later study with >2,000 patients with Kawasaki disease and 9,000 control patients from multiple independent cohorts across different populations highlighted a Kawasaki disease-associated polymorphism in the *FCGR2A* locus, which encodes FcγRIIA (CD32a), a member of the family of IgG receptors[79]. This polymorphism has important implications as the standard of care for Kawasaki disease is IVIG, a pool of plasma IgG that interacts with FcγRs on immune cells. Interestingly, 15–20% of patients with Kawasaki disease have IVIG-resistant disease and

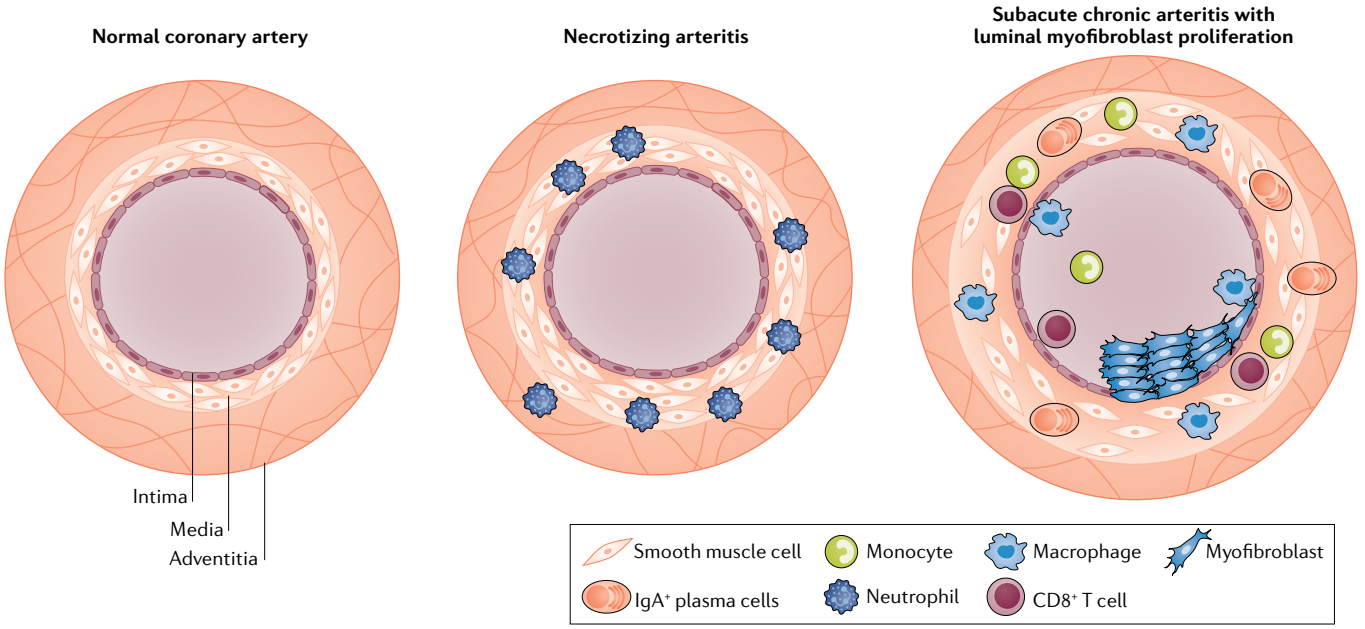

Fig. 2 | **Pathophysiology of Kawasaki disease vasculitis.** The normal coronary artery is composed of three general layers: the tunica intima, tunica media and tunica adventitia. The intima is mainly composed of endothelial cells, the media of smooth muscle cells and the adventitia of loose connective tissue. In Kawasaki disease, necrotizing arteritis develops in the first 2 weeks of the disease and is associated with neutrophilic infiltration, which gradually destroys the intima, media and some portions of the adventitia of the coronary artery. $CD8^+$ T cells, IgA^+ plasma cells, monocytes and macrophages compose the inflammatory infiltrate during subacute chronic arteritis. These cells release pro-inflammatory cytokines such as IL-1β and TNF, which contribute to luminal myofibroblast proliferation, in which myofibroblasts, mainly derived from smooth muscle cells, and their matrix products progressively obstruct the coronary lumen.

require another round of IVIG treatment or the use of adjunctive therapies[15,19,20,80]. The exact mechanisms by which IVIG mediates its therapeutic effect and how IVIG resistance develops remain unknown, and the potential involvement of this FcγRIIA polymorphism in IVIG resistance requires further investigation.

Pathophysiology of Kawasaki disease

The innate immune response. The immune response associated with Kawasaki disease is complex and involves the activation and infiltration of the coronary artery wall by both innate and adaptive immune cells (FIG. 2). On the basis of studies of post-mortem tissue from patients with Kawasaki disease, Kawasaki disease vascular pathology has been classified into three sequential linked pathological processes[81]. Necrotizing arteritis develops in the first 2 weeks of the disease and is associated with neutrophilic infiltrations, which gradually destroy the coronary artery intima, media and some portions of the adventitia. Alarmins from the S100 protein family, which are present in the cytoplasm of neutrophils, monocytes and macrophages[82], also participate in this inflammatory process. Concentrations of circulating S100A8/A9 heterodimers (calprotectin) and S100A12 are substantially higher in patients with Kawasaki disease during the acute phase than in control patients with other febrile illnesses and decline after IVIG treatment[83–85]. After the acute phase of Kawasaki disease, plasma concentrations of S100A8/A9 heterodimers only remain elevated in patients with giant CAAs[84], highlighting its potential utility as a biomarker to monitor long-term persistence of inflammation. S100A12 also contributes to the acute inflammatory response by directly stimulating monocytes to produce IL-1β, which in turn activates coronary endothelial cells[85]. Necrotizing arteritis might result in the formation of CAAs and is followed by two other processes, subacute or chronic vasculitis and luminal myofibroblast proliferation (LMP), which occur simultaneously and might be observed for months to years after disease onset[81]. The inflammatory infiltrates are composed of $CD8^+$ T cells, IgA^+ plasma cells, eosinophils and macrophages, which release pro-inflammatory cytokines contributing to cardiovascular pathology. Meanwhile, myofibroblasts, mainly derived from smooth muscle cells, and their matrix products progressively obstruct the coronary lumen[81] (FIG. 2). Persistent subacute and chronic vasculitis and LMP can lead to stenosis and thrombosis after acute illness[6,9].

Matrix metalloproteinases. Matrix metalloproteinases (MMPs; zinc-dependent endopeptidases that degrade extracellular matrix components) are known to have an important role in both inflammation and tissue remodelling processes[86]. Increased expression and activity of a diverse set of MMPs has been demonstrated in acute Kawasaki disease[87–89]. The expression levels of MMP3 and MMP9, both known to mediate vascular smooth muscle cell migration and neointimal formation[90], are increased in patients with Kawasaki disease[91], and the circulating levels of these MMPs correlate with the development of CAAs in these patients[92]. *MMP3* SNPs are also associated with the development of CAAs[88], and

this protease is considered to be a driving factor allowing IL-1-induced signalling to lead to migration of vascular smooth muscle cells and their transition to proliferating myofibroblasts[93–95]. Whereas MMP9 has been studied and implicated in elastin breakdown in the *Lactobacillus casei* cell wall extract (LCWE)-induced Kawasaki disease mouse model[96,97], information about the role of MMP3 in this mouse model is lacking.

MicroRNAs. MicroRNAs (miRNAs; a class of small non-coding RNAs that regulate mRNA expression) are emerging as critical gene regulators in a host of cellular processes, including inflammation[98]. Of human coding genes, 60–70% are estimated to be regulated by miRNAs[99]. Several studies attempting to discover Kawasaki disease biomarkers have found that the miRNA profiles of serum exosome or coronary artery tissues are associated with acute Kawasaki disease[100–104]. These miRNAs include miR-23a[100–103], miR-27b[100], miR-223 (REFS[100–103]) and miR-145 (REF.[103]). These miRNAs might provide clues as to the molecular mechanisms involved in the development of the cardiovascular lesions associated with Kawasaki disease. For example, miR-145 is highly expressed in vascular smooth muscle cells and has been reported to promote their switching to neointimal proliferating cells[105,106] and to regulate the transforming growth factor-β signalling pathway[103]. Increased levels of miR-23a contribute to cardiomyocyte apoptosis and may promote inflammatory responses by blocking macrophage autophagy activity[107,108]. However, improved understanding and characterization of the molecular and cellular mechanisms underlying the different roles of miRs during Kawasaki disease require further studies with animal models.

Myocarditis. Most attention in Kawasaki disease research and clinical practice has focused on the development of CAAs and long-term complications of coronary artery stenosis and ischaemia[109]. However, the subacute and chronic inflammation of Kawasaki disease is also associated with the development of myocarditis[3,6,110–112]. Myocarditis has been described as the 'hidden face of the moon' in Kawasaki disease[110]. Reports indicate that myocarditis occurs frequently during acute Kawasaki disease[111], and serial myocardial biopsy studies have documented that histological myocarditis develops in the majority of patients with Kawasaki disease, even in the absence of coronary aneurysms[113,114]. More recent data indicate that myocardial inflammation can be documented in 50–70% of patients using gallium citrate (^{67}Ga) scans and technetium-99 (^{99m}Tc)-labelled white blood cell scans[115]. Another study has shown that myocardial inflammatory changes and myocardial oedema in Kawasaki disease occur even before coronary artery abnormalities and without concurrent ischaemic damage[112].

Myocarditis in Kawasaki disease tends to develop early, and acute left ventricular dysfunction is generally transient and responds readily to anti-inflammatory treatment[116]. However, Kawasaki disease myocarditis might be associated with fatal arrhythmias in infants, and in certain cases might lead to long-term complications including myocardial fibrosis[81,117]. Therefore, myocarditis during Kawasaki disease and its potential consequences deserve serious investigation, and long-term studies into late adulthood are needed.

Complement and immune complexes. Kawasaki disease affects small and medium sized vessels, particularly the coronary arteries; however, dilatations and aneurysms can occur systemically, including in the axillary, subclavian, brachial, renal and iliac arteries as well as the abdominal aorta[23,118–120]. Post-mortem findings have revealed that 73% of patients with Kawasaki disease have renal artery involvement and acute kidney injury[121] involving glomerulonephritis with intracapillary changes and deposition of immune complex composed of IgA and complement component 3 (C3)[22,122,123]. These findings are comparable to those in two other human vasculitis diseases, IgA vasculitis (IgAV) and IgA nephropathy (IgAN), which are similarly characterized by IgA immune complexes with C3 deposition in kidney glomeruli (see below). Increased concentrations of circulating IgA and secretory IgA (sIgA) have been reported in the serum of children with Kawasaki disease during the acute phase[124]. IgA$^+$ plasma cells are present in the coronary artery wall and in non-vascular tissues, such as the kidney, trachea and pancreas of patients with Kawasaki disease[25,26]. This IgA response is oligoclonal, seems to be antigen driven and might be caused by Kawasaki disease-triggering agents[125,126].

The IL-1 signalling pathway. Evidence from mouse models of Kawasaki disease[11,127,128], as well as transcriptome analysis performed on whole blood of patients with Kawasaki disease during the acute or convalescent phase[129,130], demonstrate the involvement of innate immune cells and inflammasome overactivation throughout the acute phase of the disease. In vitro cultured PBMCs isolated from patients with Kawasaki disease spontaneously release IL-1β into the supernatant, and this process is substantially reduced after IVIG treatment[28]. Serum concentrations of both IL-1β and IL-18 are also higher in children with acute Kawasaki disease than in control patients with other febrile illnesses, and markedly decrease during the convalescent phase[59], supporting the concept of activation of the NLRP3 inflammasome complex. Similarly, IL-1 and NLRP3-related gene transcripts are upregulated in PBMCs from patients with acute Kawasaki disease and are decreased during the convalescent phase of the disease[59], and an *IL1B*-gene-related signature is associated with acute phase disease and IVIG resistance[130]. Furthermore, a study has shown that differential expression of IL-1β and related signalling genes might have a role in mediating the sex-based differences seen in patients with Kawasaki disease[131]. In the LCWE mouse model of Kawasaki disease, the activation of caspase 1, IL-1α and IL-1β is key to the development of coronary arteritis, aneurysms, myocarditis and abdominal aorta aneurysms[127,128,132]. IL-1 has the capacity to expand and promote the differentiation of antigen-specific CD8$^+$ T cells[133], and indeed the frequencies of circulating CD4$^+$ and CD8$^+$ T cells are increased in patients with Kawasaki

disease[134]. Infiltrations of mature dendritic cells as well as activated cytotoxic CD8$^+$ T cells have been reported in arterial layers of coronary aneurysms[24,135]. Therefore, blocking the NLRP3–IL-1β pathway seems to be a valid therapeutic option in Kawasaki disease.

Role of the gastrointestinal tract

Intestinal permeability. The intestinal barrier has a critical role in maintaining intestinal homeostasis and health by preventing harmful organisms and luminal antigens from entering the circulation. A dysfunctional intestinal barrier, characterized by increased intestinal permeability, is recognized as a pathogenic factor in many inflammatory diseases[136]. In Kawasaki disease, abdominal pain, diarrhoea and vomiting are often observed at the onset of acute illness, affecting up to 60% of diagnosed patients and indicating that the gastrointestinal tract is also affected[4,137–140]. A multicentre study of >300 patients revealed that gastrointestinal manifestations at onset of disease complicate diagnosis, delay adequate treatment and correlate with IVIG resistance and severity of CAAs[141]. Immunohistochemical studies have revealed higher numbers of activated CD4$^+$ T cells and macrophages along with lower numbers of CD8$^+$ T cells in the jejunum lamina propria in patients with Kawasaki disease than in control patients with diarrhoea from cows' milk protein intolerance[142]. However, these cellular abnormalities are specific to the acute phase of the disease and return to normal during the convalescent phase[142]. IgA$^+$ plasma cells have also been observed in a variety of different vascular and non-vascular tissues in patients with Kawasaki disease[26], and patients with Kawasaki disease also have increased concentrations of sIgA, which is produced at the intestinal mucosal surface, in their serum[124]. These studies indicate that the gastrointestinal tract is affected during Kawasaki disease and that mucosal immune activation might compensate and protect from defective intestinal barriers.

The role of gut-related immunity in the induction of inflammation in organ systems distant from the gut has been the subject of intensive investigation. We have observed increased intestinal permeability and a dysregulated intestinal immune response characterized by increased numbers of IgA$^+$ B cells in the Peyer's patches in the LCWE-induced mouse model of Kawasaki disease[143] (FIG. 3). In this model, the excessive IL-1β release associated with LCWE injection acts on intestinal epithelial cells to open tight junctions, and administration of IVIG or pharmacological agents that block intestinal permeability significantly reduces disease development[143]. Altogether, these observations link increased intestinal permeability and defective intestinal barrier function with systemic IL-1β release in Kawasaki disease.

The intestinal microbiome. Despite the strong connection between the intestinal microbiome and development of cardiovascular diseases[144,145], only a few studies have investigated the role of the intestinal microbiome during development of Kawasaki disease or treatment resistance. Microbiological culture-based methods demonstrated that, compared with healthy control individuals, patients with Kawasaki disease have a different intestinal microbiota composition characterized by a lower incidence of the *Lactobacillus* genus[146,147] and increased *Streptococcus* and *Staphylococcus*[148] species. Lactobacilli have been reported to prevent diarrhoeal disorders[149,150] and to improve intestinal barrier function by increasing the expression of intestinal tight junctions[151,152], enhancing the intestinal mucus layer[153] and modulating the intestinal microbiota composition[154]. Lactobacilli have also been shown to boost innate and immune functions against a variety of bacterial infections[155–157], and their disappearance during acute Kawasaki disease might lead to the blooming of other bacterial pathogens, which might further promote intestinal barrier dysfunction and inflammation. Intriguingly, a retrospective study of 364 patients with Kawasaki disease showed that children who received microbiome-altering antibiotics in the week before Kawasaki disease diagnosis were substantially more likely to have IVIG-resistant disease than those who did not receive antibiotics[158]. Antibiotics alter the abundance, taxonomic richness and diversity of the bacterial[159,160] as well as fungal[161] intestinal microbiome, and those alterations might persist from weeks to years after treatment discontinuation[159,160,162]. A longitudinal metagenomic study of faecal samples derived from patients with Kawasaki disease showed a marked increase of five *Streptococcus* spp. during the acute phase of Kawasaki disease[163]; however, all patients in that study were treated with antibiotics in the early stage of disease, therefore this observation might be reflective of antibiotic-induced dysbiosis and not Kawasaki disease itself. Nonetheless, how this intestinal dysbiosis occurs and how its effect on intestinal permeability affects the development of cardiovascular lesions during Kawasaki disease vasculitis remains unknown and under-appreciated.

Link with IgA vasculitis

IgAV, or Henoch-Schönlein purpura, is an IgA-mediated necrotizing vasculitis resulting in fibrinoid destruction of the affected small vessels. Renal involvement, characterized by IgA deposition in the kidney glomeruli, is also observed in IgAV[164]. IgAV nephritis is closely related to another glomerular disease, IgAN, wherein accumulation and deposition of IgA and IgA immune complexes in the kidney glomerular mesangium drive glomerular inflammation[165]. As IgA is mainly found at mucosal surfaces, a 'gut–kidney axis', influenced by a mix of genetic, microbial and dietary factors, has been suggested to be involved in the development of both IgAN[166] and IgAV in paediatric and adult patients[167]. We have demonstrated that the LCWE-induced mouse model of Kawasaki disease vasculitis is associated with the deposition of IgA and IgA–C3 immune complexes in vascular tissues, such as the inflamed coronary artery and abdominal aorta[143]. Deposited IgA and IgA–C3 immune complexes might result in overactivation of the immune cells present in the cardiovascular lesions and subsequent amplification of inflammation[143]. Substantial evidence indicates that immune complexes might promote vascular damage during human Kawasaki disease through the

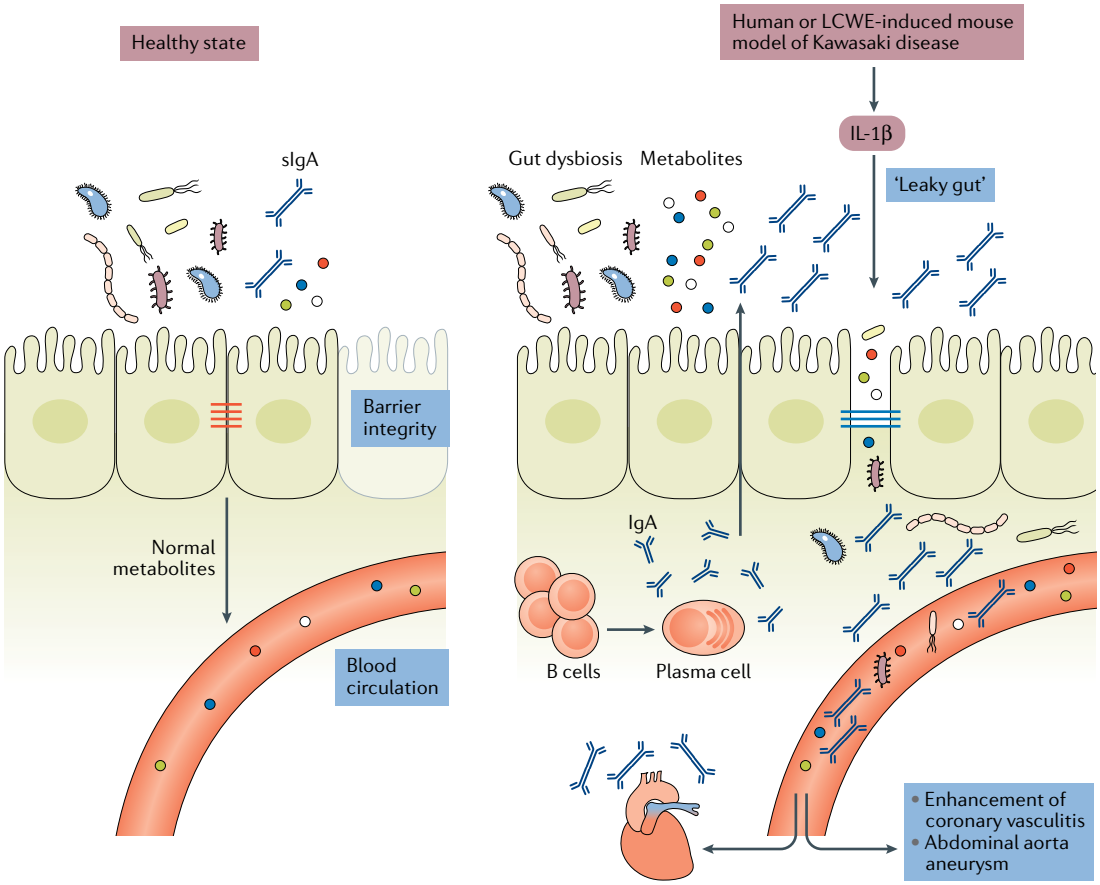

Fig. 3 | **Existence of a 'gut–vascular' axis in Kawasaki disease vasculitis.** In healthy individuals, intestinal epithelial cells are sealed together by intestinal tight junctions, and the intestinal epithelium acts as a barrier that prevents the passage of commensal bacteria and pathogens while permitting intercellular flux of ions, molecules and metabolites. *Lactobacillus casei* cell wall extract (LCWE)-induced Kawasaki disease vasculitis and human Kawasaki disease are associated with increased IL-1β production, which leads to decreased expression of intestinal tight junctions, resulting in increased intestinal permeability. Differences in intestinal microbiota composition have been observed in patients with Kawasaki disease, and intestinal dysbiosis might contribute further to the inflammatory process. LCWE injection is also associated with a dysregulated intestinal immune response characterized by increased numbers of IgA^+ B cells in the gastrointestinal tract and elevated secretory IgA (sIgA) concentrations. Intestinal barrier dysfunction results in sIgA leakage to the systemic circulation and pathogenic IgA–C3 immune complex deposition in the vascular tissues.

activation and aggregation of platelets, the release of vasoactive mediators, and the subsequent recruitment of neutrophils and leukocytes to the site of inflammation (reviewed elsewhere[168]).

Interestingly, we have also observed IgA and C3 deposition in the kidney glomeruli of LCWE-injected mice developing Kawasaki disease[143], and immune complex-mediated nephropathy has also been observed in Kawasaki disease[123]. However, to date IgA deposition has not been reported in CAAs of patients with Kawasaki disease. Given that availability of human tissue samples is limited, and those that are available are usually collected at the end stage of the disease, they might not be representative of active Kawasaki disease pathological features, and further studies are warranted. Like Kawasaki disease, IgAV develops mostly in children, affects males more than females, is more predominant in Asian countries such as Japan and Korea, and is also associated with abdominal pain, diarrhoea, skin rash and IgA deposition in the affected small vessels[169]. IgAN also shares pathological features with Kawasaki disease, such as increased intestinal permeability, low to moderate intestinal inflammation associated with activation of inflammatory cells in the small intestinal mucosa and colocalization of sIgA-complement in the glomerular mesangium[165,170]. Moreover, a polymorphism in the promoter of the lipopolysaccharide (LPS) receptor CD14 (CD14/159) is associated with coronary artery abnormalities in patients with Kawasaki disease[171] and has been linked to progression of IgAN to more severe renal disease[172]. IL-1β has a key pathogenic role during Kawasaki disease and also seems to be implicated in renal complications related to IgAV[173] and IgAN[174]. Altogether, given that Kawasaki disease shares clinical features and pathological mechanisms with both IgAV and IgAN, it is possible that Kawasaki disease is a form of IgAV. Similarly, treatments that have shown efficacy in Kawasaki disease, such as anakinra and IVIG, might be suitable and useful for treating IgAV[175] and IgAN.

Mouse models of Kawasaki disease

The lack of identification of specific aetiological agents and incomplete understanding of the molecular mechanisms involved in Kawasaki disease cardiovascular pathology have delayed the development of targeted and effective treatment options for this disease. In addition, the limited availability of tissue samples from patients with Kawasaki disease has considerably impeded progress in understanding the pathogenesis of the disease, making the availability of relevant animal models of Kawasaki disease extremely valuable. Kawasaki disease vasculitis can be induced in mice by injection of cell wall components from *L. casei*[176], *C. albicans*[177] or nucleotide-binding oligomerization domain containing 1 (Nod1) ligand[178] (TABLE 1). These mouse models of Kawasaki disease have accelerated research and have enhanced understanding of the pathogenesis of this disease. However, no animal model perfectly recapitulates human disease. Particularly in the context of Kawasaki disease, given that the aetiology remains unknown, researchers must exercise caution in interpreting results based on experimental models and confirm findings in patient cohorts. Nevertheless, even though the extrapolation of preclinical mouse data to humans is far from straightforward, mouse models are still invaluable tools to study certain pathological aspects of human inflammatory diseases and gain mechanistic insights.

The LCWE mouse model. *L. casei* is a Gram-positive bacteria that colonizes the gastrointestinal and urogenital tracts of both human and animals[179]. More than 35 years ago, Lehman et al.[180] demonstrated that a single intraperitoneal injection of LCWE induces a dose-dependent and chronic polyarthritis in rats. However, when injected into mice, LCWE induces instead a focal coronary arteritis[176]. How and which element of LCWE triggers Kawasaki disease vasculitis is unknown. LCWE is mainly composed of peptidoglycans, contains high levels of rhamnose and is resistant to lysozyme degradation[176].

The cardiovascular lesions induced in mice by LCWE are histologically similar to those observed in human disease. LCWE-induced Kawasaki disease vasculitis is characterized by infiltration of inflammatory cells in the aortic root, development of necrotizing arteritis in the coronary artery followed by luminal obstruction due to LMP that can lead to complete coronary artery stenosis[181], recapitulating the three pathological processes of human Kawasaki disease described above (FIG. 4a–d). In children with Kawasaki disease, thrombotic occlusion of the inflamed coronary artery leads to ischaemic heart disease[23,120], and similarly, occluding organizing thrombus in the coronary artery can be observed in LCWE-injected mice (FIG. 4e). Acute myocarditis and chronic scarring of the coronary arteries with the formation of stenotic fragments are also observed in LCWE-induced Kawasaki disease vasculitis (FIG. 4f), even long after the acute phase[182], which is similar to the fibrotic lesions that might lead children with Kawasaki disease to develop long-term cardiovascular sequelae in adulthood[8,9]. MRI and echocardiography in LCWE-injected mice demonstrate the presence of electrocardiographic changes (as observed in human Kawasaki disease) and myocardial dysfunction, which are responsive to anakinra therapy[183,184].

The LCWE-induced Kawasaki disease vasculitis in mice is dependent on intact TLR2 and MyD88 signalling and the subsequent release of pro-inflammatory cytokines, including IL-1β, IL-6 and TNF[10]. Genetic depletion of the TNF receptor or pharmacological blockade of the TNF signalling pathway (with infliximab (monoclonal antibodies to TNF) or etanercept (soluble TNF receptors)) protects mice from LCWE-induced Kawasaki disease vasculitis[132,185]. This model is also T cell dependent, as *Rag1*$^{-/-}$ mice develop fewer cardiovascular lesions[11]. CD8^{+} T cells are specifically required for LCWE-induced Kawasaki disease vasculitis as treatment of LCWE-injected mice with an anti-CD8-depleting antibody prevents the development of vasculitis[181]. This finding correlates with human disease, in which infiltrations of CD3^{+} T cells[135], and particularly CD8^{+} T cells, are detected in the CAAs[24]. The LCWE model has also confirmed the importance

Table 1 | **Comparison of the three mouse models of Kawasaki disease**

Characteristic	*Lactobacillus casei* cell wall extract	*Candida albicans* water-soluble fraction	Nod1 ligand (FK565)
Induction	Single intraperitoneal injection	Repeated intraperitoneal injections	Priming with LPS and Nod1 ligand intraperitoneal injection
Pathology	Aortic root inflammation; coronary arteritis; epicardial coronary arteritis; luminal myofibroblast proliferation; development of abdominal aorta aneurysms	Aortic root inflammation; coronary arteritis; inflammation focally extending to coronary arteries; development of abdominal aorta aneurysms	Aortic root inflammation; coronary arteritis
Immune characteristics	MyD88–TLR2-dependent; NLRP3 inflammasome-dependent; innate immune cell dependent (neutrophils and macrophages); T cell dependent	Dectin-2 receptor-dependent; increased antineutrophil cytoplasmic antibodies; innate immune cell dependent (neutrophils and macrophages); T cell dependent	CD11c^{+} macrophage-dependent; T cell-independent
Therapy	IVIG; anakinra; IL-1α antibody; IL-1β antibody; TNF antibody	IVIG; IL-1β antibody; GM-CSF antibody	NA

GM-CSF, granulocyte–macrophage colony-stimulating factor; IVIG, intravenous immunoglobulin; LPS, lipopolysaccharide; NA, not available.

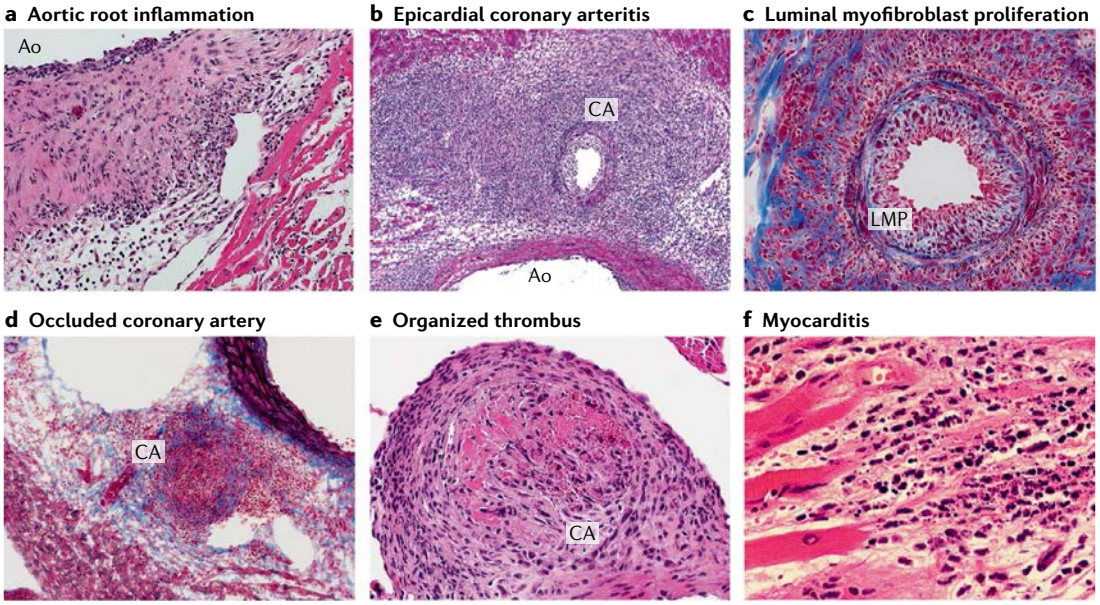

Fig. 4 | **Histological and morphological findings in the LCWE-induced mouse model of Kawasaki disease vasculitis.** Wild-type mice underwent intraperitoneal injection with *Lactobacillus casei* cell wall extract (LCWE), and heart tissues were harvested 2 weeks later. Haematoxylin and eosin (H&E) and trichrome staining were performed on heart sections. **a** | Inflammatory cell infiltration in the aortic route (H&E staining; ×40). **b** | Arteritis development in epicardial muscular coronary artery (H&E staining; ×20). **c** | Luminal myofibroblast proliferation (LMP) and non-specific neointimal proliferation injury to the arterial wall (trichrome staining; ×200). **d** | Complete occlusion of the coronary artery by LMP (trichrome staining; ×20). **e** | Organized thrombus in the coronary artery (H&E staining; ×200). **f** | Myocarditis (H&E staining; ×200). Ao; aorta, CA; coronary artery.

of the ITPKC pathway in Kawasaki disease development and demonstrated that ITPKC deficiency is associated with increased Ca^{2+} flux and levels of IL-1β in vitro[59]. Interestingly, the relatively mild development of coronary arteritis in LCWE-injected CBA/N mice — which are characterized by a defective B cell maturation process and poor humoral immune responses — suggests that the humoral immune response might participate in amplification of the disease[186]. IgA^+ plasma cells infiltrate vascular and non-vascular tissues during the acute phase of Kawasaki disease[25,26], resulting in the development of an oligoclonal IgA response in the coronary artery[125,126]. Interestingly, we have observed increased numbers of IgA^+ plasmablasts in the spleen, Peyer's patches and abdominal aorta draining lymph nodes of LCWE-injected mice, as well as increased concentrations of circulating IgA and IgA deposition in heart tissues, abdominal aorta and kidney glomureli[143].

Mouse models also provide a useful opportunity to evaluate the efficacy of therapeutic regimens on the development and healing of cardiovascular lesions. When given up to 5 days after LCWE injection, IVIG substantially decreases the severity of cardiovascular lesions in mice[187], mirroring the effects of IVIG treatment in humans. As described above, IL-1β signalling is higher in patients with Kawasaki disease than in age-matched control patients with other febrile illnesses[91,188], and studies using the LCWE model helped lead to the discovery of the importance of this pathway in the pathogenesis of the disease and the therapeutic potential of IL-1 blockade. Depletion of macrophages or blocking the IL-1 pathway either genetically using *IL1R*$^{-/-}$, *IL1α*$^{-/-}$ or *IL1β*$^{-/-}$ mice or with antibodies targeting IL-1α or IL-1β, or anakinra (IL1Ra), strongly reduces cardiovascular lesion development as well as myocardial dysfunction in LCWE-injected mice[128,132,184].

The CAWS mouse model. *C. albicans* is a harmless commensal fungus normally present in the human gastrointestinal tract that can transition into a pathogen capable of inducing inflammation in immune-impaired hosts. In 1979, Murata demonstrated that an alkaline extract made from *C. albicans* isolated from faeces from a patient with Kawasaki disease induced coronary arteritis in mice[177]. CAWS is composed of polysaccharides, mainly β-glucans and α-mannan proteins of the yeast cell wall[189], and needs to be injected intraperitoneally for five consecutive days in the first week of the disease to induce vasculitis in the aortic valves and the coronary arteries[189,190]. In this model, recognition of α-mannan proteins by the dectin-2 receptor seems to be essential, as CAWS-injected *Dectin-2*$^{-/-}$ mice do not develop vasculitis[191].

The CAWS model shares some histological similarities with human Kawasaki disease pathology in that inflammation affects both the aortic root and the proximal region of the coronary arteries[190]. Inflammation can also affect non-coronary artery sites in 25% of CAWS-injected mice and can be observed in the lymph nodes, the kidneys and the liver[190,192]. CAWS-induced coronary artery lesions resemble those of human Kawasaki disease and are typically proliferative, granulomatous and characterized by intimal thickening with destruction of the elastic lamina and media[190].

Echocardiography in CAWS-injected mice indicates a marked decrease of cardiac function, which can be restored by IL-10 supplementation[193]. IL-10 is a potent anti-inflammatory cytokine that might improve the outcome of CAWS-induced vasculitis by inhibiting the release of pro-inflammatory mediators, such as TNF and IL-1β, from tissue-infiltrating innate immune cells[194]. Interestingly, CAWS-induced Kawasaki disease vasculitis is also strain dependent, as CAWS injections lead to a high incidence of vasculitis in CD-1, C3H/HeN, DBA/2 and C57BL/6N mice, but the CBA/JN strain is resistant to coronary arteritis[190,195]. The DBA/2 strain is the most sensitive, with the highest mortality rate resulting from a more intense coronary arteritis[195]. The sensitivity of DBA/2 mice is associated with increased production of the pro-inflammatory cytokines TNF, IL-6 and IFNγ[195,196], whereas resistance of CBA/JN mice is explained by increased levels of IL-10 production in that strain[197].

Despite the presence of T cell and B cell infiltration in the inflamed coronary artery, mice lacking T cells still develop moderate to typical cardiac inflammation, indicating that T cells might not be required in the development of Kawasaki disease vasculitis in this particular model[198,199]. Absence of both T cells and B cells in *Rag1*$^{-/-}$ mice leads to lower incidence of CAWS-induced Kawasaki disease vasculitis; reconstitution of *Rag1*$^{-/-}$ mice with wild-type, but not *CCR2*$^{-/-}$, T cells and B cells restores cardiovascular lesions, suggesting roles for both T cells and B cells and the modulation of disease development by CCR2 expression[200]. The innate immune response also participates in vasculitis development; resident macrophages recognize the CAWS antigens through the dectin-2 receptor, leading to their activation, release of CCL2, and recruitment of neutrophils and inflammatory monocytes producing IL-1β in the aortic root[201].

CAWS-induced vasculitis is also associated with the rapid production of granulocyte–monocyte colony-stimulating factor in the heart, which subsequently drives inflammatory myocarditis by activating tissue macrophages and promoting recruitment of neutrophils and monocytes[199]. TNF is also produced during the acute phase of CAWS-induced Kawasaki disease vasculitis and is essential for the development of acute myocarditis, as TNF receptor-deficient mice are protected from the development of CAWS vasculitis[202]. IVIG administration substantially reduces CAWS-induced heart vessel inflammation[203]. Like the LCWE model, the CAWS model is also dependent on the IL-1 pathway, as *IL1R*$^{-/-}$, *IL1β*$^{-/-}$, *Asc*$^{-/-}$ and *Nlrp3*$^{-/-}$ mice are protected from induction of vasculitis, and treatment with anti-IL-1β agents substantially attenuates CAWS vasculitis[202,204,205].

The Nod1 ligand mouse model. Endothelial cells are equipped to sense microbial components through Toll-like receptors and nucleotide-binding oligomerization domain-containing protein like receptors. Subcutaneous injection or oral delivery of FK565, a specific synthetic Nod1 ligand, in mice primed with LPS results in a diffuse cellular inflammation of the aortic root and transmural infiltration of inflammatory cells in the coronary artery wall[178,206]. Other arteries, such as the iliac and renal arteries, also show signs of inflammation associated with a thickening of the intima[206].

The mechanisms by which FK565 induces coronary arteritis in mice remain unknown. When administered orally, FK565 does not induce intestinal mucosa inflammation, but specifically activates vascular cells to produce a diverse array of pro-inflammatory cytokines, including IL-1β[206], and chemokines such as CCL2, resulting in the recruitment of inflammatory cells in the tissues[178]. This model seems to be independent of T cells, B cells and natural killer T cells, as LPS-primed *Rag-1*$^{-/-}$ mice still develop aortitis and coronary arteritis after FK565 injection[207]. The inflammatory infiltrates observed around the inflamed aortic root and coronary arteries mainly comprise neutrophils and CD11c^{+} cardiac macrophages; their specific depletion considerably reduces the development of FK565-induced Kawasaki disease vasculitis[178,207]. The concentration of circulating IL-1β is substantially increased in the serum of FK565-injected mice compared with control or CAWS-injected animals, and higher IL-1β levels correlate with a larger inflammation area[206]. However, specific studies further investigating the role of IL-1β in this model are needed.

Treatment of Kawasaki disease

Traditional and novel therapies in humans. The current standard of care for Kawasaki disease is the use of high-dose IVIG together with aspirin. If given during the first 10 days of the disease, IVIG reduces the risk of development of coronary arteritis and aneurysms from about 30% to 5–7%[14,15]. The mechanisms by which IVIG treatment reduces the inflammatory responses are still unknown; however, IVIG is suspected to have a wide spectrum of action targeting multiple arms of the immune response[18]. IVIG has been shown to inhibit IL-1β production from in vitro stimulated macrophages and to stimulate the production of IL-1Ra[208,209]. During Kawasaki disease, IVIG reduces production of inflammatory cytokines and chemokines, and decreases the activation and number of circulating neutrophils, monocytes, macrophages and activated T cells by saturating Fc receptors[18]. The majority of patients with Kawasaki disease who are treated with IVIG improve and do not develop coronary artery damage; however, up to 20% of children with Kawasaki disease do not respond to treatment or have fever recurrence after initial IVIG treatment, and these patients are at the highest risk of developing coronary artery lesions[3,20,210].

The involvement of pro-inflammatory cytokines in the acute phase of Kawasaki disease suggests that combinational therapy, composed of IVIG associated with TNF inhibitors, steroids, calcineurin inhibitors or anakinra, might be useful to treat patients with IVIG-resistant disease. The use of TNF inhibitors in combination with IVIG has had mixed results thus far. Infliximab was associated with decreased fever duration and reduced markers of inflammation (C-reactive protein and neutrophil counts), suggesting a possible improvement of coronary artery outcomes[211]; however, etanercept treatment

resulted in a substantial reduction in IVIG resistance only in patients >1 year old[212].

An important area of research is the use of biomarkers to predict IVIG resistance in Kawasaki disease. The Kobayashi scoring system, based on a combination of laboratory test results (for example, C-reactive protein levels, neutrophil percentages, platelets counts and levels of aspartate and alanine aminotransferase) and demographic variables (sex, age and number of days of illness before the start of the treatment) has been successfully used to predict IVIG-resistance in Japanese patients[213], but not in North American children with Kawasaki disease[214]. The combination of prednisolone and IVIG to treat Japanese patients with Kawasaki disease predicted to have IVIG-resistant disease according to the Kobayashi score (RAISE study) resulted in more rapid fever resolution, reduced development of CAAs and lower incidence of additional rescue treatment[215] compared with IVIG alone.

As discussed above, Kawasaki disease susceptibility and increased coronary artery lesion risk are associated with an SNP in *ITPKC*[58] that results in a lack of NFAT regulation and activation of the T cell compartment owing to increased IL-2 production[216]. $CD8^+$ cytotoxic T cells are present in the inflamed arterial wall during Kawasaki disease[24,135]; therefore, targeting T cell expansion might be an efficient approach to preventing CAAs during Kawasaki disease. A combination treatment of IVIG and ciclosporin, a calcineurin inhibitor that suppresses IL-2 production and T cell activation, was tested in a clinical trial in Japanese patients with Kawasaki disease predicted to have IVIG-resistant disease based on the Kobayashi score (KAICA trial)[217]. In this trial, the combination treatment was shown to be safe and associated with a lower incidence of CAAs; however, treatment was linked with increased risk of relapse[217]. Furthermore, the scoring system used to identify IVIG-non-responders is poorly predictive in European children with Kawasaki disease, limiting the conclusions of this study.

The important role of the IL-1β–IL-1 receptor pathway in Kawasaki disease development has been demonstrated in both human patients[27,28,129,130] and mouse models[127,132,202,204]. Therefore, clinical trials investigating IL-1 pathway inhibition by using anakinra, which blocks both IL-1α and IL-1β, have been initiated in North America (ANAKID; ClinicalTrials.gov identifier NCT02179853)[218] and Europe (Kawakinra; European Clinical Trials number 2014-002715-4)[219]. Already, multiple case reports exist of the successful use of anakinra to treat patients with IVIG-resistant Kawasaki disease[220–224], indicating the promise of this second-line therapy.

Therapeutic insights from mouse models. Although no animal model can fully mimic human disease, the LCWE-induced Kawasaki disease mouse model has been accepted by many in the research community as a reliable experimental model providing novel insights that can be tested in patients. For example, IVIG efficiently prevents coronary arteritis development in LCWE-injected mice[187] as well as in the CAWS mouse model of Kawasaki disease[203].

The effects of the calcineurin inhibitors ciclosporin and tacrolimus have been investigated in the Nod1 ligand-induced mouse model of Kawasaki disease vasculitis[225]. This approach was rational given the established role of T cells and calcium signalling in Kawasaki disease. However, contrary to the expected outcome, these inhibitors exacerbated the coronary arteritis[225]. Notably, however, this result was probably related to the choice of mouse model, as the Nod1 ligand-mediated mouse model of Kawasaki disease vasculitis has previously been shown to be T cell-independent[207]. Indeed, in an independent study using the CAWS mouse model, which is T cell dependent, ciclosporin suppressed CAWS-induced vasculitis[226], emphasizing the importance of model selection in preclinical studies. Most importantly, results in human studies bear out the therapeutic potential of calcineurin inhibition, as the Japanese phase III trial (KAICA trial) showed that adding ciclosporin to IVIG in patients with Kawasaki disease who were at high risk of IVIG resistance was beneficial in diminishing overall incidence of CAAs[217].

The role of TNF has been investigated in both the LCWE and the CAWS mouse models of Kawasaki disease vasculitis[185]. Initially, etanercept treatment or genetic deletion of TNF receptor 1 was shown to protect mice from LCWE-induced coronary arteritis[185,202]. Infliximab treatment also prevented the development of both LCWE-induced coronary arteritis and myocarditis[132]. Similar results were obtained in the CAWS mouse model of Kawasaki disease vasculitis, in which etanercept[226,227] suppressed the incidence and decreased the severity of vasculitis. Mechanistically, TNF has been proposed to be produced by myeloid cells in the acute phase and to promote myocarditis and recruitment of immune cells by acting on cardiac stromal cells[202]. However, infliximab and etanercept might not directly target the TNF signalling pathway, and their observed effects might be indirect. Indeed, infliximab is not able to bind mouse TNF[227,228]; therefore, the anti-inflammatory effect of infliximab might be attributable to the binding of Fc receptors at the surface of activated cells[229,230].

The overwhelming evidence for the critical role of IL-1β in promoting LCWE-induced Kawasaki disease vasculitis in mice[127,128,132] led to the initiation of clinical trials testing the effect of anakinra for blocking IL-1β as a second therapy option to treat children with IVIG-resistant Kawasaki disease. Multiple case reports now outline the successful use of anakinra to treat patients with IVIG-resistant Kawasaki disease[221–224]. Alternatively, direct inhibition of the NLRP3 inflammasome might be a more targeted therapeutic strategy to treat Kawasaki disease, as it would affect several pathways beyond IL-1β, including IL-1α and IL-18. Several NLRP3 inhibitors have been identified[231] and tested in mouse models of inflammatory diseases, such as experimental autoimmune encephalomyelitis and cryopyrin-associated periodic syndrome[232]. It would be interesting to determine if such drugs could be used to prevent and reduce the cardiovascular complications in mouse models of Kawasaki disease vasculitis.

Conclusions

Over the past 40 years, research has improved our understanding of Kawasaki disease pathology and the development of coronary vasculitis. However, some questions still remain unanswered, such as the identification of the aetiological agents, how the disease is triggered, and the specific immune pathways associated with coronary vasculitis development and IVIG resistance. Owing to the rarity of human tissues from patients with Kawasaki disease, the use of animal models reproducing human Kawasaki disease features is invaluable. Many advances have been made over the decades by combining biological observations in human samples with mechanistic insights from experimental animal models. This 'bench to bedside' approach successfully led to the identification of the critical role of IL-1β in Kawasaki disease and resulted in the development of clinical trials in which anakinra is being used to treat children with IVIG-resistant Kawasaki disease.

LCWE-injected mice exhibit a dysfunctional intestinal barrier, and the increased IgA response and elevated sIgA levels in both LCWE-injected mice and children with Kawasaki disease reveal the existence of a 'gut–vascular' axis[143]. In evaluating this model system and the role of IgA, it should not be forgotten that injection of identically prepared LCWE induces chronic polyarthritis in selected inbred rat strains[180,233]. This observation implies that a common immunogenetic pathway might underlie a variety of autoimmune illnesses, with disease expression moderated not by the inducing agent, but rather by host genetics. The fact that cell wall fragments of common gut bacteria can produce varying disease manifestations in the face of inflammation-induced increased gut permeability suggests that some autoimmune diseases might not in fact be induced by the normal response to an unusual agent, but rather an unusual response to a common agent. Similarly, we hypothesize that vasculitic diseases, including Kawasaki disease, are not a usual response to an unusual environmental stimulus, but rather an unusual response (genetically determined) to a common environmental stimulus. This hypothesis has major implications for understanding the aetiology and pathogenesis of not only Kawasaki disease but also IgA-mediated diseases and perhaps others. In addition, it strongly suggests that inhibition of IL-1β might be effective for the many chronic inflammatory diseases in which IgA deposition is a key finding.

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