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Anti-granulocyte scintigraphy in early rheumatoid arthritis - does it work?

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Corresponding Author:	János Gaál, M.D. Kenézy Gyula Hospital Debrecen, HUNGARY
Corresponding Author Secondary Information:	
Corresponding Author's Institution:	Kenézy Gyula Hospital
Corresponding Author's Secondary Institution:	
First Author:	Edit Horkay, MD
First Author Secondary Information:	
Order of Authors:	Edit Horkay, MD
	Gyöngyvér Kincse, MD
	József Varga, Ph.D.
	Lajos Szabados, MD
	Ildikó Garai, MD, Ph.D.
	János Gaál, M.D.
Order of Authors Secondary Information:	
Abstract:	Objective: To compare the performance of anti-granulocyte scintigraphy with those of widely used prognostic indices (such as DAS28, anti-CCP, early MRI imaging). Methods: Twenty-five patients with early arthritis were enrolled into the study. Following the review of clinical data and the evaluation of disease activity, we performed MRI imaging of the hands, anti-granulocyte scintigraphy, and determined anti-CCP positivity. The relationship between the changes of MRI scores and the above prognostic factors were analyzed statistically. Results: At baseline, values were as follows: DAS28 3.86±1.19, CRI 0.15±0.12, MRI erosions and synovitis scores 25.11±12.82 and 4.32±4.02 (respectively), the ratio of anti-CCP positivity was 7/12 (58%). After the follow-up period of 13.6±2.52 months, erosion and synovitis scores were 43.11±22.23, and 5.32±6.16, respectively (p=0,001 and p=0,015). The occurrence of new erosions was correlated with baseline erosion score (k=0.523, p=0.022) and anti-CCP positivity (p=0.021).The relationship between CRI and baseline synovitis score was strong (=0.518, p=0.023), whereas it was weak only between the former and baseline erosion score (=0.402, p=0.08). Conclusion: As shown by this study, potential markers for predicting subsequent destructiveness in early RA include MRI and anti-CCP testing, primarily. 99mTc-labeled anti-granulocyte joint scintigraphy is appropriate for the objective and quantitative appraisal of disease activity.

Introduction

Rheumatoid arthritis (RA) is a chronic, immune-mediated, polyarticular inflammatory disorder, which afflicts approximately one per-cent of the adult population and leads to the gradual destruction of involved joints. RA is associated with a substantial deterioration in quality of life; moreover, it also reduces life expectancy. Therefore, early recognition, along with the prediction of subsequent prognosis and therapeutic efficacy are issues of utmost importance. In early RA (diagnosed 2 years before at the earliest), the literature keeps account of several prognostic factors, potentially useful for predicting the expected outcome of the disease. Predictors, implicated with a higher probability of a poorer prognosis, and the evolution of destructive lesions include rheumatoid factor positivity [1], the presence of IgA-type rheumatoid factors [2] and rheumatic nodules, female gender [3], and the abnormalities of the synovial fluid (leukocytosis, acidosis) [4]. New biological markers providing information on the destruction and renewal of articular cartilage have been identified. Cartilage oligomeric matrix protein (COMP), for example, has been suggested by TSENG *et al* as a potential diagnostic and prognostic marker, also suitable for monitoring therapeutic efficacy [5]. FUJIKAWA *et al* [6] reported similar findings. SYVERSEN *et al* presumed a weak correlation between joint destruction and the elevated serum level of collagen cross-linked C-telopeptide (CTX-1), an established marker of bone resorption [7]. The serum levels of a variety of aggrecan molecules and their fragments have also been suggested as prognostic markers. ROUSSEAU *et al* detected a significant reduction of total aggrecan level, along with the presence of at least one specific subpopulation of aggrecan fragments [8]. Remarkable laboratory indices of prognostic value include anti-citrullinated protein/peptide antibodies (ACPA), and anti-mutant-citrullinated vimentin antibodies (anti-MCV). According to SYVERSEN *et al*, the presence of the latter is as reliable a predictor of subsequent articular

destruction as anti-CCP positivity [9]. Imaging studies are additional, important supplements to laboratory testing.

According to clinical observations, neutrophil leukocytes predominate the cell population present in joints afflicted by active RA. Therefore, the accumulation of leukocytes may be regarded as an essential event in rheumatoid synovitis [10]. Although leukocyte infiltration into the joints of patients with early arthritis can be detected by scintigraphy, the value of this finding regarding the subsequent evolution of erosion is unknown [11].

Our study, conducted on patients with early rheumatoid arthritis (characterized by a disease duration shorter than a year), intended to ascertain the prognostic value of ^{99m}Tc-labeled leukocyte scintigraphy in predicting the eventual outcome of the disease process, especially the occurrence of erosions.

Materials and methods

Patients

Twenty-five patients, who had been followed up at the Early Arthritis Center of 'Kenézy Gyula' Hospital were enrolled. The patient collection period started in December, 2008 and ended in December, 2009. All patients fulfilled the ACR 1987 criteria of RA [12] . By the end of the follow-up period of one year on average, clinical, and laboratory data were available on 19 out of the 25 patients. 6 patient lost for follow up: 2 patients withdrew the informed consent, 3 patients refused the regular controls during the study because of traffic difficulties and 1 patient moved off from the city and became unreachable.

Methods

Upon inclusion, we reviewed clinical and laboratory data. An MRI of the hands was obtained to diagnose and quantify the synovitis and after that a planar joint scintigraphy was performed using Fab' fragments of monoclonal, ^{99m}Tc-labeled anti-granulocyte antibodies (anti-NCA-90, sulesomab, LeukoScan®). MRI and scintigraphy were performed at an interval of two weeks

at the most. The clinical and laboratory patameters of the patients were followed up for a year on average. RA was managed with pharmacotherapy chosen by the rheumatologist responsible for the patient's medical care, in view of clinical disease activity. MRI was repeated after a year on average. We correlated the severity of new erosions and of synovitis to baseline findings of MRI and scintigraphy.

The used statistical methods were: the Kolmogorov–Smirnov test to investigate the normality of the data distribution, the Pearson's correlation test for seeking the correlation between the clinical and scintigraphic data, the independent samples *t*-test to seek the relationship between anti-CCP positivity and the development of new erosions and synovitis and paired samples t test for evaluation of the changes in MRI scores (erosion and synivitis).

The results were regarded to be significant if p value was <0,05. The study protocol was approved by the Institutional Review Board of University of Debrecen, Medical & Health Sciences Center; and an informed consent was obtained from all patients.

MRI was performed using equipment specifically designed for imaging of the extremities (E-scan 0.2 T, Esaote Biomedica, Geneva, Italy) on recumbent patients. Native, and gadolinium-enhanced (0.1 mmol/kg b. w. Gd-DTPA, Magnevist®, Schering, Berlin, Germany) coronal T1- and T2-weighted, STIR, as well as 3D T1-weighted images were obtained along with axial T1-weighted scans (Figure 2.). We evaluated the images according to the method described by KLARLUND *et al* in 2005 [13]. We appraised the erosions individually, in 14 localizations altogether (including the radial and ulnar epiphyses, all carpal bones, and the 2nd through 5th metacarpal heads), by absolute numbers, as well as by greatest diameters. Using the latter, the mean surface area of erosions in the studied regions were determined and then, aggregated.

The synovitis score was evaluated semi-quantitatively, according to a four-grade scale (0 = no synovial thickening, 1 through 3 = mild/moderate/severe thickening) for each region

and then, individual scores were aggregated. The extent of synovitis was determined in the distal radioulnar, intercarpal, and in the second through fifth metacarpophalangeal joints (6 localizations altogether) – thus, a maximal score of 18 could be achieved. This was increased further by the tenosynovitis score, which was "1" in the presence and "0" in the absence of detectable synovial inflammation.

During the scintigraphy, 800 MBq ^{99m}Tc LeukoScan (Immunomedics GmbH, Germany) was injected intravenously. The injected radioactivity was measured with a dose calibrator, and converted to counts/sec using the calibration factor measured for the camera-collimator system previously. Static images of the wrists and hands were acquired 4 h after the administration of the radiopharmaceutical. The images were acquired into 128*128 matrices over 5 minutes, using MB-9200 gamma camera (Gamma Move, Hungary) equipped with a LEOP collimator. The images were a) evaluated visually in a qualitative manner (for increased radiopharmaceutical uptake); and (b) the regional uptake of the radiopharmaceutical was calculated for the hands. Regions of interest (ROI-s) were drawn around the wrists and the hands (Figure 3.), and their uptake was expressed as a percentage of the injected dose (corrected for Tc decay). We aggregated regional radiopharmaceutical uptake measured in the joints into a single uptake parameter (Cumulative Radiopharmaceutical-uptake Index-CRI).

Results

The essential clinical parameters of the study population are shown in Figure 1. At baseline, the mean age of patients was 53.5±11.9 years; the female-to-male ration was 15/4; and mean disease duration was 4.97±3.03 months. The clinical activity of synovitis was in the moderate range, as shown by the three-item activity index evaluating 28 joints (DAS28) of 3.83±1.19. Seven patients received methotrexate, one patient each was on hydroxychloroquine or sulfasalazine monotherapy; 5 underwent combination therapy with hydroxychloroquine and methotrexate. Additional combinations comprised methotrexate with sulfasalazine,

methotrexate with sulfasalazine and hydroxychloroquine, and methotrexate with hydroxychloroquine and leflunomide, in one patient each. Finally, two patients were treated with methotrexate, sulfasalazine, and leflunomide, as first-line combination therapy. Fifteen patients required transient steroid therapy over a period of 4.28±1.46 months on average. Eleven of then nineteen patients (58%) were rheumatoid factor-positive, whereas the proportion of anti-CCP (anti-cyclic citrullinated peptide) positive patients was 37% (7/19 patients).

Baseline MRI erosion score of the hands was 25.11±12.82; the initial synovitis score was 4.32±4.02, and the cumulative radiopharmaceutical uptake index (CRI) was 0.15±0.12. By the end of the follow-up period of 13.26 ±2.52 months, the MRI erosion and synovitis scores changed to 43.11±22.23 and 5.32±6.16, respectively (p=0,001 and p=0,015).

We did not find statistically significant relationships between patient age, gender, and disease duration, as well as rheumatoid factor positivity, or the occurrence of erosions.

DAS28 and initial synovitis scores, by contrast, were closely related (κ=0.622, p=0.042). The occurrence of new erosions was related both to the initial erosion score (κ=0.523, p=0.022), and to anti-CCP positivity (p=0.021).

As shown in Figure 4. the cumulative radiopharmaceutical uptake index (CRI) measured by scintigraphy was correlated with anti-CCP positivity (p=0.048), as well as with baseline synovitis score (κ=0.518, p=0.023), but not with the extent of erosions that had evolved over the one-year-long follow-up period.

Discussion

The prevention of articular damage is the ultimate goal of the management of RA. The current therapeutic strategy is essentially determined by inflammatory activity, as this has been

reasonably related to subsequent erosiveness. A variety of disease activity indexes are in use for characterizing the intensity of arthritis. Disease Activity Score 28 (DAS28), which is the most widely used tool for this purpose [14], evaluates both objective components (number of swollen joints) and subjective elements (number of tender joints, subjective pain intensity assessed by the patient on a visual analogue scale), along with partially non-specific variables (ESR, C-reactive protein level). As a result, the reproducibility of articular indexes is at least questionable, considering the diverse pain perception thresholds of patients, or the non-specific character of ESR.

Many attempts have been made for the quantitation of the activity of arthritis by diagnostic imaging modalities, including radionuclide studies, some of which may prove adequate for determining the activity of RA. Having evaluated the extent of bone marrow edema with MRI of the hand, wrist, and feet in their study of 2-years duration, HETLAND *et al* consider this index the strongest, independent predicting factor of radiological progression [15]. PALOSAARI *et al* found a close relationship between bone marrow edema and subsequent articular erosions in their 2-year study [16]. OSTGENDORF *et al* studied the diagnostic efficacy of duplex ultrasound, positron emission tomography (PET), and mini-arthroscopy and found this combination useful for the early detection of pathomorphological abnormalities as well as for the prediction of prognosis [17].

VAN DER LAKEN found that the PET imaging of macrophages, undertaken after the [11C](R)-PK11195-labeling of their benzodiazepine receptors, offers non-invasive means both for the detection of incipient synovitis and for the subsequent monitoring of its activity [18]. Studying 13 RA patients with ^{99m}Tc-MDP joint scintigraphy, MÖTTÖNEN *et al* demonstrated a correlation between high scintigraphic activity and subsequent erosiveness. Their findings also suggest that erosions do not evolve in scintigraphically inactive joints [19].

DE BOIS *et al* had performed ^{99m}Tc-IgG joint scintigraphy in 30 patients with early rheumatoid arthritis and found, after one-year of follow-up, that this form of radionuclide imaging might prove useful for the prediction of joint destruction [20]. TAKALO *et al* reported similar results with of ^{99m}Tc-nanokolloid joint scintigraphy [21]. Using ^{99m}Tc-HMPAO-leukocyte scintigraphy, our work group showed a relationship between the magnitude of articular leukocyte infiltration and the number of swollen joints [14].

No studies have been conducted to date to clarify the relationship between the extent of leukocyte infiltration and disease activity, or erosiveness. As suggested by the relationship between CRI (calculated radionuclide uptake index) of the hands and anti-CCP positivity, as well as the MRI synovitis score, ^{99m}Tc-labeled anti-granulocyte joint scintigraphy is appropriate for the objective and quantitative appraisal of disease activity, primarily.

Nevertheless, ^{99m}Tc-labeled anti-granulocyte scintigraphy should be performed when access to MRI is limited, non-existent, or the waiting list for this test is long. Moreover, the cost of ^{99m}Tc-labeled anti-granulocyte scintigraphy is significantly lower than that of MRI, and being a ubiquitously available (in any radioisotope diagnostic laboratory equipped with a gamma camera), rapid, and relatively straightforward test, it is a realistic alternative to MRI in cases where the objective appraisal of the activity of arthritis is important. High scintigraphic activity detected over the hands suggests a substantial leukocyte infiltration, which might require the early initiation of aggressive therapy.

Study limitations: the relatively low patient number, and the lack of a uniform treatment algorhytm may be considered a potential flaw of this study.

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Declaration: The first two authors contributed equally to this work.

Figure legends

Figure 1.

Essential clinical data of the study population

Figure 2.

3DT1-weighted MRI images of the region from the radiocarpal to the MCP joints of the same patient. *Left:* No erosions are visible on this baseline scan. *Right:* On this image obtained 13 months later, erosions are depicted on the lateral-radial surface of the distal radioulnar joint.

Figure 3.

99mTc anti-granulocyte scintiscan of the hands, with superimposed ROIs: the involvement of the left wrist and of the second MCP joint of the right hand are visible.

Figure 4.

The relationship between the cumulative index of radioisotope uptake and baseline synovitis score.

Figure 1.
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Mean age (years)	53.5 ± 11.9
Female-to-male ratio	15/4
Mean disease duration (months)	4.97 ± 3.03
Baseline DAS28	3.86 ± 1.19
DMARD monotherapy (%)	9 (47%)
Combination therapy	10 (53%)
Steroid therapy	15 (79%)
RF positivity	11 (58%
aCCP positivity	7 (37%)

Figure 2.
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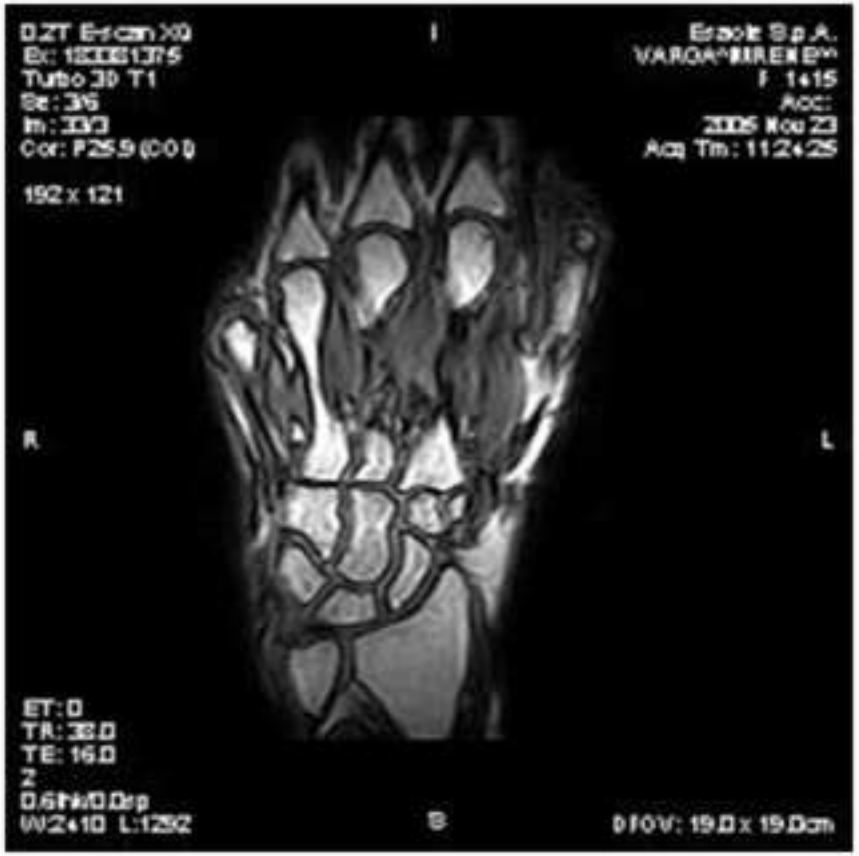

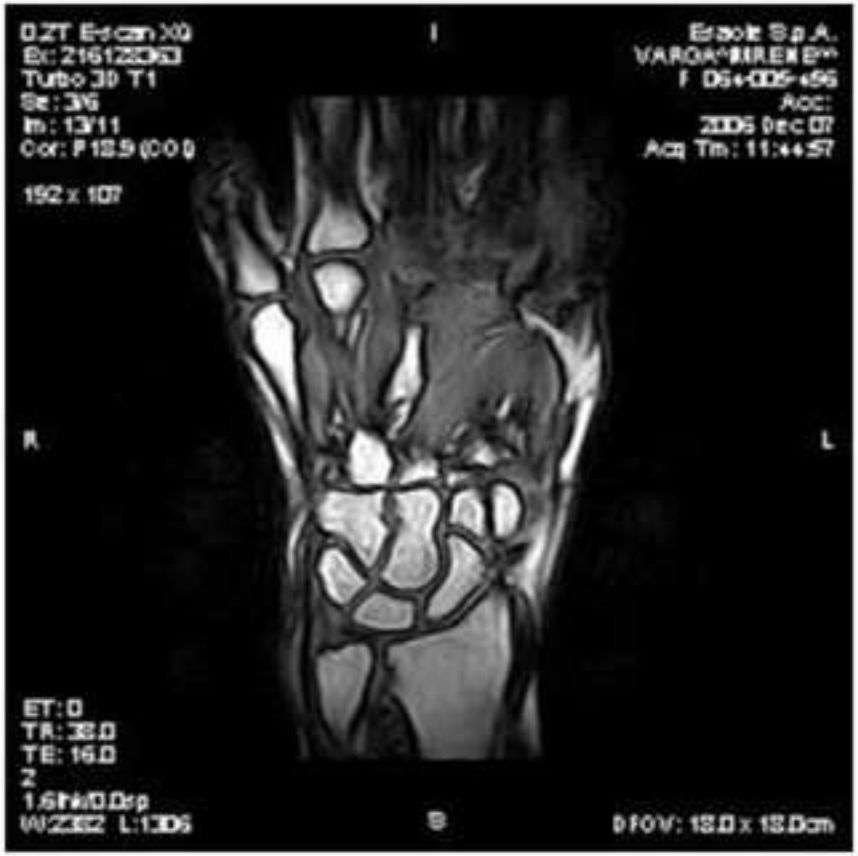

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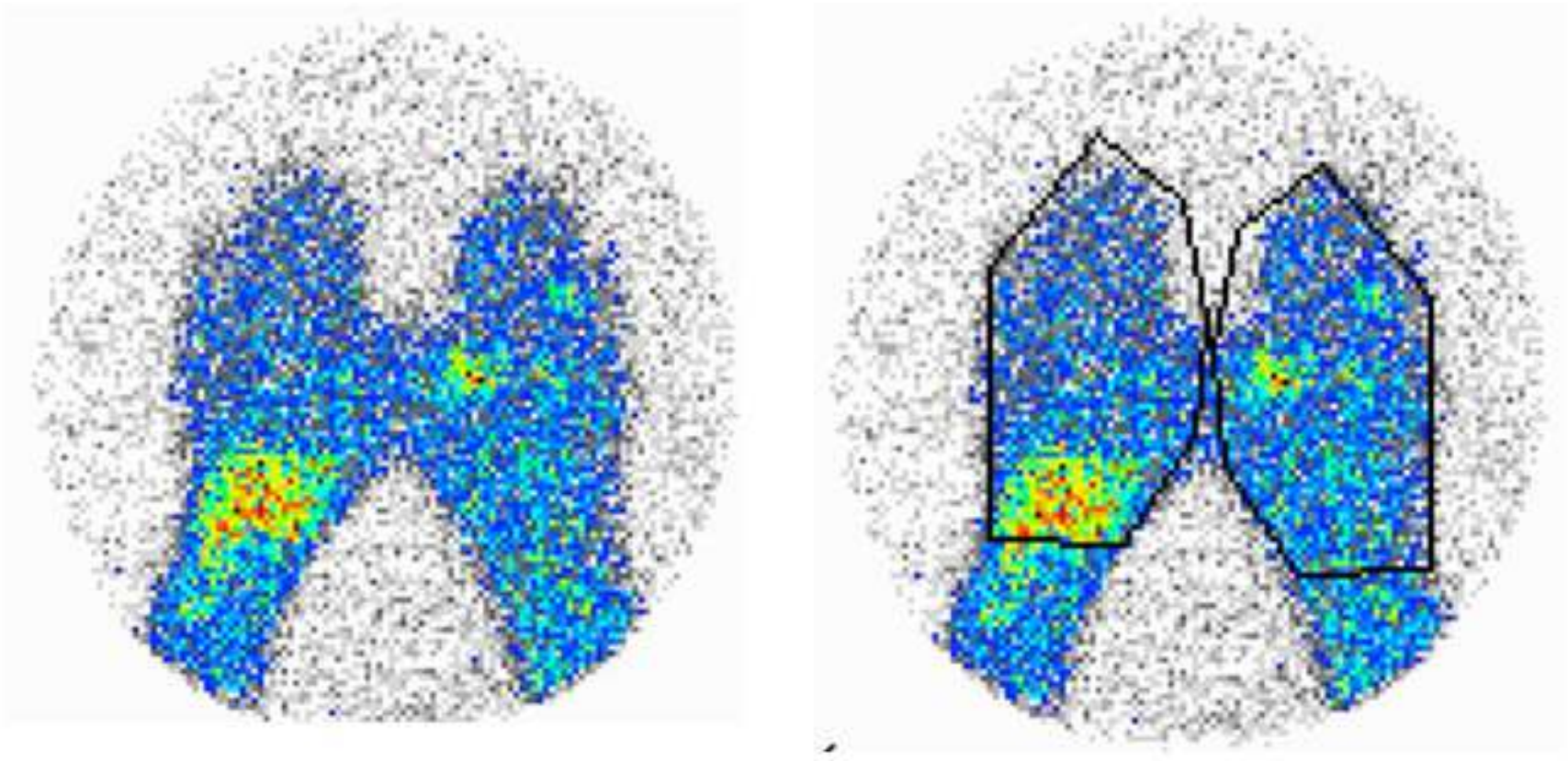

Figure 4.
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