CLINICAL SCIENCE

Investigating changes in disease activity as a mediator of cardiovascular risk reduction with methotrexate use in rheumatoid arthritis

Tate M Johnson ,[1,2] Harlan R Sayles,[1,3] Joshua F Baker,[4,5] Michael D George,[4,5] Punyasha Roul,[1] Cheng Zheng,[3] Brian Sauer,[6,7] Katherine P Liao,[8] Daniel R Anderson,[9] Ted R Mikuls ,[1,2] Bryant R England [1,2]

Handling editor Josef S Smolen

► Additional online supplemental material is published online only. To view, please visit the journal online (http://dx.doi.org/10.1136/annrheumdis-2021-220125).

For numbered affiliations see end of article.

Correspondence to
Dr Bryant R England, Medicine & Research Service, VA Nebraska-Western Iowa Health Care System, Omaha, Nebraska, USA;
bryant.england@unmc.edu

Received 8 February 2021
Accepted 19 May 2021
Published Online First 28 May 2021

ABSTRACT

Objective Examine the association of methotrexate (MTX) use with cardiovascular disease (CVD) in rheumatoid arthritis (RA) using marginal structural models (MSM) and determine if CVD risk is mediated through modification of disease activity.

Methods We identified incident CVD events (coronary artery disease (CAD), stroke, heart failure (HF) hospitalisation, CVD death) within a multicentre, prospective cohort of US Veterans with RA. A 28-joint Disease Activity Score with C-reactive protein (DAS28-CRP) was collected at regular visits and medication exposures were determined by linking to pharmacy dispensing data. MSMs were used to estimate the treatment effect of MTX on risk of incident CVD, accounting for time-varying confounders between receiving MTX and CVD events. A mediation analysis was performed to estimate the indirect effects of methotrexate on CVD risk through modification of RA disease activity.

Results Among 2044 RA patients (90% male, mean age 63.9 years, baseline DAS28-CRP 3.6), there were 378 incident CVD events. Using MSM, MTX use was associated with a 24% reduced risk of composite CVD events (HR 0.76, 95% CI 0.58 to 0.99) including a 57% reduction in HF hospitalisations (HR 0.43, 95% CI 0.24 to 0.77). Individual associations with CAD, stroke and CVD death were not statistically significant. In mediation analyses, there was no evidence of indirect effects of MTX on CVD risk through disease activity modification (HR 1.03, 95% CI 0.80 to 1.32).

Conclusions MTX use in RA was associated with a reduced risk of CVD events, particularly HF-related hospitalisations. These associations were not mediated through reductions in RA disease activity, suggesting alternative MTX-related mechanisms may modify CVD risk in this population.

Key messages

What is already known about this subject?

- Several studies have demonstrated a reduced risk of atherothrombotic cardiovascular disease (CVD) with methotrexate (MTX) use, but its effect on heart failure (HF) and the mechanisms underlying its cardioprotective effect are poorly understood.

What does this study add?

- An evaluation of MTX treatment effect on CVD events, using novel marginal structural models accounting for the effect of time-varying confounders on the propensity to receive MTX, demonstrated a 24% reduction in composite CVD events and a 57% reduction in HF hospitalisation.
- Using mediation analysis, we observed that the effects of MTX use on CVD risk reduction are not mediated by reduction in rheumatoid arthritis (RA) disease activity.

How might this impact on clinical practice or future developments?

- These preliminary findings suggest that MTX-related mechanisms other than disease activity control may provide CVD, and particularly HF, benefit in patients with RA.

INTRODUCTION

Cardiovascular disease (CVD) is the most common cause of death in rheumatoid arthritis (RA)[1 2] owed largely to a heightened risk of coronary artery disease (CAD), stroke and heart failure (HF)[3–5] only partially explained by traditional CVD risk factors.[6] Controlling RA disease activity and select disease-modifying antirheumatic drugs (DMARDs) are speculated to provide cardioprotective benefits. Though methotrexate (MTX) did not reduce risk in non-RA patients with established CVD in the absence of systemic inflammation,[7] multiple observational studies have demonstrated benefit in patients with RA. MTX was associated with a 28% lower risk of CVD events in a meta-analysis of observational studies including over 200 000 patients with RA.[8] Recent cohort studies report 20%–60% reductions in CVD events with MTX treatment.[9–11] While associations with atherothrombotic CVD is more extensively studied, MTX has also been associated with a reduced risk of HF.[12 13]

The mechanisms by which MTX reduces CVD in RA remain poorly understood. Indirectly, MTX may lower CVD risk by reducing RA disease activity. Potential direct cardioprotective mechanisms of MTX include increased cholesterol efflux capacity, improved endothelial function, reduced foam cell generation and the ability to scavenge free radicals

To cite: Johnson TM, Sayles HR, Baker JF, *et al*. *Ann Rheum Dis* 2021;**80**:1385–1392.

implicated in CVD pathogenesis.[14–17] Improved understanding of the mechanisms by which MTX reduces CVD in RA may help clinicians optimally use MTX as therapeutic options expand.

In this study, we evaluated the association of MTX with incident CVD in a prospective cohort of US Veterans with RA to test the hypothesis that CVD risk reduction with MTX in RA is mediated through modification of RA disease activity.

PATIENTS AND METHODS

Study design

We conducted a cohort study in the Veterans Affairs Rheumatoid Arthritis (VARA) registry, a multicentre, prospective cohort of US Veterans with RA initiated in 2003.[18] Participants are >18 years of age and satisfy 1987 American College of Rheumatology (ACR) classification criteria for RA.[19] Participants signed informed consent for participation. Demographic data are obtained at enrolment while clinical data, including ACR Core Measures for RA disease activity, are collected at each visit. We followed patients from the latter of VARA enrolment or 1 April 2005 until incident CVD event, death or end of study period (1 April 2015).

DMARD exposure

Prescription fills for MTX and other conventional DMARDs (hydroxychloroquine, sulfasalazine, leflunomide), tumour necrosis factor (TNF) inhibitors (adalimumab, etanercept, infliximab, certolizumab, golimumab), non-TNF biologics (rituximab, abatacept, tocilizumab) and prednisone were identified from pharmacy dispensing records in the VA Corporate Data Warehouse (CDW).[20] Drug courses were defined from dispensing data using established algorithms.[21] Because our hypothesis focused on whether current disease activity (affected by prior MTX exposure) mediated future CVD risk, we used a 90-day lookback (180 days for rituximab) at each encounter to define drug exposure (online supplemental figure 1).

RA disease activity

Patient and provider global assessments of disease activity, swollen and tender joint counts, and C reactive protein (CRP) values were used to calculate time-varying 28-joint Disease Activity Scores (DAS28-CRP) at each VARA encounter. Missing disease activity components (joint counts 3.6%, global assessment 10.8% and acute phase reactant 5.1% of observations) were imputed using the last observation carried forward for subsequent missing data.

Covariates

Covariates were selected a priori and included age, sex, race, body mass index (BMI) categories, smoking status, comorbidities (including the Rheumatic Disease Comorbidity Index[22]), and use of aspirin, statins and non-steroidal anti-inflammatory drugs (NSAIDs). Demographics and smoking status were obtained at baseline. Time-varying BMI was obtained from the VA CDW, using the value closest to the visit date.[23] Comorbidities including hypertension, hyperlipidaemia, diabetes, CAD, cerebrovascular disease, HF, chronic kidney disease and chronic liver disease were defined by ≥2 inpatient or outpatient International Classification of Disease, Ninth Revision (ICD-9) codes prior to the index date (online supplemental table 1). Statin use was defined at baseline by the presence of at least one dispensing episode in the year prior to index. Aspirin and NSAIDs were defined at each visit using registry data as these agents are frequently obtained over the counter.

Outcomes

The primary outcome was a composite of incident (occurring after the index date) CAD (myocardial infarction (MI), percutaneous coronary intervention (PCI) or coronary artery bypass grafting (CABG)), stroke, HF hospitalisation or CVD-related death. Secondary outcomes included a composite of atherothrombotic major adverse cardiovascular events (MACE; CAD, stroke or CVD-related death) and individual CVD outcomes. CVD-related deaths were determined from the National Death Index (NDI), using ICD-10 codes I00–I99. Non-fatal events were identified using an administrative algorithm incorporating previously validated ICD-9 codes (online supplemental table 2).[24–30] We queried the VA CDW, requiring a primary or secondary hospital discharge diagnosis for CAD or stroke, including transient ischaemic attack. Since it is a chronic condition, HF was required to be a primary discharge diagnosis. Coronary revascularisation was identified using ICD-Procedure and Current Procedural Terminology codes for PCI and CABG. A random sample of CVD events was validated by medical record review using previously described adjudication criteria,[24 26] yielding a positive predictive value (PPV) of 90% for CAD and 83% for HF hospitalisations. The PPV for stroke was 70%; thus, all stroke events were validated by medical record review. Since acute events such as MI and stroke may result in treatment outside the VA, we identified outpatient CAD and stroke diagnoses in patients without a hospitalised event in the VA (online supplemental table 2). All events identified through this approach were validated by medical record review.

Statistical analysis

Baseline characteristics were stratified by ever-exposure or never-exposure to MTX during the study period. Crude incidence rates (IRs) and IR ratios (IRRs) for CVD events were estimated based on time-varying MTX exposure.

Marginal structural models (MSM) were used to evaluate the association of time-varying MTX use with CVD events. As time-varying covariates may dynamically affect the likelihood of receiving MTX, traditional analytical methods assessing the association of MTX with CVD events are at risk for substantial residual confounding.[31] MSM uses inverse probability weighting to account for the influence of time-fixed and time-varying confounders on the propensity to receive MTX, providing a more robust measure of causality between MTX and incident CVD risk.[31] MSM methods are described in detail in online supplemental appendix, following the approach described by Fewell *et al*.[32] Variables from current and prior visits were used to estimate the propensity to receive MTX at the current visit. Standardised mean differences (SMD) were calculated to ensure covariate balance.[33] In secondary analyses, we stratified patients by baseline DAS28-CRP (moderate/high vs remission/low) and high-sensitivity CRP (hs-CRP, with a previously used cut-off of 2 mg/L[34]), restricted analyses to those without a history of MACE or HF, and examined these associations in those with RA duration of <5 years.

Recognising that RA disease activity represents a potential confounder and/or mediator in the exposure–outcome pathway between MTX and CVD (figure 1), additional MSMs were used to estimate natural indirect effects through disease activity modification and direct effects of MTX exposure on CVD risk based on this causal structure.[35] Specific methods for this approach are detailed in online supplemental appendix. DAS28-CRP at the current visit served as the mediator between treatment with MTX and subsequent CVD events. Secondary analyses

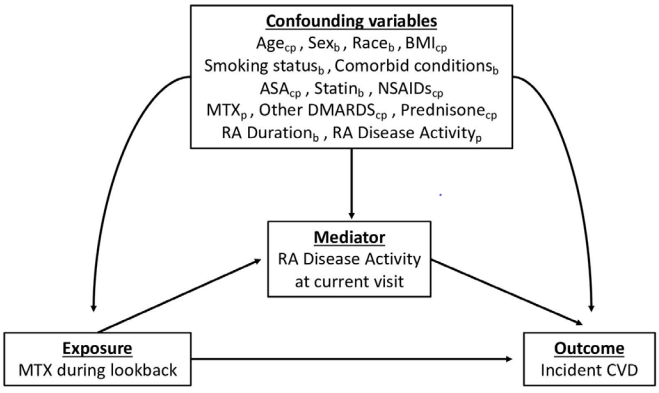

Figure 1 Directed acyclic graph modelling the causal framework between methotrexate (exposure) and incident cardiovascular disease (outcome). RA disease activity is both affected by MTX exposure and influences CVD risk, thus acts as the primary mediator of interest in this analysis. All confounding variables listed are assumed to influence the exposure, mediator and CVD outcome. RA disease activity measures included the DAS28-CRP as well as measures of systemic inflammation (erythrocyte sedimentation rate and CRP). ASA, aspirin; BMI, body mass index; CRP, C reactive protein; CVD, cardiovascular disease; DMARDs, disease modifying anti-rheumatic drugs; MTX, methotrexate; NSAIDs, non-steroidal anti-inflammatory drugs; RA, rheumatoid arthritis.

examined measures of systemic inflammation (erythrocyte sedimentation rate or CRP) as mediators between MTX exposure and CVD events. All analyses were completed using Stata V.15.1 software (StataCorp) within the VA Informatics and Computing Infrastructure.[20]

Patient and public involvement

Patients and the public were not directly involved in the design or completion of this study.

RESULTS

Baseline characteristics

We followed 2044 Veterans with RA over 10 360 patient-years (median follow-up of 4.9 years, mean of 11.4 visits per patient). Patients were predominantly male (90.0%), white (77.0%), in their seventh decade (mean age 63.9 years) and seropositive (79.2% rheumatoid factor (RF) positive, 77.8% anti-cyclic citrullinated peptide (anti-CCP) positive) with a mean DAS28-CRP of 3.6. Of 1090 patients on MTX at baseline, 561 discontinued MTX during the follow-up. Among patients not on MTX at baseline, 272 patients initiated MTX during the follow-up. Follow-up time was similar between patients treated with (5.0 years) or without (4.8 years) MTX at baseline. Demographics, BMI, smoking status and CVD-related comorbidities were similar by exposure group, with a modestly higher frequency of hyperlipidaemia in the MTX ever-exposed population (table 1). Chronic kidney disease and liver disease were more common in MTX unexposed patients, as was the use of leflunomide and sulfasalazine. Across all observations, biological DMARD and prednisone use was similar between MTX exposure groups, while other conventional DMARDs were more frequent in MTX unexposed patients (52.8% vs 36.9% of observations).

Table 1 Baseline characteristics of veterans with rheumatoid arthritis, stratified by methotrexate (MTX) ever-exposure (N=2044)

	MTX ever-exposed (N=1369)	MTX never-exposed (N=675)
Age (years)	63.8 (11.2)	64.1 (10.6)
Male sex, N (%)	1238 (90.4)	602 (89.2)
White race, N (%)	1046 (76.4)	527 (78.1)
BMI (kg/m^2)	28.7 (5.8)	28.1 (5.3)
Healthcare utilisation*		
Inpatient	0.2 (0.6)	0.2 (0.7)
Outpatient	16.8 (15.0)	17.8 (16.2)
High school graduate, N (%)	1116 (86.6)	536 (86.7)
Smoking status, N (%)		
Current	348 (25.5)	189 (28.1)
Former	719 (52.7)	363 (54.0)
Never	298 (21.8)	120 (17.9)
Comorbid conditions, N (%)		
RDCI	1.29 (1.31)	1.40 (1.39)
Hypertension	858 (62.7)	436 (64.6)
Hyperlipidaemia	682 (49.8)	315 (46.7)
Diabetes	301 (22.0)	154 (22.8)
Coronary artery disease	327 (23.9)	173 (25.6)
Cerebrovascular disease	91 (6.6)	53 (7.9)
Heart failure	109 (8.0)	63 (9.3)
Chronic kidney disease	66 (4.8)	55 (7.5)
Chronic liver disease	22 (1.6)	60 (8.9)
Lung disease	303 (22.1)	204 (30.2)
RA-related factors		
Rheumatoid factor positive, N (%)	1035 (78.2)	520 (81.1)
Anti-CCP positive, N (%)	1013 (76.7)	513 (80.2)
RA disease duration (years)	10.5 (11.1)	13.4 (11.6)
Tender joint count	5.5 (6.9)	4.3 (6.2)
Swollen joint count	4.5 (5.6)	3.6 (5.0)
Patient Global	41.2 (25.7)	39.5 (24.7)
Provider Global	35.0 (23.1)	32.9 (22.6)
ESR (mm/hr)	25.6 (22.6)	28.3 (24.4)
CRP (mg/dL)	1.5 (2.4)	1.6 (2.6)
hs-CRP (mg/L)	1.2 (2.0)	1.2 (1.9)
DAS28-CRP	3.7 (1.4)	3.4 (1.3)
MDHAQ	0.9 (0.6)	0.9 (0.6)
Medication use, N (%)		
Hydroxychloroquine	306 (25.9)	175 (22.4)
Sulfasalazine	106 (7.7)	92 (13.6)
Leflunomide	52 (3.8)	144 (21.3)
Prednisone	605 (44.2)	252 (37.3)
TNF inhibitor	343 (25.1)	210 (31.1)
Non-TNF biologic	23 (1.7)	15 (2.2)
Statins	599 (43.8)	290 (43.0)
Aspirin	85 (6.2)	41 (6.1)
NSAIDs	475 (34.7)	234 (34.7)

Baseline characteristics are stratified by whether a patient was ever exposed to MTX during the study period. Values mean (SD) unless otherwise noted.
*Number of visits in the 12 months prior to index date.
.BMI, body mass index; DAS28, disease activity score with 28-joint count; ESR, erythrocyte sedimentation rate; hs-CRP, high sensitivity C-reactive protein; MDHAQ, multidimensional health assessment questionnaire; MTX, methotrexate; NSAIDs, non-steroidal anti-inflammatory drugs; RA, rheumatoid arthritis; RDCI, Rheumatic Disease Comorbidity Index.

Incidence of CVD events

We identified 378 composite CVD events with an incidence of CAD, stroke, HF hospitalisation and CVD-related death of 14.2, 6.0, 7.9 and 15.0 events per 1000 person-years, respectively (table 2). Crude IRRs demonstrated a reduced incidence of HF hospitalisation (IRR 0.48, 95% CI 0.30 to 0.77) as well as composite CVD events (IRR 0.83, 95% CI 0.67 to 1.02) and CVD-related death (IRR 0.76, 95% CI 0.55 to 1.05) in MTX

Table 2 Incidence rates of cardiovascular events in veterans with rheumatoid arthritis (per 1000 person-years)

Event category*	N	Follow-up time (PY)	Mean time to event, years	Incidence rates (per 1000 PY)		IRR (95% CI)
				MTX ever-exposed	MTX never-exposed	
Composite	378	10 360	3.42	33.05	39.88	0.83 (0.67 to 1.02)
HF hospitalisation	87	10 956	3.47	5.17	10.66	**0.48 (0.30 to 0.77)**
MACE	342	10 443	3.45	30.32	35.14	0.86 (0.69 to 1.07)
CAD	151	10 605	3.08	13.50	14.97	0.90 (0.65 to 1.26)
Stroke	66	10 934	3.36	6.13	5.95	1.03 (0.62 to 1.72)
CVD Death	167	11 098	3.99	13.00	17.03	0.76 (0.55 to 1.05)

Values listed in bold indicate p<0.05.
*MACE is a composite of outcomes included in CAD, stroke and CVD death. CAD includes acute myocardial infarction, percutaneous coronary intervention or coronary artery bypass.
.CAD, coronary artery disease; CVD, cardiovascular disease; HF, heart failure; IRR, incidence rate ratio; MACE, major adverse cardiovascular event; MTX, methotrexate; PY, person years; RA, rheumatoid arthritis.

exposed patients in unadjusted analyses, although the latter two were not statistically significant. IRRs for MACE, CAD and stroke were not significantly different between MTX exposed and unexposed patients (table 2).

Mtx and CVD risk using MSM

In MSM including time-varying variables from the current and prior encounter, covariate balance was achieved between exposure groups (SMD <0.1 for all variables). MTX was associated with a reduced risk of composite CVD events (HR 0.76, 95% CI 0.58 to 0.99) and HF-related hospitalisation (HR 0.43, 95% CI 0.24 to 0.77). While MTX was associated with a numeric reduction in MACE (HR 0.82, 95% CI 0.63 to 1.06), CAD (HR 0.84, 95% CI 0.55 to 1.28), stroke (HR 0.72, 95% CI 0.34 to 1.53) and CVD-related death (HR 0.78, 95% CI 0.55 to 1.13), these differences were not statistically significant (table 3).

When stratified by baseline DAS28-CRP, associations between MTX and composite CVD events and MACE were generally numerically stronger among those with moderate to high DAS28-CRP at baseline, though 95% CIs were overlapping (online supplemental table 3). Minimal differential associations between MTX and CVD events were observed based on a hs-CRP cut-off of 2 g/dL at baseline (online supplemental table 3). When restricting analyses to those without a history of MACE or HF (online supplemental table 4) as well as RA duration <5 years (online supplemental table 5), similar estimates were observed, although this did not reach statistical significance for HF (HR 0.52, 95% CI 0.23 to 1.18). Sensitivity analysis including education, lung disease, and baseline MDHAQ were similar, but less precise (composite HR 0.79, 95% CI 0.59 to 1.05, HF hospitalisation 0.45, 95% CI 0.24 to 0.84) (online supplemental table 6).

Table 3 Association of methotrexate use with cardiovascular disease events in veterans with rheumatoid arthritis*

Event category†	HR (95% CI)	P value
Composite	**0.76 (0.58 to 0.99)**	**0.04**
HF hospitalisation	**0.43 (0.24 to 0.77)**	**0.005**
MACE	0.82 (0.63 to 1.06)	0.12
CAD	0.84 (0.55 to 1.28)	0.42
Stroke	0.72 (0.34 to 1.53)	0.39
CVD Death	0.79 (0.55 to 1.13)	0.19

Values listed in bold indicate p<0.05.
*Marginal structural models adjusting for time-varying age, BMI, RA disease activity, and medication use (prednisone, conventional and biological DMARDs, ASA and NSAIDs). Time-invariant variables assessed at baseline included sex, race, smoking status, RA duration, comorbidities and statin use. Primary analysis includes variables from the current and prior visit into the propensity to receive MTX.
†MACE is a composite of CAD, stroke and CVD death. CAD includes acute myocardial infarction, percutaneous coronary intervention or coronary artery bypass.
.ASA, aspirin; BMI, body mass index; CAD, coronary artery disease; CVD, cardiovascular disease; DMARD, disease-modifying anti-rheumatic drug; HF, heart failure; MACE, major adverse cardiovascular event; MTX, methotrexate; NSAIDs, non-steroidal anti-inflammatory drugs; RA, rheumatoid arthritis.

Mediation analysis of MTX and CVD risk

In mediation analyses modelling RA disease activity as the primary mediator between MTX exposure and CVD events, indirect effects of MTX on CVD events through modification of RA disease activity were not significant (range HR 0.87–1.06, all ps>0.05, figure 2). Direct effects of MTX were associated with a lower risk of HF hospitalisation (HR 0.41, 95% CI 0.18 to 0.92) and a numeric, but non-significant, reduction in composite CVD events (HR 0.74, 95% CI 0.51 to 1.07). Similarly, secondary analyses did not demonstrate that reductions in inflammatory markers mediated MTX-related CVD risk (range HR 0.91–1.48, figure 3).

DISCUSSION

In this multicentre, prospective cohort of US Veterans with RA, we observed a significantly reduced risk of CVD events associated with MTX use, independent of age, BMI, traditional CVD risk factors, RA disease activity and other RA therapies using state-of-the-art causal inference techniques. To our knowledge, this is among the first studies to demonstrate a greater risk reduction for HF hospitalisations with MTX use compared with CAD and stroke, supporting a potential cardioprotective role of MTX extending beyond atherothrombotic events. While the association of MTX with lower CVD risk in RA has been consistently reported,[3 4 6] whether this is driven primarily by reductions in RA disease activity has not been elucidated. Through mediation analysis, we observed that the cardioprotective effect of MTX was not attributable to modification of disease activity. This suggests that use of MTX may have additional direct cardiovascular benefits in patients with RA that are independent of its role in controlling clinical RA disease activity, though this requires further study.

We estimated that MTX exposed patients carried a 24% lower risk of CVD, similar to prior studies supporting a cardioprotective effect of MTX.[8 36] Non-significant trends suggesting a protective effect against MACE (HR 0.82, 95% CI 0.63 to 1.06) or the MACE components of CAD (HR 0.84, 95% CI 0.55 to 1.28) and stroke (HR 0.72, 95% CI 0.34 to 1.53) are similar

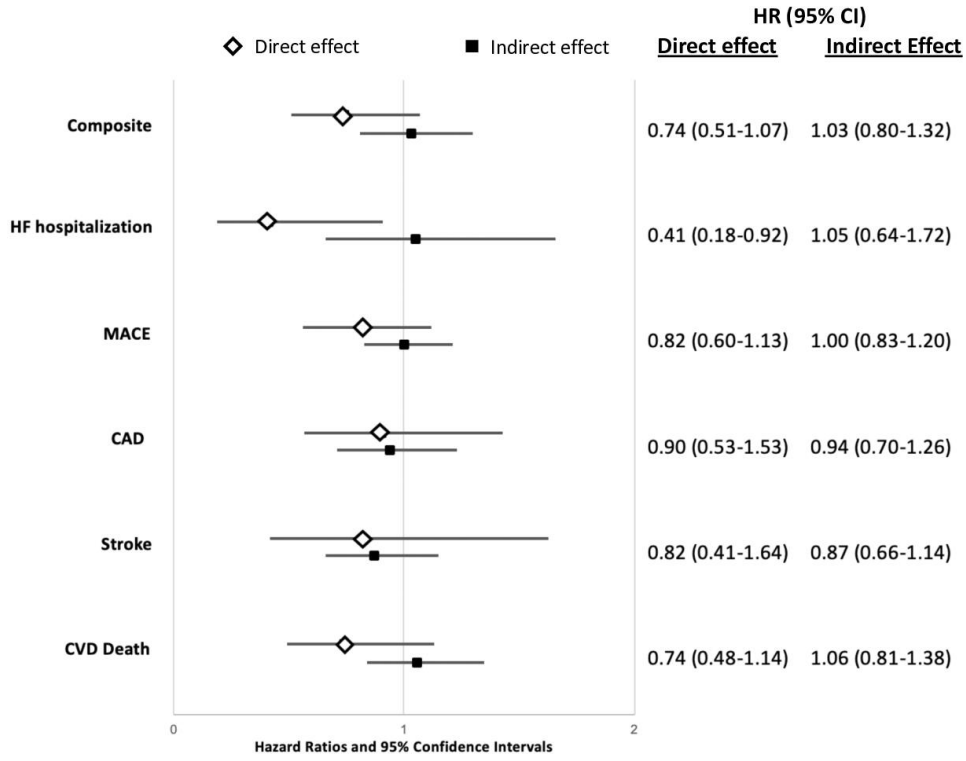

Figure 2 Forest plot illustrating the natural direct and natural indirect effects of MTX mediated by DAS28-CRP with CVD events using a counterfactual-based mediation analysis abbreviations: CAD, coronary artery disease; CVD, cardiovascular disease; DAS, Disease Activity Score; HF, heart failure; MACE, major adverse cardiovascular event; MTX, methotrexate.

to prior estimates.[8 36] While most prior studies use covariate adjustment in proportional hazards regression models, their adjustment for time-varying covariates that confound MTX use is limited. We used MSM, an approach that more accurately accounts for time-varying variables that influence the propensity to receive MTX and develop CVD events.[31] Importantly, this represents a novel methodological approach in addressing this high-impact clinical question.

Patients with RA are at a 20%–50% increased risk of HF referent to the general population, independent of ischaemic heart disease and suffer higher mortality rates after HF diagnosis.[5 37–42] Given the chronic nature of HF often complicated by recurrent hospitalisations, poorly controlled HF poses a substantial burden to patients and healthcare systems.[43] Effective prevention and management of HF in patients with RA could yield considerable benefits in this population. Initial case–control and cohort studies in RA demonstrated a 20%–50% reduction in HF risk associated with MTX use.[12 13 37] Our study builds on these findings, demonstrating a 57% lower risk of HF hospitalisation in those with MTX exposure in a large, multicentre, prospective RA cohort while accounting for longitudinal RA disease activity and other DMARD use. When restricting to those without HF at enrolment, a similar, but less precise, reduction in new-onset HF was seen (online supplemental table 4).

Mechanisms underpinning CVD risk in RA are multifactorial, including traditional CVD risk factors, RA disease activity, systemic inflammation, lipoprotein dysfunction, endothelial dysfunction and oxidative stress.[44] We demonstrated that indirect effects of MTX through modification of RA disease activity and systemic inflammation did not mediate CVD risk reduction in this cohort. Our findings suggest potential benefit of MTX in the context of CVD risk management beyond its important role of improving disease activity, particularly as it relates to reducing HF hospitalisations. Supporting this, a previous study of patients initiating biological DMARDs demonstrated a 24% lower risk of incident CVD in those on concomitant MTX.[45] Continued research is warranted to elucidate direct effects of MTX, such as improvements in lipoprotein and endothelial function,[14–17] on CVD outcomes in RA. Although not observed as an indirect effect from MTX in this study, evidence linking disease activity with CVD risk necessitates tight control of RA for lowering CVD risk.

The effect of antirheumatic drugs on CVD risk in non-RA patients is of increasing interest.[34 46] Whether MTX-related mechanisms might translate to effective CVD prevention outside of RA was investigated in the Cardiovascular Inflammation Reduction Trial.[7] In non-RA patients with prior CAD, low-dose MTX treatment did not reduce CVD events compared with placebo. These findings may relate to the inclusion of patients without high levels of systemic inflammation,[7] though in our study baseline hs-CRP minimally influenced the association between MTX and CVD events. RA likely portends a more complex inflammatory milieu than the non-RA population, thus, it remains unclear whether a specific inflammatory phenotype in the non-RA population might receive CVD benefit from MTX.

There are limitations to this study. This is an older, male-predominant cohort of US Veterans, which may limit generalisability of these findings. Sample size did not allow for a new-user, active-comparator design, thus changes in disease activity related to MTX may be underestimated and further study is required to specifically compare the observed CVD benefits of MTX, alone or in combination, with other DMARD regimens. As in any observational study, residual confounding is possible and manifests as a direct effect in mediation analyses, which must be considered when interpreting a causal link between MTX and CVD risk. While longitudinal measures allowed for robust

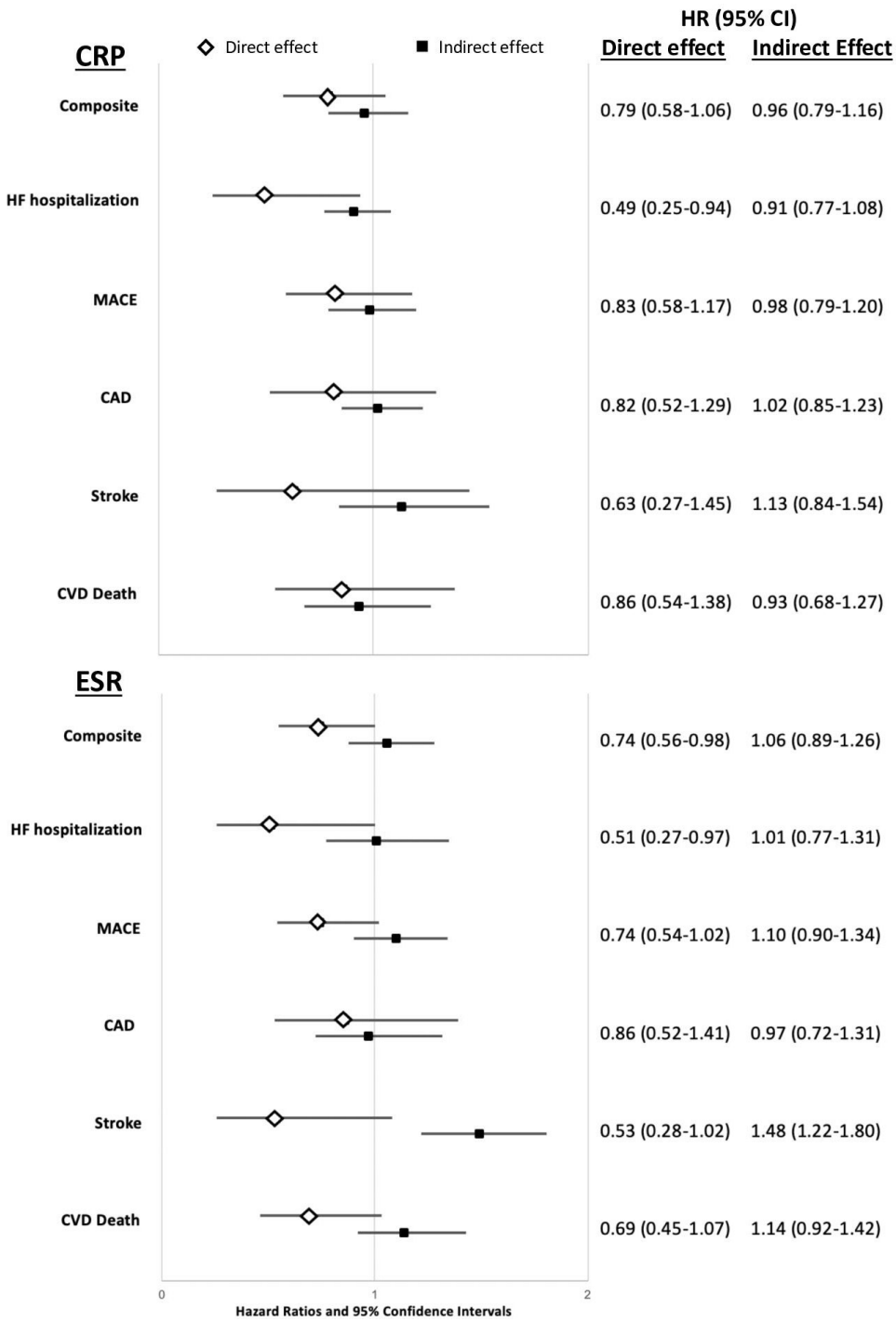

Figure 3 Forest plot illustrating the natural direct and natural indirect effects of MTX with cardiovascular disease events mediated by CRP (A) and ESR (B) using a counterfactual-based mediation analysis. CAD, coronary artery disease; CRP, C reactive protein; CVD, cardiovascular disease; ESR, erythrocyte sedimentation rate; HF, heart failure; MACE, major adverse cardiovascular event; MTX, methotrexate.

assessment of indirect effects of MTX through modification of disease activity, additional studies could explore the contribution of alternative mediators to CVD risk reduction with MTX use, such as reduced glucocorticoid dose. Additionally, folic acid supplementation may lead to modest reductions in atherothrombotic CVD,[47] although this has not been shown to affect HF risk. Though time-varying data were available for many variables, smoking status, statin use and comorbid conditions were available only at baseline. As this is an older population with a high degree of established comorbidity, the new incidence of traditional CVD comorbidities during follow-up has been shown to be relatively infrequent in this cohort.[48] Left truncation of our cohort limits the ability to account for accumulated RA disease activity and DMARD exposure prior to index, though including surrogate measures of prior RA disease activity including RA duration, DMARD (both conventional and biologic) and prednisone exposure, as well as MDHAQ did not substantially affect results. Because we relied on administrative data to identify CVD events, an event outside of the VA may be missed, though efforts were made to systematically minimise outcome misclassification by identifying new outpatient diagnoses and validating events through medical record review.

Causal inference methodology is rapidly evolving, offering means for more robust estimates of treatment effects and disease and/or treatment mechanisms in the observational setting. The use of propensity score-based methods is increasing, particularly in pharmacoepidemiological studies.[49] There remains underutilisation of MSM and causal mediation analyses in longitudinal observational studies of rheumatic diseases. Continued implementation of these methods in epidemiological studies is needed to improve the validity of study findings and to elucidate the mechanisms underlying observed associations.

In conclusion, MTX exposure was associated with a 24% reduced risk of CVD events including a 57% reduced risk of HF hospitalisation in a large RA registry linked to national VA data. These cardioprotective effects were not attributable to indirect effects through modification of RA disease activity or systemic inflammation. These data suggest MTX may offer CVD benefit, particularly in HF, beyond its role in reducing RA disease activity, and these roles should be especially considered in RA patients at high risk of CVD.

Author affiliations
[1]Medicine & Research Service, VA Nebraska-Western Iowa Health Care System, Omaha, Nebraska, USA
[2]Department of Internal Medicine, Division of Rheumatology, University of Nebraska Medical Center, Omaha, Nebraska, USA
[3]Department of Biostatistics, University of Nebraska Medical Center, Omaha, Nebraska, USA
[4]Department of Medicine, Division of Rheumatology, University of Pennsylvania Perelman School of Medicine, Philadelphia, Pennsylvania, USA
[5]Rheumatology, Corporal Michael J Crescenz VA Medical Center, Philadelphia, Pennsylvania, USA
[6]Rheumatology, VA Salt Lake City Health Care System, Salt Lake City, Utah, USA
[7]Department of Medicine, Division of Rheumatology, University of Utah Medical Center, Salt Lake City, Utah, USA
[8]Rheumatology, VA Boston Healthcare System, West Roxbury, Massachusetts, USA
[9]Department of Internal Medicine, Division of Cardiology, University of Nebraska Medical Center, Omaha, Nebraska, USA

Acknowledgements Work supported by Centre of Excellence for Suicide Prevention, Joint Department of Veterans Affairs and Department of DefenseDefence Mortality Data Repository—National Death Index

Contributors TMJ, TRM and BRE designed the study. TMJ, JFB, PR, BS, TRM and BRE were responsible for acquisition of data. TMJ, HRS, PR, CZ and BRE analysed the data. All authors were responsible for interpretation of the data and for drafting, revising and approving the final submitted manuscript.

Funding TMJ was supported by a Rheumatology Research Foundation Resident Research Preceptorship. TRM is supported by a VA Merit Award (BX004600) and grants from the Rheumatology Research Foundation and the National Institutes of Health: National Institute of General Medical Sciences (U54GM115458), National Institute on Alcohol Abuse and Alcoholism (R25AA020818) and National Institute of Arthritis and Musculoskeletal and Skin Diseases (2P50AR60772). BRE is supported by a Rheumatology Research Foundation Scientist Development Award and the VA (CX002203).

Disclaimer The views expressed in this article are those of the authors and do not necessarily reflect the position or policy of the Department of Veterans Affairs or the United States government.

Competing interests MDG receives grant support from Bristol Myers Squibb for unrelated work.

Patient consent for publication Not required.

Ethics approval Each individual site has institutional review board approval.

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement Data are available on reasonable request from the VA Rheumatoid Arthritis Registry and Biorepository after obtaining the required ethical approval.

ORCID iDs
Tate M Johnson http://orcid.org/0000-0003-0335-4157
Ted R Mikuls http://orcid.org/0000-0002-0897-2272
Bryant R England http://orcid.org/0000-0002-9649-3588

REFERENCES

1 England BR, Sayles H, Michaud K, *et al*. Cause-Specific mortality in male US veterans with rheumatoid arthritis. *Arthritis Care Res* 2016;68:36–45.
2 Sokka T, Abelson B, Pincus T. Mortality in rheumatoid arthritis: 2008 update. *Clin Exp Rheumatol* 2008;26:S35–61.
3 Aviña-Zubieta JA, Choi HK, Sadatsafavi M, *et al*. Risk of cardiovascular mortality in patients with rheumatoid arthritis: a meta-analysis of observational studies. *Arthritis Rheum* 2008;59:1690–7.
4 Avina-Zubieta JA, Thomas J, Sadatsafavi M, *et al*. Risk of incident cardiovascular events in patients with rheumatoid arthritis: a meta-analysis of observational studies. *Ann Rheum Dis* 2012;71:1524–9.
5 Løgstrup BB, Ellingsen T, Pedersen AB, *et al*. Cardiovascular risk and mortality in rheumatoid arthritis compared with diabetes mellitus and the general population. *Rheumatology* 2021;60:1400–9.
6 Solomon DH, Karlson EW, Rimm EB, *et al*. Cardiovascular morbidity and mortality in women diagnosed with rheumatoid arthritis. *Circulation* 2003;107:1303–7.
7 Ridker PM, Everett BM, Pradhan A, *et al*. Low-Dose methotrexate for the prevention of atherosclerotic events. *N Engl J Med* 2019;380:752–62.
8 Roubille C, Richer V, Starnino T, *et al*. The effects of tumour necrosis factor inhibitors, methotrexate, non-steroidal anti-inflammatory drugs and corticosteroids on cardiovascular events in rheumatoid arthritis, psoriasis and psoriatic arthritis: a systematic review and meta-analysis. *Ann Rheum Dis* 2015;74:480–9.
9 Ajeganova S, de Faire U, Jogestrand T, *et al*. Carotid atherosclerosis, disease measures, oxidized low-density lipoproteins, and atheroprotective natural antibodies for cardiovascular disease in early rheumatoid arthritis -- an inception cohort study. *J Rheumatol* 2012;39:1146–54.
10 Davis LA, Cannon GW, Pointer LF, *et al*. Cardiovascular events are not associated with MTHFR polymorphisms, but are associated with methotrexate use and traditional risk factors in US veterans with rheumatoid arthritis. *J Rheumatol* 2013;40:809–17.
11 Widdifield J, Abrahamowicz M, Paterson JM, *et al*. Associations between methotrexate use and the risk of cardiovascular events in patients with Elderly-onset rheumatoid arthritis. *J Rheumatol* 2019;46:467–74.
12 Bernatsky S, Hudson M, Suissa S. Anti-Rheumatic drug use and risk of hospitalization for congestive heart failure in rheumatoid arthritis. *Rheumatology* 2005;44:677–80.
13 Myasoedova E, Crowson CS, Nicola PJ, *et al*. The influence of rheumatoid arthritis disease characteristics on heart failure. *J Rheumatol* 2011;38:1601–6.
14 Reiss AB, Carsons SE, Anwar K, *et al*. Atheroprotective effects of methotrexate on reverse cholesterol transport proteins and foam cell transformation in human THP-1 monocyte/macrophages. *Arthritis Rheum* 2008;58:3675–83.
15 Ronda N, Greco D, Adorni MP, *et al*. Newly identified antiatherosclerotic activity of methotrexate and adalimumab: complementary effects on lipoprotein function and macrophage cholesterol metabolism. *Arthritis Rheumatol* 2015;67:1155–64.
16 Thornton CC, Al-Rashed F, Calay D, *et al*. Methotrexate-mediated activation of an AMPK-CREB-dependent pathway: a novel mechanism for vascular protection in chronic systemic inflammation. *Ann Rheum Dis* 2016;75:439–48.
17 Zimmerman MC, Clemens DL, Duryee MJ, *et al*. Direct antioxidant properties of methotrexate: inhibition of malondialdehyde-acetaldehyde-protein adduct formation and superoxide scavenging. *Redox Biol* 2017;13:588–93.
18 Mikuls TR, Reimold A, Kerr GS, *et al*. Insights and implications of the Va rheumatoid arthritis registry. *Fed Pract* 2015;32:24–9.
19 Arnett FC, Edworthy SM, Bloch DA, *et al*. The American rheumatism association 1987 revised criteria for the classification of rheumatoid arthritis. *Arthritis Rheum* 1988;31:315–24.
20 , 2014. Available: http://www.hsrd.research.va.gov/for_researchers/vinci/cdw.cfm [Accessed cited 2018 November 21].
21 Cannon GW, Mikuls TR, Hayden CL, *et al*. Merging Veterans Affairs rheumatoid arthritis registry and pharmacy data to assess methotrexate adherence and disease activity in clinical practice. *Arthritis Care Res* 2011;63:1680–90.
22 England BR, Sayles H, Mikuls TR, *et al*. Validation of the rheumatic disease comorbidity index. *Arthritis Care Res* 2015;67:865–72.
23 Baker JF, Billig E, Michaud K, *et al*. Weight loss, the obesity paradox, and the risk of death in rheumatoid arthritis. *Arthritis Rheumatol* 2015;67:1711–7.
24 Davis LA, Mann A, Cannon GW, *et al*. Validation of diagnostic and procedural codes for identification of acute cardiovascular events in US veterans with rheumatoid arthritis. *EGEMS* 2013;1:8.
25 Floyd JS, Blondon M, Moore KP, *et al*. Validation of methods for assessing cardiovascular disease using electronic health data in a cohort of veterans with diabetes. *Pharmacoepidemiol Drug Saf* 2016;25:467–71.

26 Grijalva CG, Chung CP, Stein CM, *et al*. Computerized definitions showed high positive predictive values for identifying hospitalizations for congestive heart failure and selected infections in Medicaid enrollees with rheumatoid arthritis. *Pharmacoepidemiol Drug Saf* 2008;17:890–5.
27 Huang H, Turner M, Raju S, *et al*. Identification of acute decompensated heart failure hospitalizations using administrative data. *Am J Cardiol* 2017;119:1791–6.
28 Kiyota Y, Schneeweiss S, Glynn RJ, *et al*. Accuracy of Medicare claims-based diagnosis of acute myocardial infarction: estimating positive predictive value on the basis of review of hospital records. *Am Heart J* 2004;148:99–104.
29 Lakshminarayan K, Larson JC, Virnig B, *et al*. Comparison of Medicare claims versus physician adjudication for identifying stroke outcomes in the women's health Initiative. *Stroke* 2014;45:815–21.
30 Sundbøll J, Adelborg K, Munch T, *et al*. Positive predictive value of cardiovascular diagnoses in the Danish national patient registry: a validation study. *BMJ Open* 2016;6:e012832.
31 Robins JM, Hernán MA, Brumback B. Marginal structural models and causal inference in epidemiology. *Epidemiology* 2000;11:550–60.
32 Fewell Z, Hernán MA, Wolfe F, *et al*. Controlling for time-dependent confounding using marginal structural models. *Stata J* 2004;4:402–20.
33 Austin PC, Grootendorst P, Anderson GM. A comparison of the ability of different propensity score models to balance measured variables between treated and untreated subjects: a Monte Carlo study. *Stat Med* 2007;26:734–53.
34 Ridker PM, Everett BM, Thuren T, *et al*. Antiinflammatory therapy with canakinumab for atherosclerotic disease. *N Engl J Med* 2017;377:1119–31.
35 Pearl J. Interpretation and identification of causal mediation. *Psychol Methods* 2014;19:459–81.
36 Micha R, Imamura F, Wyler von Ballmoos M, *et al*. Systematic review and meta-analysis of methotrexate use and risk of cardiovascular disease. *Am J Cardiol* 2011;108:1362–70.
37 Ahlers MJ, Lowery BD, Farber-Eger E, *et al*. Heart failure risk associated with rheumatoid Arthritis-Related chronic inflammation. *J Am Heart Assoc* 2020;9:e014661.
38 Crowson CS, Nicola PJ, Kremers HM, *et al*. How much of the increased incidence of heart failure in rheumatoid arthritis is attributable to traditional cardiovascular risk factors and ischemic heart disease? *Arthritis Rheum* 2005;52:3039–44.
39 Davis JM, Roger VL, Crowson CS, *et al*. The presentation and outcome of heart failure in patients with rheumatoid arthritis differs from that in the general population. *Arthritis Rheum* 2008;58:2603–11.
40 Khalid U, Egeberg A, Ahlehoff O, *et al*. Incident heart failure in patients with rheumatoid arthritis: a nationwide cohort study. *J Am Heart Assoc* 2018;7. doi:10.1161/JAHA.117.007227. [Epub ahead of print: 19 01 2018].
41 Mantel Ängla, Holmqvist M, Andersson DC, *et al*. Association Between Rheumatoid Arthritis and Risk of Ischemic and Nonischemic Heart Failure. *J Am Coll Cardiol* 2017;69:1275–85.
42 Nicola PJ, Maradit-Kremers H, Roger VL, *et al*. The risk of congestive heart failure in rheumatoid arthritis: a population-based study over 46 years. *Arthritis Rheum* 2005;52:412–20.
43 Heidenreich PA, Albert NM, Allen LA, *et al*. Forecasting the impact of heart failure in the United States: a policy statement from the American heart association. *Circ Heart Fail* 2013;6:606–19.
44 England BR, Thiele GM, Anderson DR, *et al*. Increased cardiovascular risk in rheumatoid arthritis: mechanisms and implications. *BMJ* 2018;361:k1036.
45 Xie F, Chen L, Yun H, *et al*. Benefits of methotrexate use on cardiovascular disease risk among rheumatoid arthritis patients initiating biologic disease-modifying antirheumatic drugs. *J Rheumatol* 2020. doi:10.3899/jrheum.191326. [Epub ahead of print: 15 Oct 2020].
46 Tardif J-C, Kouz S, Waters DD, *et al*. Efficacy and safety of low-dose colchicine after myocardial infarction. *N Engl J Med* 2019;381:2497–505.
47 Li Y, Huang T, Zheng Y, *et al*. Folic acid supplementation and the risk of cardiovascular diseases: a Meta-Analysis of randomized controlled trials. *J Am Heart Assoc* 2016;5.
48 Baker JF, England BR, George M, *et al*. Disease activity, cytokines, chemokines and the risk of incident diabetes in rheumatoid arthritis. *Ann Rheum Dis* 2021;80:566–72.
49 Johnson SR, Tomlinson GA, Hawker GA, *et al*. Propensity score methods for bias reduction in observational studies of treatment effect. *Rheum Dis Clin North Am* 2018;44:203–13.