Cureus
Part of SPRINGER NATURE

Open Access Review Article

Duration of Methotrexate Interruption and Vaccine Immunogenicity in Rheumatoid Arthritis: A Systematic Review of Hold Intervals Stratified by Vaccine Platform

Review began 10/21/2026
Review ended 11/15/2026
Published 11/18/2026

© Copyright 2026
Mahmoud Rezk et al. This is an open access article distributed under the terms of the Creative Commons Attribution License CC-BY 4.0., which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

DOI: 10.7759/cureus.119884

Mohamed Sobhy Mahmoud Rezk [1], Hager Musa Ahmed Ali [2], Ahmed Abdelateef Ahmed Abdelmageed [3], Mohamed Kamal Gomaa Abdelfattah [4], Mahmoud Abdelghany Eldyasty Ahmed Eid [5], Khadija Dafa Alla Osman [6], Ahmed Mohammed [7], Fatima Nadeem Salih [8], Hanady ME M Osman [9]

1. Rheumatology, Ministry of Health, Hail Health Cluster, King Khalid Hospital, Hail, SAU 2. General Practice, Abu Dhabi Health Services Company (SEHA), Abu Dhabi, ARE 3. Medicine, University Hospitals of Derby and Burton, Derby, GBR 4. Rheumatology, King Salman Specialist Hospital, Hail, SAU 5. Dermatology, Al-Azhar University Hospitals, Cairo, EGY 6. Internal Medicine, Specialized Rehabilitation Hospital, Abu Dhabi, ARE 7. Gastroenterology, St. Columcille's Hospital, Dublin, IRL 8. General Practice, Kasr Al-Ainy Educational Hospital, Cairo, EGY 9. Quality Improvement and Patient Safety, Ministry of Defense Health Services, Najran Armed Forces Hospital, Najran, SAU

Corresponding author: Ahmed Abdelateef Ahmed Abdelmageed, ahmedfrinds@hotmail.com

Abstract

Methotrexate (MTX) blunts humoral responses to several vaccines in rheumatoid arthritis (RA), and temporary interruption, hereafter termed a hold, is widely recommended, yet the optimal hold interval and whether it differs by vaccine platform remain uncertain. We systematically reviewed original peer-reviewed studies that compared an MTX hold of defined duration, or an MTX-free post-vaccination window, with continued MTX, or that compared hold durations, in adults with RA alone or as the predominant group within a mixed inflammatory arthritis cohort. PubMed/MEDLINE, Embase, Scopus, and Web of Science were searched from inception to September 30, 2026, following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses 2020 guidelines. Risk of bias was assessed with Risk-of-Bias Tool 2 for randomized trials and Risk of Bias in Non-randomized Studies - of Interventions for non-randomized studies, and certainty of evidence with the Grading of Recommendations, Assessment, Development, and Evaluation. Eleven studies (nine randomized, two observational cohorts) including 2,099 analyzed MTX-treated participants met the eligibility criteria, covering inactivated influenza vaccines, inactivated whole-virion, adenoviral-vector and mRNA SARS-CoV-2 vaccines, and pneumococcal conjugate vaccine. Across platforms, holding MTX for about two weeks after vaccination consistently improved humoral responses: an absolute gain in satisfactory influenza response of 21.0 percentage points (95% confidence interval (CI): 10.6-31.7), a 2.08-fold higher anti-spike receptor-binding domain (RBD) titer after mRNA-predominant boosters (95% CI: 1.59-2.70) that persisted to 26 weeks, and 2.2- to 2.6-fold higher median anti-RBD titers after ChAdOx1. In the one trial that tested it, holding MTX only before vaccination conferred no benefit, whereas holds spanning the post-vaccination window did, and observational data suggested a useful duration of about 10 days. A one-week hold was noninferior to two weeks for influenza (difference = -6.1 percentage points, 95% CI: -19.3 to 7.1; margin = -20) and carried fewer flares, but one-week data for other platforms are sparse. Repeating the hold after each dose of a two-dose series increased flares, whereas a single hold after the final dose preserved the immunologic gain without a significant excess of flares. Deferring MTX initiation by one month after pneumococcal conjugate vaccination improved responses in early RA without compromising one-year disease control. Certainty was moderate for influenza, mRNA, and pneumococcal conjugate vaccines; low for the adenoviral-vector vaccine; and very low for the inactivated whole-virion vaccine. A post-vaccination MTX hold of one to two weeks is supported with moderate certainty for influenza and mRNA vaccines, but with low or very low certainty for vector and whole-virion vaccines, with one week a reasonable option for influenza; trials testing one-week holds for other platforms and linking titers to clinical protection are needed.

Categories: Rheumatology
Keywords: covid-19 vaccine, drug holiday, immunogenicity, influenza vaccine, methotrexate, pneumococcal vaccine, rheumatoid arthritis, systematic review, vaccination

Introduction And Background

Rheumatoid arthritis (RA) is a chronic, systemic, immune-mediated inflammatory disease that causes synovitis, progressive joint destruction, disability, and excess mortality when inadequately controlled [1]. Low-dose weekly methotrexate (MTX) is the anchor conventional synthetic disease-modifying antirheumatic drug and is recommended as part of the first treatment strategy for nearly all patients with active RA, either alone or combined with biological or targeted synthetic agents [1]. Patients with RA carry a substantially higher burden of infection than the general population, including serious respiratory tract infections, a risk that reflects both disease-related immune dysregulation and treatment-induced

How to cite this article
Mahmoud Rezk M, Ahmed Ali H, Ahmed Abdelmageed A, et al. (November 18, 2026) Duration of Methotrexate Interruption and Vaccine Immunogenicity in Rheumatoid Arthritis: A Systematic Review of Hold Intervals Stratified by Vaccine Platform. Cureus 18(11): e119884. DOI 10.7759/cureus.119884

immunosuppression [2,3]. Vaccination against influenza, *Streptococcus pneumoniae*, SARS-CoV-2, and herpes zoster is therefore a central element of preventive care in RA, and the immunogenicity of these vaccines in MTX-treated patients is of considerable practical importance.

MTX exerts its anti-inflammatory effects through several mechanisms, including inhibition of purine and pyrimidine synthesis, promotion of extracellular adenosine release, interference with transmethylation reactions, and modulation of nuclear factor-κB and Janus kinase-signal transducer and activator of transcription (JAK-STAT) signaling [4]. These same pathways are required for the clonal expansion of antigen-specific B and T lymphocytes and the germinal-center reactions that follow vaccination. Consistent with this biology, a meta-analysis found that MTX reduces humoral responses to pneumococcal vaccines and may impair responses to influenza vaccines in RA [5]. During the COVID-19 pandemic, MTX emerged as one of the agents most strongly associated with attenuated humoral and cellular responses to mRNA vaccines, with adequate antibody responses in only 62% of MTX-treated patients with immune-mediated inflammatory disease compared with more than 90% of healthy controls and of patients on anticytokine biologics [6]. In practical terms, more than one in three MTX-treated patients failed to mount an adequate antibody response, compared with fewer than one in ten controls, a gap of a magnitude that could plausibly erode protection [6].

Because MTX is administered weekly and acts on rapidly proliferating lymphocytes, a brief interruption, hereafter termed a hold and defined as the temporary omission of scheduled MTX doses, timed to the period of antigen-driven lymphocyte expansion, has been proposed as a way to restore vaccine responsiveness without sacrificing long-term disease control. The 2022 American College of Rheumatology (ACR) guideline accordingly conditionally recommends holding MTX for two weeks after influenza vaccination, provided disease activity allows, but advises continuing MTX around other non-live vaccines, including pneumococcal and recombinant zoster vaccines, a position that reflects the limited direct evidence for those vaccine types [7]. The duration of the hold is the critical, unresolved variable: a longer hold may yield more immunogenicity but increases the risk of RA flare, and repeated holds are required for multi-dose vaccine series. Moreover, vaccine platforms differ fundamentally in antigen persistence and in the kinetics of the immune responses they induce, so a hold interval validated for a single-dose inactivated influenza vaccine cannot be assumed to be optimal for mRNA, adenoviral-vector, whole-virion inactivated, or polysaccharide-conjugate vaccines. Existing syntheses have quantified the overall effect of MTX on influenza and pneumococcal vaccine responses [5] or pooled COVID-19 hold trials into a single standardized estimate [8], but none has compared hold durations and hold timing across vaccine platforms while weighing the immunologic gain against the risk of flare. This systematic review therefore aimed to synthesize original evidence on the duration and timing of MTX holds in relation to vaccine immunogenicity and disease activity in patients with RA, stratified by vaccine platform.

Review

Methodology

Protocol and Reporting

This systematic review was conducted and reported in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 statement [9]. The review question was structured using the PICO (Population, Intervention, Comparator, Outcome) framework, which formalizes a clinical question into the four elements that determine eligibility and guide the search [10]. No review protocol was prospectively registered. The review question, eligibility criteria, outcomes, risk-of-bias tools, and the plan for a narrative synthesis stratified by vaccine platform were defined a priori and are reported in full in this section. Because there is no registered protocol against which deviations could be verified, this is acknowledged as a limitation; the handling of multiple reports of one trial, and of one report describing two trials, is described under Study Selection.

Eligibility Criteria

Studies were eligible if they enrolled adults (≥18 years) with RA receiving a stable dose of MTX or about to initiate it (P); applied a temporary discontinuation of MTX of a defined duration in relation to vaccination, or a defined MTX-free interval created by deferring initiation until after vaccination (I); compared this with uninterrupted MTX, immediate initiation, or an alternative hold duration or schedule (C); and reported vaccine immunogenicity as seroconversion, seroprotection, satisfactory vaccine response, geometric mean titers or ratios, neutralizing activity, or serotype-specific responder rates, with disease flare, disease activity, cellular responses, and adverse events as secondary outcomes (O). Randomized controlled trials, non-randomized interventional studies, and observational cohort studies were eligible, and studies of mixed inflammatory or autoimmune rheumatic disease cohorts were retained when RA predominated and platform-specific hold results were reported, as excluding them would have omitted the principal randomized evidence on mRNA platforms. Reviews, meta-analyses, conference abstracts, editorials, commentaries, letters without original data, protocols, preprints, case reports, and animal or in vitro work were excluded, as were studies confined to live-attenuated vaccines, studies of other immunosuppressants without MTX-specific hold data, studies in which the hold duration could not be determined, and articles

not available in English.

For consistency, the term hold is used throughout for any temporary omission of scheduled MTX doses in relation to vaccination, whatever term the source study used (interruption, discontinuation, withdrawal, pause, or suspension). Flare, defined by study-specific criteria as an episode of disease worsening, and disease activity, measured with validated composite indices at fixed time points, were treated as distinct secondary outcomes.

Information Sources and Search Strategy

PubMed/MEDLINE, Embase, Scopus, and Web of Science were searched from inception to September 30, 2026. The search strings for all databases are provided in the Appendices. Reference lists of included articles and of relevant reviews and guidelines were hand-searched, and ClinicalTrials.gov (https://ClinicalTrials.gov) and the Clinical Trials Registry-India were screened to identify trials whose results had subsequently been published in peer-reviewed form. No date or design filters were applied at the search stage.

Study Selection

Records were exported to a reference manager and deduplicated. Titles and abstracts were screened against the eligibility criteria, and the full texts of potentially eligible records were then assessed, with reasons recorded for every exclusion at full-text stage. Screening, full-text assessment, data extraction, and risk-of-bias assessment were each performed by a single reviewer. Where a trial had generated more than one peer-reviewed primary report, such as an interim and a final analysis of the same randomized cohort, the reports were linked to a single study so that participants were not counted twice, and the most complete report was used for data extraction; accordingly, all VROOM (vaccine response on, off MTX) estimates were taken from the final analysis, and interim results are cited for comparison only.

Data Extraction

A standardized, piloted extraction form captured first author, year, country, design and registration number, analyzed sample size, population and disease activity criteria, mean MTX dose, vaccine type and platform, number of doses and dosing interval, hold duration and its timing relative to vaccination, comparator, immunogenicity outcome definitions and serology timing, flare and disease activity outcomes, and effect estimates. Extracted values were checked against the source articles before synthesis. Effect estimates and their 95% confidence intervals (CIs) were extracted as published. Where a study reported group-level counts without a CI, the absolute risk difference and the risk ratio were calculated with 95% CIs using the Newcombe hybrid score method and the log method, respectively. Where only medians were reported, ratios of medians were calculated for descriptive comparison only, because CIs cannot be derived from medians and interquartile ranges. For noninferiority comparisons, the prespecified margin was extracted together with the CI of the between-group difference.

Risk-of-Bias Assessment

Randomized studies were appraised with the revised Cochrane risk-of-bias tool for randomized trials (RoB 2) across its five domains: the randomization process, deviations from intended interventions, missing outcome data, measurement of the outcome, and selection of the reported result [11]. Non-randomized studies were appraised with the Risk of Bias in Non-randomized Studies of Interventions (ROBINS-I) tool across its seven domains: confounding, selection of participants, classification of interventions, deviations from intended interventions, missing data, measurement of outcomes, and selection of the reported result [12]. Because participants and treating clinicians could not be blinded to a hold in any interventional study, masking of outcome assessors and laboratory personnel was recorded for each trial. For the observational cohorts, age, disease activity, concomitant immunosuppression, prior SARS-CoV-2 infection, and vaccine type were considered the principal potential confounders of the association between holding MTX and the antibody response. Domain-level and overall judgements are presented as traffic-light plots.

Data Synthesis

Studies were grouped by vaccine platform (inactivated split-virion influenza; inactivated whole-virion SARS-CoV-2; adenoviral-vector SARS-CoV-2; mRNA SARS-CoV-2; pneumococcal conjugate) and, within platform, by hold duration and timing. Findings were tabulated and summarized narratively, with effect estimates reported as published. Ratios of the reported response measure in the hold arm to that in the comparator arm were displayed for each study arm to convey direction and approximate magnitude; because these ratios refer to different response measures, they are study-specific and descriptive and were neither pooled nor compared formally across studies. No meta-analysis was performed because of heterogeneity in assays, outcome definitions, hold schedules, and populations; accordingly, no assessment of publication bias by funnel plot was undertaken. Certainty in the evidence for each platform was rated using the Grading of Recommendations Assessment, Development and Evaluation (GRADE) approach [13], considering risk of

bias, inconsistency, indirectness, imprecision, and publication bias, with randomized evidence starting at high certainty and observational evidence at low certainty. Because no effect estimates were pooled, ratings were applied to the body of evidence for each platform rather than to a summary estimate.

Results

Study Selection

The searches returned 321 records (PubMed/MEDLINE 99, Embase 72, Scopus 57, Web of Science 93), of which 122 were duplicates. Of the 199 records screened, 163 were excluded at title and abstract, leaving 36 reports that were all retrieved and assessed in full. In total, 25 were excluded: seven did not report the duration of the MTX hold, seven enrolled populations not meeting RA diagnostic criteria, and 11 were reviews, meta-analyses, or cohort reports that did not meet the eligibility criteria. Citation searching and registry screening identified no additional eligible reports. Overall, 11 reports describing 11 studies were included (Figure *1*) [14-24]. Two of these reports were an interim and a final analysis of the same randomized trial and were treated as a single study [17,18], and one report described two independently randomized parallel trials, MIVAC I and MIVAC II, which are counted and presented separately throughout [16].

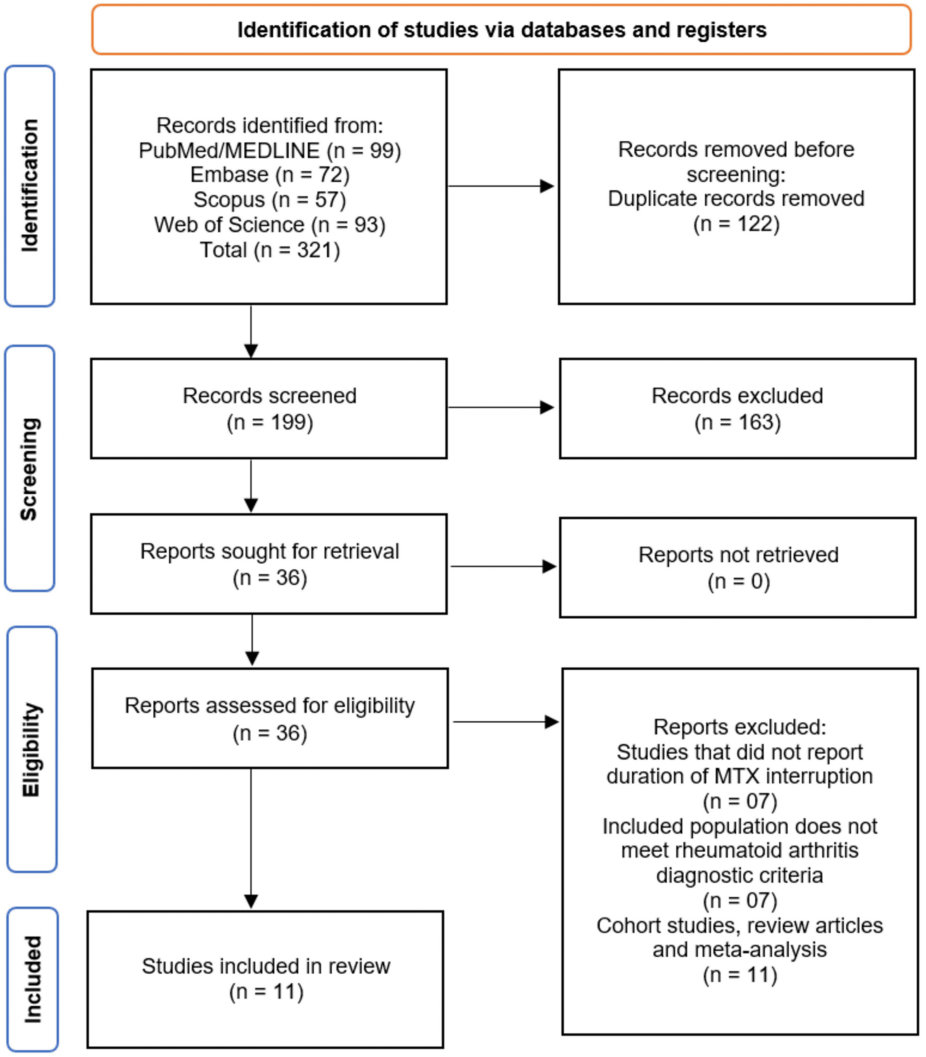


FIGURE 1: Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 flow diagram of study identification, screening, and inclusion.

Counts reflect searches of PubMed/MEDLINE, Embase, Scopus, and Web of Science on September 30, 2026. Citation searching and registry screening yielded no additional eligible reports. MIVAC I and II, reported together, are counted as two studies, and the two vaccine response on, off methotrexate (VROOM) reports as one.

Characteristics of Included Studies

The 11 included studies analyzed 2,099 MTX-treated participants (the sum of the analyzed populations in Table *1*; outcome-specific denominators were smaller in some trials, for example, 92 of 129 participants contributed to the immunogenicity analysis of the CoronaVac trial [20]) and were conducted in South Korea, Brazil, India, the United Kingdom, Germany, Spain, and France. Nine were randomized, and two were observational cohorts. Five studies, with 1,071 participants, were restricted to RA [14,15,19,20,24]. Four randomized studies also enrolled psoriatic arthritis or other immune-mediated inflammatory diseases, and RA accounted for 677 of their 914 participants (74%) [16,18,21]; the two observational cohorts enrolled mixed autoimmune rheumatic disease populations [22,23]. Hold durations ranged from one to four weeks, with additional arms testing holds placed before vaccination and one trial deferring MTX initiation by one month. Study characteristics are summarized in Table *1*.

Study	Country; design	Population (n analyzed)	Vaccine (platform)	Hold	Comparator
Park et al., 2017 [14]	Korea; randomized, four parallel arms	RA on stable MTX (n = 199)	Trivalent influenza (inactivated)	Arm 2: 4 wk before; arm 3: 2 wk before plus 2 wk after; arm 4: 4 wk after	Arm 1: MTX continued
Park et al., 2018 [15]	Korea; randomized, investigator-blinded, multicenter	RA on stable MTX (n = 316)	Quadrivalent influenza (inactivated)	2 wk after vaccination	MTX continued
Skaria et al., 2022 (MIVAC I) [16]	India; randomized, assessor-masked, per-protocol analysis	RA (n = 141) or PsA (n = 17) on stable MTX (n = 158)	ChAdOx1, two doses about 13 wk apart (adenoviral vector)	2 wk after each dose	MTX continued
Skaria et al., 2022 (MIVAC II) [16]	India; randomized, assessor-masked, per-protocol analysis	RA (n = 150) or PsA (n = 7) who had continued MTX at dose 1 (n = 157)	ChAdOx1, dose 2 (adenoviral vector)	2 wk after the second dose only	MTX continued
Abhishek et al., 2022/2024 (VROOM) [17,18]	UK; randomized, open-label, serology masked, multicenter	Immune-mediated inflammatory disease, RA 208 (54%) (n = 383)	COVID-19 booster, 95% mRNA (76% BNT162b2) (mRNA)	2 wk after the booster	MTX continued
Park et al., 2023 [19]	Korea; randomized, investigator-blinded, noninferiority	RA on stable MTX (n = 178)	Quadrivalent influenza (inactivated)	1 wk after vaccination	MTX held 2 wk after vaccination
Araujo et al., 2022 [20]	Brazil; randomized, investigator-blinded, per-protocol analysis	RA with CDAI 10 or less (n = 129; 92 for immunogenicity)	CoronaVac, two doses 28 days apart (inactivated whole-virion)	2 wk after each dose	MTX continued
Martínez-Fleta et al., 2024 [21]	Spain; randomized, three arms	RA (n = 178) and PsA (n = 38) on MTX (n = 216)	mRNA or vector, primary series	1 wk, or 2 wk, after each dose	MTX maintained
Arumahandi de Silva et al., 2022 [22]	Germany; observational, retrospective cohort	Autoimmune rheumatic disease on MTX (n = 64)	mRNA or vector, two doses	Hold by patient or physician choice	MTX continued
Habermann et al., 2022 [23]	Germany; observational, prospective cohort	Rheumatic disease on MTX (n = 50)	COVID-19 mRNA booster	Hold around the booster	MTX continued
Morel et al., 2025 (VACIMRA) [24]	France; randomized, open-label, multicenter	Early RA starting MTX, none in prior 3 months (n = 249)	PCV13 then PPSV23 (conjugate)	MTX initiation deferred 1 month after PCV13	MTX started at the time of vaccination

TABLE 1: Characteristics of included studies.

n denotes participants analyzed in each study (intention-to-treat population of the final report for VROOM); masking refers to outcome assessors or laboratory personnel, as participants and treating clinicians were aware of allocation in every interventional study. Skaria et al. (2022) [16] appears as two entries because that single report describes two separately randomized, independently conducted trials: MIVAC I (Methotrexate Interruption and VACcination trial I), in which methotrexate was held after both ChAdOx1 doses, and MIVAC II, in which it was held only after the second dose in patients who had continued methotrexate at the first. References [17] and [18] are the interim and final reports of one trial, VROOM, and are counted as a single study.

CDAI: Clinical Disease Activity Index; MTX: methotrexate; PCV13: 13-valent pneumococcal conjugate vaccine; PPSV23: 23-valent pneumococcal polysaccharide vaccine; PsA: psoriatic arthritis; RA: rheumatoid arthritis; VROOM: vaccine response on, off methotrexate; wk: weeks

Immunogenicity by Vaccine Platform and Hold Interval

Immunogenicity findings for each vaccine platform, with the corresponding hold intervals, are summarized in Table *2* and set out by study arm below.

Vaccine platform	Studies	Hold interval tested	Effect of hold versus comparator
Inactivated influenza	3 RCTs [14,15,19]	4 wk before; 2 wk before plus 2 wk after; 2 wk after; 4 wk after; 1 wk after	Hold before vaccination alone: no gain. 2 wk after: 75.5% vs 54.5%, RD 21.0 percentage points (95% CI: 10.6-31.7). 1 wk vs 2 wk: 68.9% vs 75.0%, RD -6.1 percentage points (95% CI: -19.3 to 7.1); noninferior against a -20-point margin
Inactivated whole-virion SARS-CoV-2	1 RCT [20]	2 wk after each dose	Seroconversion 78.4% vs 54.5% (RD 23.8 percentage points, 95% CI: 3.9-40.4*); GMT 34.2 (95% CI: 25.2-46.4) vs 16.8 (11.9-23.6)
Adenoviral-vector SARS-CoV-2	2 RCTs [16]	2 wk after each dose; 2 wk after dose 2 only	Median anti-RBD 2484.0 vs 1147.5 IU/mL (ratio 2.16; p = 0.0014) and 2,553.5 vs 990.5 IU/mL (ratio 2.58; p < 0.0001); equivalent titers with either schedule
mRNA SARS-CoV-2	1 RCT [17,18]; 1 randomized three-arm study [21]; 2 cohorts [22,23]	2 wk after booster; 1 or 2 wk after each dose; variable	GMR 2.08 (95% CI: 1.59-2.70) at 4 wk, 1.88 (1.44-2.46) at 12 wk, 1.50 (1.12-2.01) at 26 wk; higher neutralization and interferon-gamma; optimum about 10 days (observational)
Pneumococcal conjugate	1 RCT [24]	Initiation deferred 1 month	Responders 88% vs 75% by ELISA (adjusted RR 1.46, 95% CI: 1.10-1.92); 97% vs 87% by OPA (adjusted RR 1.65, 95% CI: 1.25-2.19)

TABLE 2: Summary of immunogenicity by vaccine platform and hold interval.

The comparator is continued methotrexate except for the one-week arm of Park et al. (2023) [19], where it is a two-week hold. A satisfactory vaccine response is a fourfold or greater rise in hemagglutination-inhibition titer. *Calculated from reported counts (Newcombe hybrid score method). Ratios of medians are descriptive, and no CI can be derived for them. The VACIMRA risk ratios are adjusted for the randomization strata, as reported, and therefore differ from crude ratios of the percentages. The VROOM estimates are from the final analysis [18].

ELISA: enzyme-linked immunosorbent assay; GMR: geometric mean ratio; GMT: geometric mean titer; OPA: opsonophagocytic assay; RBD: receptor-binding domain; RCT: randomized controlled trial; RD: risk difference; RR: risk ratio

Inactivated split-virion influenza vaccine: Three randomized trials examined this platform [14,15,19]. The four-arm dose-finding trial showed that timing, not the hold itself, determines benefit: a four-week hold placed entirely before vaccination resulted in no improvement, whereas arms spanning the post-vaccination period did, with satisfactory responses of 51.0% versus 31.5% for the arm holding two weeks before and two weeks after vaccination (risk difference = 19.5 percentage points, 95% CI: 0.6-36.8, calculated from reported counts) [14]. The confirmatory multicenter trial then raised satisfactory vaccine responses from 54.5% to 75.5%, an absolute gain of 21.0 percentage points (95% CI: 10.6-31.7), with higher seroprotection against all four antigens and no increase in post-vaccination disease activity [15]. The subsequent noninferiority trial found a one-week hold noninferior to two weeks (satisfactory response = 68.9% vs 75.0%; difference = -6.1 percentage points, 95% CI: -19.3 to 7.1, against a prespecified margin of -20 percentage points), with similar response rates at 16 weeks and results unaffected by MTX dose [19].

Inactivated whole-virion SARS-CoV-2 vaccine: One randomized trial addressed this platform [20]. Holding MTX for two weeks after each of two CoronaVac doses given 28 days apart raised anti-S1/S2 IgG seroconversion from 54.5% to 78.4% (risk difference = 23.8 percentage points, 95% CI: 3.9-40.4) with a parallel doubling of geometric mean titers, although neutralizing antibody positivity did not differ significantly. These estimates derive from a per-protocol population of 37 of 60 participants randomized to the hold and 55 of 69 randomized to continuation.

Adenoviral-vector SARS-CoV-2 vaccine: The paired MIVAC trials provide the only randomized data for this platform and isolate the contribution of each dose [16]. Holding MTX after both ChAdOx1 doses more than doubled the median anti-receptor-binding domain (RBD) titer, and holding it only after the second dose produced an essentially equivalent titer, indicating that the benefit concentrates at the dose driving the recall response. The two doses were given a median of about 13 weeks apart, and the comparison between the hold arms of the two trials is between parallel trials rather than within one randomization.

mRNA SARS-CoV-2 vaccine: Four studies, two of them randomized, addressed mRNA platforms [17,18,21-23]. In the final VROOM analysis of 383 randomized participants, in whom 95% of boosters were mRNA vaccines, a two-week hold roughly doubled anti-S1-RBD titers at four weeks (adjusted geometric mean ratio = 2.08, 95% CI: 1.59-2.70); the advantage was attenuated but sustained at 12 weeks (1.88, 1.44-2.46) and 26 weeks (1.50, 1.12-2.01), and live-virus neutralization of the ancestral strain and Omicron BA.1 was also higher at four weeks [18]. The interim analysis of the first 254 participants had reported a ratio of 2.19 (1.57-

3.04) [17]. The Spanish three-arm study found that a hold in infection-naive patients improved anti-RBD IgG and neutralizing antibodies, most clearly in the two-week arm, and additionally raised spike-specific interferon-gamma secretion, extending the benefit from the humoral to the cellular compartment [21]. The two German cohorts both found the post-vaccination rather than the pre-vaccination interval to be the more influential. The retrospective cohort estimated an optimum of approximately 10 days after vaccination [22], and the booster cohort, which measured pseudovirus neutralization of the ancestral strain and the Omicron BA.1 and BA.2 variants, reported a consistent pattern [23]; in both, the benefit was most pronounced at age 60 years and older.

Pneumococcal conjugate vaccine: One randomized trial addressed this platform with a mechanistically analogous strategy: in early RA about to start MTX, deferring MTX initiation for one month after PCV13 raised responder rates from 75% to 88% by enzyme-linked immunosorbent assay (ELISA) (adjusted risk ratio = 1.46, 95% CI: 1.10-1.92) and from 87% to 97% by opsonophagocytic assay (adjusted risk ratio = 1.65, 95% CI: 1.25-2.19), with higher serotype-specific responses persisting at 12 months [24]. Hold windows and their outcomes across all study arms are shown in Figure *2*.

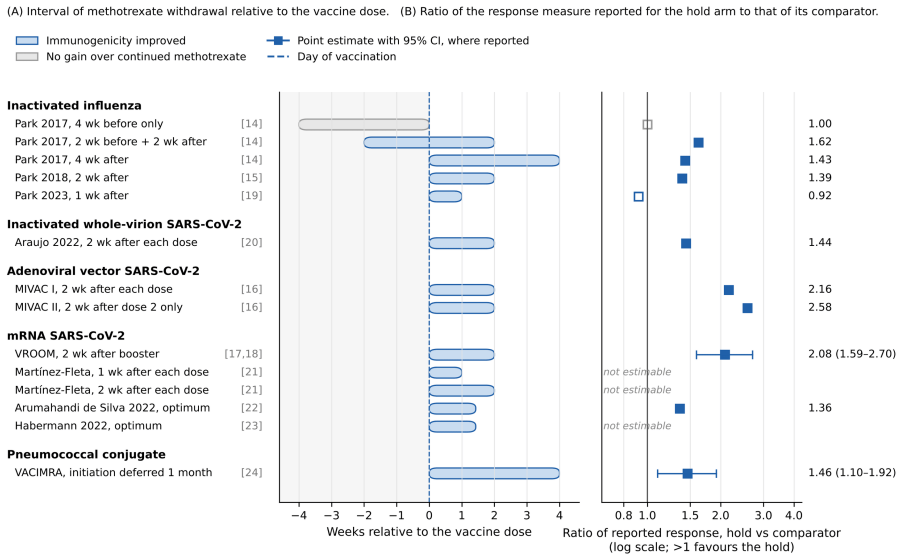


FIGURE 2: Methotrexate hold windows and the associated gain in vaccine response, by vaccine platform.

Panel A, on the left, shows the interval of methotrexate withdrawal relative to the vaccine dose, in weeks; the dashed vertical line marks the day of vaccination. In panel A, blue bars denote arms in which immunogenicity improved and grey bars arms with no gain over continued methotrexate. Panel B, on the right, shows the ratio of the response measure reported for the hold arm to that of its comparator, on a logarithmic scale, with 95% confidence intervals where reported by the trial or, for the one-week influenza arm, the CoronaVac trial, and arm 3 of Park 2017, calculated from reported counts; values above 1.0 favor the hold. The response measures summarized in panel B differ between studies; the ratios are study-specific and descriptive, were not pooled, and are not directly comparable across studies. MIVAC values are ratios of medians, for which no CI can be derived; the VACIMRA value is an adjusted risk ratio; the VROOM value is from the final analysis [18]. Park et al. (2023) [19] compared a one-week with a two-week hold, so its value indicates noninferiority rather than loss of benefit; VACIMRA [24] deferred methotrexate initiation rather than interrupting established therapy.

Disease Activity and Flare

Disease activity was measured with the Disease Activity Score in 28 joints (DAS28) [25] and the Clinical Disease Activity Index (CDAI) [26], and flare was defined by study-specific criteria; the two constructs are reported separately. The flare signal varied with the number and spacing of holds rather than with vaccine platform (Table *3*). A single two-week hold after a single-dose vaccine was not associated with any significant change in disease activity [15]. In the CoronaVac trial, in which holds followed two doses given 28 days apart, CDAI above 10 was comparable at day 28 but more frequent in the hold group at day 69, which the investigators attributed to the short dosing interval [20]. In MIVAC I, in which the two ChAdOx1 doses were about 13 weeks apart, flares after the first dose occurred in 25% of the hold group versus 8% of controls (risk difference = 17.3 percentage points, 95% CI: 5.8-28.6), with a smaller, non-significant difference after the second dose [16]. Confining the hold to the second dose in MIVAC II gave 12% versus 5% (risk difference = 6.9 percentage points, 95% CI: -2.1 to 16.5) while preserving the antibody gain [16]. In the final VROOM analysis, a single two-week hold after a booster was followed by more self-reported flares at four weeks (53% vs 33%; adjusted odds ratio = 2.28, 95% CI: 1.72-3.66) and 12 weeks (65% vs 46%; 1.98, 1.33-2.90) but not at 26 weeks (69% vs 61%; 1.37, 0.72-2.17); most flares were self-managed, the proportion seeking medical help

was similar in both groups, and glucocorticoid use for flares was numerically higher with the hold [18]. The shorter hold in the Korean noninferiority trial was associated with fewer flares than the standard two-week hold [19]. In VACIMRA, deferring initiation produced a higher DAS28 at one month only, with no difference thereafter and no excess of serious adverse events [24].

Study	Hold vs comparator	Outcome (timepoint)	Hold group	Comparator group	Effect estimate or p-value
Park et al., 2017 [14]	4 wk before; 2 wk before plus 2 wk after; 4 wk after vs continued	Physician-assessed flare (4 wk)	21.2%; 34.1%; 38.8%	24.1%	Not significant
Park et al., 2018 [15]	2 wk after vs continued	Disease activity (4 wk)	No significant change	No significant change	Not significant
Skaria et al., 2022 (MIVAC I) [16]	2 wk after each dose vs continued	Flare after dose 1; after dose 2	20/80 (25%); 19/80 (24%)	6/78 (8%); 10/78 (13%)	RD 17.3 (5.8 to 28.6)*, p = 0.005; p = 0.10
Skaria et al., 2022 (MIVAC II) [16]	2 wk after dose 2 only vs continued	Flare after dose 2	9/76 (12%)	4/81 (5%)	RD 6.9 (-2.1 to 16.5)*, p = 0.15
Abhishek et al., 2024 (VROOM) [18]	2 wk after booster vs continued	Self-reported flare (4; 12; 26 wk)	53%; 65%; 69%	33%; 46%; 61%	aOR 2.28 (1.72-3.66); 1.98 (1.33-2.90); 1.37 (0.72-2.17)
Park et al., 2023 [19]	1 wk vs 2 wk after vaccination	Physician-assessed flare (4 wk)	4.5%	12.9%	p = 0.05
Araujo et al., 2022 [20]	2 wk after each of 2 doses vs continued	CDAI above 10 (day 28; day 69)	Comparable; more frequent	Comparable; less frequent	p = 0.12; p = 0.02
Martínez-Fleta et al., 2024 [21]	1 wk or 2 wk after each dose vs maintained	Flare during follow-up (reported as relapse)	No increase in either hold arm	Reference	Not significant
Morel et al., 2025 (VACIMRA) [24]	Initiation deferred 1 month vs started at vaccination	DAS28 (1 month; 3-12 months)	Higher; similar	Lower; similar	Significant at 1 month; not significant thereafter

TABLE 3: Disease activity and flare outcomes by hold schedule.

Values separated by semicolons correspond, in order, to the arms or timepoints listed. *RD in percentage points, calculated from reported counts (Newcombe hybrid score method). Flare definitions were study-specific and are not directly comparable across trials: Park et al. (2017) and Park et al. (2023) used physician-assessed flare, Araujo et al. (2022) a CDAI threshold, the MIVAC trials a predefined rise in DAS28 or in the Clinical Disease Activity Index for Psoriatic Arthritis, or physician intent to escalate therapy, assessed by a masked assessor, VROOM patient-reported flare, and Martínez-Fleta et al. (2024) relapse during follow-up. Where a trial reported a direction of effect without a numerical p value, the direction is stated. The comparator for Park et al. (2023) [19] is a two-week hold rather than continued methotrexate.

aOR, adjusted odds ratio as reported; values in parentheses are 95% CIs. CDAI: Clinical Disease Activity Index [26]; DAS28: Disease Activity Score in 28 joints [25]; MTX: methotrexate; RA: rheumatoid arthritis; RD: risk difference

Risk of Bias

Domain-level judgements are shown in Figure *3* for the randomized studies and Figure *4* for the non-randomized studies. Five of the nine randomized studies were at low risk of bias across all five RoB 2 domains [14,15,17-19,24], the two VROOM reports being assessed together as one trial. The two MIVAC trials and the CoronaVac trial raised some concerns for deviations from intended interventions and for missing outcome data, because each analyzed only participants who completed the protocol after post-randomization exclusions: 158 of 250 and 157 of 178 randomized participants in MIVAC I and II [16], and 37 of 60 and 55 of 69 participants in the CoronaVac hold and continuation arms [20]. The three-arm Spanish study raised some concerns additionally for the randomization process, as allocation concealment was not fully described [21]. Masking differed across trials (Table *1*): disease-activity assessors or investigators were masked in the MIVAC trials, the CoronaVac trial, and two Korean trials [15,16,19,20], and VROOM measured its serological outcomes masked to allocation [18]. Measurement of the outcome was judged at low risk in every randomized study because the primary outcomes were objective laboratory assays. In both observational cohorts, the decision to hold MTX was made by patients or physicians, so bias due to

confounding was judged moderate, as age, disease activity, concomitant immunosuppression, and prior infection could influence both the decision and the antibody response; the remaining six ROBINS-I domains were judged at low risk because exposure and outcomes were defined from documented vaccination dates and laboratory assays, giving an overall judgement of moderate risk of bias [22,23].

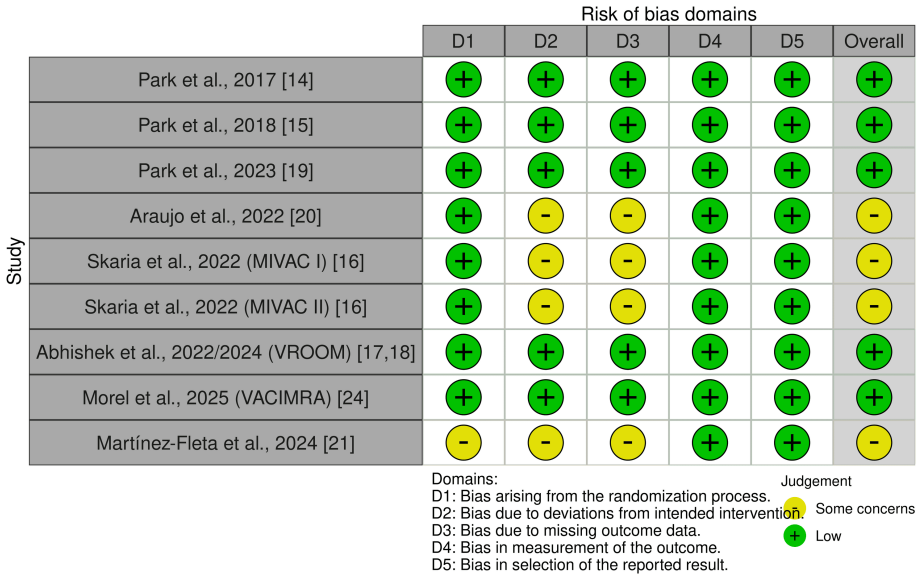


FIGURE 3: Risk of bias in the nine randomized studies (RoB 2).

D1: randomization process; D2: deviations from intended interventions; D3: missing outcome data; D4: measurement of the outcome; D5: selection of the reported result. Some concerns in D2 and D3 for the MIVAC and CoronaVac trials reflect per-protocol analyses after post-randomization exclusions.

RoB: Risk of Bias Tool 2

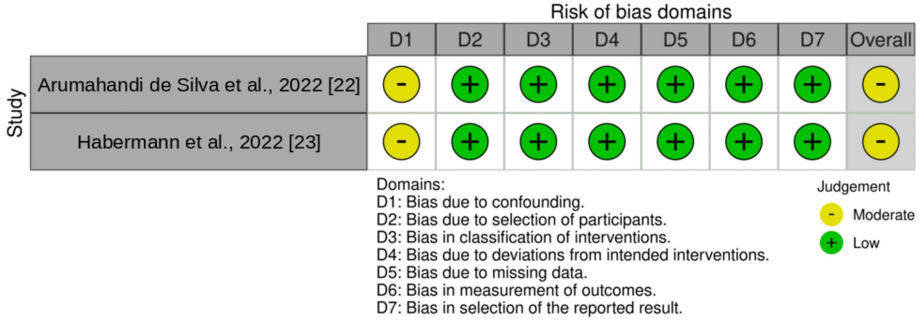


FIGURE 4: Risk of bias in the two non-randomized studies (ROBINS-I).

D1: confounding; D2: selection of participants; D3: classification of interventions; D4: deviations from intended interventions; D5: missing data; D6: measurement of outcomes; D7: selection of the reported result. Bias due to confounding was judged moderate because the decision to hold methotrexate was made by patients or physicians.

ROBINS-I: Risk of Bias in Non-randomized Studies - of Interventions

Certainty of the Evidence

Certainty was rated separately for each platform (Table 4). The body of evidence for inactivated influenza vaccine was of moderate certainty: three randomized trials at low risk of bias gave consistent results, but all came from one research group, and the outcome was immunogenicity rather than clinical infection, so the

rating was downgraded once for indirectness. Evidence for mRNA vaccines was also moderate, supported by one large multicenter trial with a sustained effect to 26 weeks and corroborating randomized and observational data, downgraded once for indirectness because the pivotal trial enrolled a mixed immune-mediated inflammatory disease population and measured antibody titer. Evidence for the pneumococcal conjugate vaccine was moderate, from a single adequately powered multicenter trial downgraded once for indirectness. Certainty was low for the adenoviral-vector platform, where two single-center trials were downgraded for indirectness and for risk of bias, and very low for the inactivated whole-virion platform, where a single small trial analyzed per protocol was downgraded for risk of bias, indirectness, and imprecision. Indirectness was rated serious for every platform because all outcomes were immunological surrogates rather than clinical infection.

Vaccine platform	Evidence base	Risk of bias	Inconsistency	Indirectness	Imprecision	Certainty
Inactivated influenza	3 RCTs [14,15,19]	Not serious	Not serious	Serious (surrogate outcome; single research group)	Not serious	Moderate
Inactivated whole-virion SARS-CoV-2	1 RCT [20]	Serious (per-protocol analysis after exclusions)	Not assessable (single study)	Serious (surrogate outcome)	Serious (92 analyzed; neutralization not significant)	Very low
Adenoviral-vector SARS-CoV-2	2 RCTs [16]	Serious (some concerns on RoB 2)	Not serious	Serious (surrogate outcome; single center)	Not serious	Low
mRNA SARS-CoV-2	1 RCT [17,18]; 1 randomized three-arm study [21]; 2 cohorts [22,23]	Not serious	Not serious	Serious (surrogate outcome; mixed IMID population)	Not serious	Moderate
Pneumococcal conjugate	1 RCT [24]	Not serious	Not assessable (single study)	Serious (surrogate outcome)	Not serious	Moderate

TABLE 4: GRADE certainty of the evidence, by vaccine platform.

Randomized evidence started at high certainty and observational evidence at low certainty. Surrogate outcome denotes that immunogenicity, not clinical infection, was measured. Publication bias could not be assessed formally because no estimates were pooled, and it was not used to downgrade.

GRADE: Grading of Recommendations Assessment, Development and Evaluation [13]; IMID: immune-mediated inflammatory disease; RCT: randomized controlled trial; RoB 2: revised Cochrane risk-of-bias tool for randomized trials

Discussion

This systematic review assembled 11 studies comprising 2,099 MTX-treated participants that compared defined hold intervals around vaccination in RA and closely related inflammatory arthritides. Three findings emerge, although their certainty differs by platform. First, a hold was associated with improved immunogenicity on every platform studied to date: inactivated split-virion influenza, inactivated whole-virion SARS-CoV-2, adenoviral-vector SARS-CoV-2, mRNA SARS-CoV-2, and pneumococcal conjugate; however, the inactivated whole-virion, adenoviral-vector, and pneumococcal conjugate platforms each rest on a single trial or trial report, at very low, low, and moderate certainty, respectively. Second, in the one trial that compared holds before and after vaccination and in two observational cohorts, the window following vaccination appeared to matter more than the one preceding it, with a useful length of approximately one to two weeks. Third, the risk associated with a hold appears to be governed less by the vaccine platform than by how many holds are required and how closely they are spaced, so the number of doses in a series and the interval between them are as relevant to the safety calculus as the duration of any single hold.

Why the Post-vaccination Window May Matter Most

The apparent primacy of the post-vaccination window is the most mechanistically informative observation in this literature, although its randomized support comes from a single influenza trial and the remainder from two observational cohorts. In the four-arm dose-finding trial, withholding MTX for four weeks before vaccination conferred no measurable advantage, whereas arms extending across the post-vaccination period did [14]. The German cohorts, working with different vaccine platforms and assays, reached the same

qualitative conclusion, and the retrospective cohort placed the point beyond which further abstinence yielded little additional gain at approximately 10 days after vaccination [22,23]. Because this threshold rests on observational data, it should be read as indicative rather than validated. This pattern fits what is known about MTX pharmacology and about germinal-center kinetics. Low-dose MTX does not deplete the pre-existing memory compartment; it constrains the proliferation and differentiation of lymphocytes that are actively dividing, through inhibition of purine and pyrimidine synthesis, enhanced adenosine release, and interference with nuclear factor-kappa B and JAK-STAT signaling [4]. Antigen-driven expansion begins within days of immunization and peaks over the following one to two weeks. A drug-free interval covering this peak therefore lifts a constraint precisely when it is biologically consequential, whereas a pre-vaccination interval merely lowers drug exposure when there is no antigen-specific expansion to protect. If confirmed, the practical implication would be that the clinically relevant instruction is not to skip a dose in the abstract, but to omit the dose or doses falling in the one to two weeks after the injection.

How Long the Hold Should Be, by Platform

How long that interval should be is now partly answered for influenza and largely unanswered elsewhere. The Korean noninferiority trial is the only study designed to compare hold durations head to head, and it found a one-week hold noninferior to two weeks for satisfactory vaccine response at four weeks, with similar responses at 16 weeks, comparable seroprotection and titer rises, and numerically fewer flares in the shorter-hold arm [19]. Its prespecified margin of -20 percentage points was, however, close to the 21.0-percentage-point gain that a two-week hold achieved over continued MTX [15]. The lower confidence bound of -19.3 points therefore cannot exclude that a one-week hold retains only part of that gain, although the point estimate of -6.1 points and the concordant secondary outcomes favor near-equivalence. The Spanish three-arm study provides the only other direct comparison of one versus two weeks. Although it was not powered as a formal noninferiority trial, it pointed in a different direction, with the two-week arm showing the clearest gains in anti-RBD IgG, neutralizing antibodies, and spike-specific interferon-gamma production among infection-naive patients [21]. Reconciling these results requires attention to what differs between them. Influenza vaccination in a population with extensive prior exposure elicits a recall response that may be relatively resistant to brief immunosuppression, whereas primary mRNA or vector immunization of an antigen-naive host engages a more protracted germinal-center program that could plausibly benefit from a longer drug-free interval. Until a dedicated noninferiority trial of one versus two weeks is conducted for mRNA and vector platforms, the pragmatic position is that one week is a reasonable option for influenza while two weeks remains the better-supported default for SARS-CoV-2 vaccination, individualized according to how tightly the patient's disease is controlled.

The magnitude of the immunologic gain was broadly similar across platforms once differences in outcome metrics are taken into account. For influenza, roughly one additional patient in every five vaccinated achieved a satisfactory response with the hold, an absolute difference of 21.0 percentage points (95% CI: 10.6-31.7) [15]. For the inactivated whole-virion vaccine, seroconversion rose from 54.5% to 78.4% with a doubling of geometric mean titers [20]. For mRNA boosters, the adjusted geometric mean ratio was 2.08 at four weeks and remained 1.50 at 26 weeks, which addresses the concern that a transient titer increase might reflect accelerated kinetics rather than a durable difference in the quantity of immunity generated [18]. For the adenoviral-vector vaccine, median anti-RBD titers were approximately two- to two-and-a-half-fold higher with the hold, although no CI can be derived for these ratios of medians [16]. This apparent convergence on a roughly twofold effect, in four countries with different assays, supports the direction of the effect, but because three platforms rest on a single trial or trial report and the response measures differ, it does not establish that the effect is of similar size across platforms. It is consistent with the pooled estimate reported in a meta-analysis of the COVID-19 trials, which found a standardized mean difference of 0.70 in antibody titer favoring the hold strategy [8]. The ratios displayed in Figure *2* are nonetheless study-specific and descriptive; they summarize different response measures and should not be read as a common effect size.

Safety, Flare, and the Number of Holds

The MIVAC trials deserve particular emphasis because they answer a question the other studies could not: within a multi-dose primary series, which dose matters? Holding MTX after both ChAdOx1 doses and holding it only after the second produced median anti-RBD titers of 2484.0 and 2553.5 IU/mL, respectively, essentially indistinguishable, while flare rates diverged sharply, at 25% versus 8% when the hold followed the first dose and 12% versus 5%, not significantly different, when it was confined to the second [16]. Because the doses were about 13 weeks apart, the excess in MIVAC I followed the first hold itself rather than a compressed sequence of holds; and because the two strategies were tested in parallel trials, their comparison is not protected by randomization. The interpretation that the second dose drives the recall response, and therefore captures most of the available benefit, offers a straightforward way to decouple efficacy from risk in any two-dose schedule. The Brazilian trial illustrates the complementary risk of close spacing, as excess disease activity appeared only after the second of two holds separated by 28 days [20]. Two holds in under two months amount to omitting a substantial fraction of the cumulative dose, which is a different exposure from a single omission.

Placing the flare data in context requires separating what patients report from what clinicians measure. The

Korean trials, which used physician-assessed disease activity, found no meaningful change in DAS28 with a two-week hold, a conclusion reinforced by a dedicated post-hoc analysis of both trials showing that a two-week hold produced no clinically meaningful change whereas a four-week hold caused a transient rise that reversed on reintroduction [27]. VROOM, which used patient-reported flare in an open-label design, reported more flares with the hold at four and 12 weeks but not at 26 weeks; most were self-managed, and similar proportions of participants in both groups sought medical help [18]. Both sets of observations can be true. Patients who know they have stopped their anchor drug are more likely to notice and report symptoms, and a brief hold can also produce genuine symptomatic worsening without moving composite indices. For the individual patient, the practical consequence is that a hold should be offered with an explicit plan for what to do if symptoms flare, and probably avoided in those with active disease, recent flares, or a history of difficult-to-regain control.

Deferring Initiation in Early Disease

The VACIMRA trial extends the principle in a direction with distinct clinical logic [24]. Rather than interrupting established therapy, it deferred MTX initiation by one month after pneumococcal conjugate vaccination in early RA, raising responder rates from 75% to 88% by ELISA and from 87% to 97% by opsonophagocytic assay, with higher serotype-specific responses persisting at 12 months. Disease activity was higher in the delay arm at one month, as expected of untreated early disease, and equivalent thereafter. This matters because pneumococcal vaccination is the setting in which MTX's inhibitory effect on humoral immunity has been most consistently demonstrated [5], yet the 2022 ACR guideline did not recommend holding MTX around pneumococcal vaccination for want of trial data [7]. The trial reframes the question for newly diagnosed patients: the optimal manoeuvre may be to vaccinate before starting the drug rather than to interrupt it later. It also raises a tension with the treat-to-target imperative to start MTX promptly, which a one-year ancillary analysis addressed by finding no detriment to remission rates, radiographic progression, or treatment escalation [28].

Certainty of the Evidence and Knowledge Gaps

No platform reached high certainty on GRADE assessment, and the reasons point directly to the knowledge gaps. The first and largest is that every included study used antibody titers or responder definitions as the endpoint, and none was powered to detect a difference in clinical infection; this indirectness alone capped certainty at moderate for every platform, and the inactivated whole-virion evidence fell to very low because its single trial was small and analyzed per protocol. The threshold of humoral response required for protection against influenza, COVID-19, or pneumococcal disease in immunosuppressed patients is not established, so the translation from a twofold titer difference to a reduction in pneumonia, hospitalization, or death remains an inference rather than a demonstration. The second gap concerns platforms not yet represented by published trials. It is most conspicuous for the recombinant adjuvanted zoster vaccine, which the 2022 ACR guideline strongly recommends for adults with rheumatic disease receiving immunosuppression, yet around which it advises continuing MTX, reflecting the absence of trial data on holding [7]. An interim analysis of a randomized phase 4 trial of a two-week hold has been presented in abstract form but is not yet available as a full peer-reviewed report; it was therefore excluded here, and whether an adjuvanted subunit vaccine behaves like the platforms studied so far remains open. The third gap is the near-absence of data on cellular immunity: only the Spanish study measured antigen-specific T-cell responses, finding higher interferon-gamma secretion with the hold [21]. This matters because cellular immunity contributes disproportionately to protection against severe disease and because MTX blunts CD8+ T-cell activation after vaccination [6]. The fourth concerns subgroups. The observational cohorts found the benefit of holding concentrated in patients aged 60 and older [22,23], and a modelling analysis of a Korean cohort found that MTX use was not independently associated with reduced humoral response to COVID-19 vaccination once other factors were accounted for [29], which together suggest the effect is not uniform and that targeted strategies might outperform a blanket policy. Finally, no study has examined whether partial dose reduction, as opposed to complete omission, captures some of the immunologic benefit at lower flare risk.

Despite this heterogeneity, the direction of effect was consistent: in every randomized comparison of a hold spanning the post-vaccination period, or of deferred initiation, against continued or immediate MTX, the antibody response was higher with the hold. Consistency of direction is not, however, confirmation of benefit. Three platforms rest on a single trial or trial report; certainty was low or very low for the adenoviral-vector and inactivated whole-virion vaccines and no higher than moderate for any platform; and no trial measured clinical infection. What remains to be settled is the fine calibration: one week or two for each platform, which dose of a series to target, which patients benefit most, and whether any of this changes the outcomes patients care about. Future trials that adopt a common responder definition, standardized assays, and arm-level reporting would allow these questions to be addressed by quantitative pooling with formal exploration of heterogeneity, as is possible when outcome measures are comparable.

Limitations

Several limitations qualify these conclusions. Heterogeneity in vaccine platform, hold schedule, serological assay, and outcome definition precluded quantitative pooling, so effect estimates are presented as reported,

supplemented where necessary by risk differences and ratios calculated from published data, and cross-platform comparisons are qualitative (Appendices). Six of the eleven studies enrolled populations broader than RA, and although RA predominated in each (74% of participants in the four mixed randomized studies), disease-specific estimates could not always be isolated. All three influenza trials came from one research group in a single country, which limits the generalizability of the influenza findings. Two studies were observational, and because the decision to hold MTX was made by patients or physicians rather than assigned, both were judged at moderate risk of bias from confounding, and residual confounding cannot be excluded. Participants and treating clinicians were aware of allocation in every interventional study, which chiefly threatens patient-reported flare outcomes; laboratory outcomes were objective and, where reported, were measured without knowledge of allocation. Three trials analyzed only participants who completed the protocol after post-randomization exclusions, which may have biased their estimates. Every included study used immunogenicity rather than clinical infection as its endpoint, so the clinical benefit of improved titers is inferred rather than demonstrated. Study selection, data extraction, and risk-of-bias assessment were performed by a single reviewer, which raises the possibility of selection, extraction, and appraisal error. Restricting eligibility to full peer-reviewed primary reports excluded conference abstracts and preprints, including interim data on the recombinant zoster vaccine, so the review reflects the published rather than the entire evidence base. Finally, no review protocol was prospectively registered.

Conclusions

Across five vaccine platforms and 11 studies, a brief hold of MTX timed to the period immediately after vaccination was associated with improved humoral responses in RA, with gains of roughly twofold in antibody titer or about 20 absolute percentage points in responder rate, although the strength of this evidence varies by platform and three platforms are supported by a single trial or trial report. The post-vaccination interval appears to matter most; in the only trial that tested it, holding MTX only before vaccination conferred no benefit. For inactivated influenza vaccine, a one-week hold is noninferior to two weeks and may carry a lower flare risk, making it a reasonable option, although the wide noninferiority margin leaves uncertainty about how much of the two-week gain it retains (moderate certainty). For SARS-CoV-2 vaccines across inactivated, adenoviral-vector, and mRNA platforms, a two-week hold after vaccination has the most support, although for the adenoviral-vector and inactivated whole-virion vaccines this rests on a single trial report each (moderate certainty for mRNA, low for adenoviral-vector, and very low for inactivated whole-virion vaccines), with observational data suggesting that about 10 days captures most of the benefit. In the two-dose primary series, in one pair of parallel trials, confining the hold to the final dose preserved the immunologic gain without a significant excess of flares, whereas holds after each dose increased flares, particularly when the doses were four weeks apart. In early RA, giving the pneumococcal conjugate vaccine one month before starting MTX improved and sustained responses in a single trial without compromising one-year disease control (moderate certainty). These strategies should be offered through shared decision-making, reserved for patients with stable or low disease activity, and accompanied by a plan for managing flare. Adequately powered noninferiority trials comparing one- with two-week holds for mRNA, vector, and adjuvanted subunit platforms, studies incorporating cellular immune endpoints, and trials linking enhanced immunogenicity to clinical infection outcomes are the priorities for future research.

Appendices

Database	Search string	Records
PubMed/MEDLINE	(“Methotrexate”[Mesh] OR “methotrexate”[tiab] OR “MTX”[tiab]) AND (“Arthritis, Rheumatoid”[Mesh] OR “rheumatoid arthritis”[tiab] OR “RA”[tiab]) AND (“Vaccination”[Mesh] OR “Vaccines”[Mesh] OR vaccin*[tiab] OR immuniz*[tiab] OR immunis*[tiab]) AND (hold[tiab] OR holding[tiab] OR interrupt*[tiab] OR discontinu*[tiab] OR withdraw*[tiab] OR pause[tiab] OR pausing[tiab] OR “temporary”[tiab] OR “withholding”[tiab] OR “cessation”[tiab] OR “delay”[tiab] OR “time”[tiab] OR “duration”[tiab] OR “interval”[tiab])	99
Embase (Elsevier)	(‘methotrexate’/exp OR methotrexate:ti,ab OR mtx:ti,ab) AND (‘rheumatoid arthritis’/exp OR ‘rheumatoid arthritis’:ti,ab OR ra:ti,ab) AND (‘vaccination’/exp OR ‘vaccine’/exp OR vaccin*:ti,ab OR immuniz*:ti,ab OR immunis*:ti,ab) AND (hold:ti,ab OR holding:ti,ab OR interrupt*:ti,ab OR discontinu*:ti,ab OR withdraw*:ti,ab OR pause:ti,ab OR pausing:ti,ab OR temporary:ti,ab OR withholding:ti,ab OR cessation:ti,ab OR delay:ti,ab OR time:ti,ab OR duration:ti,ab OR interval:ti,ab)	72
Scopus	TITLE-ABS-KEY(methotrexate OR mtx) AND TITLE-ABS-KEY("rheumatoid arthritis" OR ra) AND TITLE-ABS-KEY(vaccin* OR immuniz* OR immunis*) AND TITLE-ABS-KEY(hold OR holding OR interrupt* OR discontinu* OR withdraw* OR pause OR pausing OR temporary OR withholding OR cessation OR delay OR time OR duration OR interval)	57
Web of Science Core Collection	TS=(methotrexate OR MTX) AND TS=(“rheumatoid arthritis” OR RA) AND TS=(vaccin* OR immuniz* OR immunis*) AND TS=(hold OR holding OR interrupt* OR discontinu* OR withdraw* OR pause OR pausing OR temporary OR withholding OR cessation OR delay OR time OR duration OR interval)	93

TABLE 5: Full search strategies and records retrieved, inception to September 30, 2026.

No date, language, or design filters were applied at the search stage; the English-language restriction was applied at eligibility assessment.

Study	Comparison	Outcome (timepoint)	n (hold/ comparator)	Estimate (95% CI)	Basis
Park et al., 2017 [14]	2 wk before plus 2 wk after vs continued	Satisfactory response (4 wk)	49/54	51.0% vs 31.5%; RD 19.5 (0.6 to 36.8); RR 1.62 (1.00 to 2.62)	Calculated
Park et al., 2018 [15]	2 wk after vs continued	Satisfactory response (4 wk)	160/156	75.5% vs 54.5%; RD 21.0 (10.6 to 31.7)	Reported
Park et al., 2023 [19]	1 wk vs 2 wk after	Satisfactory response (4 wk)	90/88	68.9% vs 75.0%; RD -6.1 (-19.3 to 7.1); margin -20	Reported
Araujo et al., 2022 [20]	2 wk after each dose vs continued	Seroconversion (day 69)	37/55	78.4% vs 54.5%; RD 23.8 (3.9 to 40.4); RR 1.44 (1.07 to 1.93)	Calculated
Skaria et al., 2022 (MIVAC I) [16]	2 wk after each dose vs continued	Median anti-RBD, IU/mL (4 wk after dose 2)	80/78	2484.0 vs 1147.5; ratio of medians 2.16; p = 0.0014	Ratio calculated; CI not derivable
Skaria et al., 2022 (MIVAC II) [16]	2 wk after dose 2 vs continued	Median anti-RBD, IU/mL (4 wk after dose 2)	76/81	2553.5 vs 990.5; ratio of medians 2.58; p < 0.0001	Ratio calculated; CI not derivable
Abhishek et al., 2024 (VROOM) [18]	2 wk after booster vs continued	Anti-S1-RBD (4; 12; 26 wk)	180/187; 179/184; 137/151	Adjusted GMR 2.08 (1.59 to 2.70); 1.88 (1.44 to 2.46); 1.50 (1.12 to 2.01)	Reported
Morel et al., 2025 (VACIMRA) [24]	Initiation deferred 1 month vs immediate	Responders by ELISA; by OPA (1 month)	249 in total	88% vs 75%, adjusted RR 1.46 (1.10 to 1.92); 97% vs 87%, adjusted RR 1.65 (1.25 to 2.19)	Reported
Skaria et al., 2022 (MIVAC I) [16]	2 wk after each dose vs continued	Flare after dose 1	80/78	25% vs 8%; RD 17.3 (5.8 to 28.6)	Calculated
Skaria et al., 2022 (MIVAC II) [16]	2 wk after dose 2 vs continued	Flare after dose 2	76/81	12% vs 5%; RD 6.9 (-2.1 to 16.5)	Calculated
Abhishek et al., 2024 (VROOM) [18]	2 wk after booster vs continued	Self-reported flare (4; 12 wk)	191/192	Adjusted OR 2.28 (1.72 to 3.66); 1.98 (1.33 to 2.90)	Reported

TABLE 6: Point estimates, 95% confidence intervals, and sample sizes for each comparison.

RD values are in percentage points. Calculated risk differences use the Newcombe hybrid score method, and calculated risk ratios use the log method, from the per-protocol or modified intention-to-treat counts reported by each study. Comparisons not listed (Martínez-Fleta et al. (2024) and the two observational cohorts) were reported as group comparisons or threshold analyses from which a common effect estimate with a CI could not be derived.

ELISA: enzyme-linked immunosorbent assay; GMR: geometric mean ratio; OPA: opsonophagocytic assay; OR: odds ratio; RBD: receptor-binding domain; RD: risk difference; RR: risk ratio

Additional Information

Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

Concept and design: Hanady ME M Osman, Hager Musa Ahmed Ali, Mohamed Kamal Gomaa Abdelfattah, Mahmoud Abdelghany Eldyasty Ahmed Eid, Khadija Dafa Alla Osman, Fatima Nadeem Salih

Acquisition, analysis, or interpretation of data: Hanady ME M Osman, Mohamed Sobhy Mahmoud Rezk, Mohamed Kamal Gomaa Abdelfattah, Mahmoud Abdelghany Eldyasty Ahmed Eid, Khadija Dafa Alla Osman, Ahmed Mohammed, Fatima Nadeem Salih, Ahmed Abdelateef Ahmed Abdelmageed

Critical review of the manuscript for important intellectual content: Hanady ME M Osman, Mohamed

Sobhy Mahmoud Rezk, Hager Musa Ahmed Ali, Mahmoud Abdelghany Eldyasty Ahmed Eid, Ahmed Mohammed, Ahmed Abdelateef Ahmed Abdelmageed

Drafting of the manuscript: Mohamed Sobhy Mahmoud Rezk, Hager Musa Ahmed Ali, Mohamed Kamal Gomaa Abdelfattah, Khadija Dafa Alla Osman, Ahmed Mohammed, Fatima Nadeem Salih

Supervision: Ahmed Abdelateef Ahmed Abdelmageed

Disclosures

Conflicts of interest: In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work. **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

Acknowledgements

Hager Musa Ahmed Ali equally contributed to this work and should be considered a joint first author of this work. All authors have contributed substantially to this work in accordance with the ICMJE authorship criteria. Although we come from different institutions, we share a common research interest and actively collaborate on joint research projects. We are grateful for this partnership, which has made the present study possible.

References

1. Smolen JS, Landewé RB, Bergstra SA, et al.: EULAR recommendations for the management of rheumatoid arthritis with synthetic and biological disease-modifying antirheumatic drugs: 2022 update. Ann Rheum Dis. 2023, 82:3-18. 10.1136/ard-2022-223356
2. Doran MF, Crowson CS, Pond GR, O'Fallon WM, Gabriel SE: Frequency of infection in patients with rheumatoid arthritis compared with controls: a population-based study. Arthritis Rheum. 2002, 46:2287-93. 10.1002/art.10524
3. Doran MF, Crowson CS, Pond GR, O'Fallon WM, Gabriel SE: Predictors of infection in rheumatoid arthritis . Arthritis Rheum. 2002, 46:2294-300. 10.1002/art.10529
4. Cronstein BN, Aune TM: Methotrexate and its mechanisms of action in inflammatory arthritis . Nat Rev Rheumatol. 2020, 16:145-54. 10.1038/s41584-020-0373-9
5. Hua C, Barnetche T, Combe B, Morel J: Effect of methotrexate, anti-tumor necrosis factor α, and rituximab on the immune response to influenza and pneumococcal vaccines in patients with rheumatoid arthritis: a systematic review and meta-analysis. Arthritis Care Res (Hoboken). 2014, 66:1016-26. 10.1002/acr.22246
6. Haberman RH, Herati R, Simon D, et al.: Methotrexate hampers immunogenicity to BNT162b2 mRNA COVID-19 vaccine in immune-mediated inflammatory disease. Ann Rheum Dis. 2021, 80:1339-44. 10.1136/annrheumdis-2021-220597
7. Bass AR, Chakravarty E, Akl EA, et al.: 2022 American College of Rheumatology Guideline for Vaccinations in Patients With Rheumatic and Musculoskeletal Diseases. Arthritis Rheumatol. 2023, 75:333-48. 10.1002/art.42386
8. Imran M, Ali S, Ibrahim AA, et al.: Effect of methotrexate hold on COVID-19 vaccine response in the patients with autoimmune inflammatory disorders: a systematic review and meta-analysis. Clin Rheumatol. 2024, 43:2203-14. 10.1007/s10067-024-07013-3
9. Page MJ, McKenzie JE, Bossuyt PM, et al.: The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. BMJ. 2021, 372:n71. 10.1136/bmj.n71
10. Richardson WS, Wilson MC, Nishikawa J, Hayward RS: The well-built clinical question: a key to evidence-based decisions. ACP J Club. 1995, 123:A12-3. 10.7326/ACPJC-1995-123-3-A12
11. Sterne JA, Savović J, Page MJ, et al.: RoB 2: a revised tool for assessing risk of bias in randomised trials . BMJ. 2019, 366:l4898. 10.1136/bmj.l4898
12. Sterne JA, Hernán MA, Reeves BC, et al.: ROBINS-I: a tool for assessing risk of bias in non-randomised studies of interventions. BMJ. 2016, 355:i4919. 10.1136/bmj.i4919
13. Guyatt GH, Oxman AD, Vist GE, Kunz R, Falck-Ytter Y, Alonso-Coello P, Schünemann HJ: GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. BMJ. 2008, 336:924-6. 10.1136/bmj.39489.470347.AD
14. Park JK, Lee MA, Lee EY, Song YW, Choi Y, Winthrop KL, Lee EB: Effect of methotrexate discontinuation on efficacy of seasonal influenza vaccination in patients with rheumatoid arthritis: a randomised clinical trial. Ann Rheum Dis. 2017, 76:1559-65. 10.1136/annrheumdis-2017-211128
15. Park JK, Lee YJ, Shin K, et al.: Impact of temporary methotrexate discontinuation for 2 weeks on immunogenicity of seasonal influenza vaccination in patients with rheumatoid arthritis: a randomised clinical trial. Ann Rheum Dis. 2018, 77:898-904. 10.1136/annrheumdis-2018-213222
16. Skaria TG, Sreeprakash A, Umesh R, et al.: Withholding methotrexate after vaccination with ChAdOx1 nCov19 in patients with rheumatoid or psoriatic arthritis in India (MIVAC I and II): results of two, parallel, assessor-masked, randomised controlled trials. Lancet Rheumatol. 2022, 4:e755-64. 10.1016/S2665-9913(22)00228-4
17. Abhishek A, Boyton RJ, Peckham N, et al.: Effect of a 2-week interruption in methotrexate treatment versus continued treatment on COVID-19 booster vaccine immunity in adults with inflammatory conditions

(VROOM study): a randomised, open label, superiority trial. Lancet Respir Med. 2022, 10:840-50. 10.1016/S2213-2600(22)00186-2

18. Abhishek A, Peckham N, Pade C, et al.: Effect of a 2-week interruption in methotrexate treatment on COVID-19 vaccine response in people with immune-mediated inflammatory diseases (VROOM study): a randomised, open label, superiority trial. Lancet Rheumatol. 2024, 6:e92-e104. 10.1016/S2665-9913(23)00298-9
19. Park JK, Lee YJ, Shin K, et al.: A multicenter, prospective, randomized, parallel-group trial on the effects of temporary methotrexate discontinuation for one week versus two weeks on seasonal influenza vaccination in patients with rheumatoid arthritis. Arthritis Rheumatol. 2023, 75:171-7. 10.1002/art.42318
20. Araujo CS, Medeiros-Ribeiro AC, Saad CG, et al.: Two-week methotrexate discontinuation in patients with rheumatoid arthritis vaccinated with inactivated SARS-CoV-2 vaccine: a randomised clinical trial. Ann Rheum Dis. 2022, 81:889-97. 10.1136/annrheumdis-2021-221916
21. Martínez-Fleta P, Vicente-Rabaneda EF, Triguero-Martínez A, et al.: Beneficial effect of temporary methotrexate interruption on B and T cell responses upon SARS-CoV-2 vaccination in patients with rheumatoid arthritis or psoriatic arthritis. NPJ Vaccines. 2024, 9:21. 10.1038/s41541-024-00805-3
22. Arumahandi de Silva AN, Frommert LM, Albach FN, et al.: Pausing methotrexate improves immunogenicity of COVID-19 vaccination in elderly patients with rheumatic diseases. Ann Rheum Dis. 2022, 81:881-8. 10.1136/annrheumdis-2021-221876
23. Habermann E, Gieselmann L, Tober-Lau P, et al.: Pausing methotrexate prevents impairment of Omicron BA.1 and BA.2 neutralisation after COVID-19 booster vaccination. RMD Open. 2022, 8:e002639. 10.1136/rmdopen-2022-002639
24. Morel J, Dernis E, Roux C, et al.: Effect of a 1-month methotrexate delay on pneumococcal vaccine immunogenicity and disease control in patients with early rheumatoid arthritis (VACIMRA): an open-label randomised trial. Lancet Rheumatol. 2025, 7:e675-86. 10.1016/S2665-9913(25)00071-2
25. Prevoo ML, van 't Hof MA, Kuper HH, van Leeuwen MA, van de Putte LB, van Riel PL: Modified disease activity scores that include twenty-eight-joint counts. Development and validation in a prospective longitudinal study of patients with rheumatoid arthritis. Arthritis Rheum. 1995, 38:44-8. 10.1002/art.1780380107
26. Aletaha D, Nell VP, Stamm T, Uffmann M, Pflugbeil S, Machold K, Smolen JS: Acute phase reactants add little to composite disease activity indices for rheumatoid arthritis: validation of a clinical activity score. Arthritis Res Ther. 2005, 7:R796-806. 10.1186/ar1740
27. Park JK, Kim MJ, Choi Y, Winthrop K, Song YW, Lee EB: Effect of short-term methotrexate discontinuation on rheumatoid arthritis disease activity: post-hoc analysis of two randomized trials. Clin Rheumatol. 2020, 39:375-9. 10.1007/s10067-019-04857-y
28. Than T, Mouterde G, Immediato Daien C, et al.: One-month delay in MTX initiation on RA outcomes: 1-year ancillary analysis of the VACIMRA trial. Rheumatology (Oxford). 2026, 65:270. 10.1093/rheumatology/keag270
29. Kim YK, Choi Y, Jung JI, Kim JY, Kim MH, Curtis J, Lee EB: Modeling of antibody responses to COVID-19 vaccination in patients with rheumatoid arthritis. Sci Rep. 2024, 14:1335. 10.1038/s41598-024-51535-4