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Original Article

Serum biomarkers for diagnosis and severity evaluation in knee osteoarthritis

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Abstract: Objective: This study aimed to investigate the serum levels of signal transducer and activator of transcription 3 (STAT3), vascular endothelial growth factor (VEGF), and interleukins (IL)-2 and IL-4 in patients with knee osteoarthritis (KOA), and to evaluate their diagnostic value for disease severity. Methods: A total of 115 KOA patients and 50 healthy controls were enrolled. Serum levels of STAT3, VEGF, IL-2, and IL-4 were measured in all participants. The correlations between these biomarkers and KOA severity were further analyzed. Results: Compared to healthy controls, KOA patients exhibited significantly elevated serum levels of STAT3, VEGF, IL-2, and IL-4 (all $P < 0.05$). Following standard treatment, the concentrations of these biomarkers significantly decreased. Receiver operating characteristic (ROC) curve analysis revealed that the area under the curve (AUC) values for diagnosing KOA were 0.710 for STAT3, 0.726 for VEGF, 0.704 for IL-2, and 0.797 for IL-4. Furthermore, the serum concentrations of these factors showed strong positive correlations with KOA severity ($r = 0.436$ for STAT3, $r = 0.467$ for VEGF, $r = 0.497$ for IL-2, and $r = 0.509$ for IL-4). Their AUC values for assessing disease severity were 0.723 (STAT3), 0.742 (VEGF), 0.769 (IL-2), and 0.757 (IL-4). Combined detection of these biomarkers improved diagnostic accuracy. Conclusions: Abnormally elevated serum levels of STAT3, VEGF, IL-2, and IL-4 in KOA patients are closely associated with disease severity and treatment efficacy. These biomarkers may serve as auxiliary indicators for KOA diagnosis and severity assessment.

Keywords: Knee osteoarthritis, signal transducer and activator of transcription 3, vascular endothelial growth factor, interleukin-2, interleukin-4

Introduction

Knee osteoarthritis (KOA) is a progressive inflammatory condition that primarily affects the articular cartilage, subchondral bone, and surrounding synovial tissues [1]. As a major cause of disability, KOA is particularly prevalent among the elderly. Globally, it affects approximately 600 million people, with a higher prevalence in women (10.3%) compared to men (5.7%), and incidence rates reaching nearly 20% in individuals over 60 years of age [2-4]. Clinically, KOA is characterized by joint pain, transient morning stiffness, and limited mobility, all of which significantly impair patients' quality of life [5, 6]. Current treatments focus on reducing inflammation, alleviating pain, and slowing disease progression; however, no therapies can halt or reverse joint degeneration. Surgical interventions are often considered for advanced-stage patients but rarely achieve full recovery [7, 8]. Therefore, elucidating the underlying pathogenic mechanisms and identifying reliable diagnostic markers are crucial for improving early detection and treatment outcomes in KOA.

Radiographic imaging remains the gold standard for KOA diagnosis, enabling visualization of joint space narrowing, osteophyte formation, subchondral sclerosis, and cysts. However, its limited sensitivity and specificity highlight the urgent need for more reliable diagnostic alternatives [9]. In contrast, serological biomarkers offer high sensitivity and specificity in various diseases, along with operational simplicity, convenience, and enhanced safety [10].

Signal transducer and activator of transcription 3 (STAT3), a key cytokine-responsive transcrip-

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tion factor, is closely associated with angiogenesis in multiple diseases and cancers, and contributes to KOA progression by regulating angiogenesis-mediated osteogenic differentiation and chondrocyte pathology [11, 12]. Similarly, vascular endothelial growth factor (VEGF), a central regulator of angiogenesis acting through tyrosine kinase receptors, has been linked to joint pain development in KOA [13]. Emerging evidence also suggests that nanoparticle-based suppression of VEGF receptors can alleviate pain and cartilage damage in KOA patients [14]. Additionally, interleukins (IL)-2 and IL-4, as inflammatory mediators, play roles in bone marrow pathology and functional impairment in KOA, indicating their involvement in disease-related inflammation [15].

Despite these insights, few studies have explored the serum concentrations of STAT3, VEGF, IL-2, and IL-4 in KOA patients and their clinical significance. This study hypothesizes that these four biomarkers could serve as reliable tools for diagnosing KOA and assessing its severity, thereby providing new perspectives for clinical evaluation and management. The key innovations of this study are as follows: First, it is the first to elucidate the diagnostic value of STAT3, VEGF, IL-2, and IL-4 in KOA, establishing them as potential biomarkers for distinguishing KOA patients from healthy controls. Second, this research clarifies the association between these serum markers and KOA severity, along with their specific predictive values, paving the way for novel severity prediction approaches. Most notably, this study proposes a high-performance multi-parameter diagnostic framework that substantially improves KOA detection accuracy and disease staging, offering clinicians a more precise tool for practical application.

Materials and methods

Participant selection

This research protocol was approved by the Ethics Committee of Hebei General Hospital. The study enrolled 115 KOA patients treated at Hebei General Hospital and recruited 50 healthy individuals as controls. Participant recruitment and data collection were conducted over a 24-month period, from January 2022 to January 2024. Based on a power analysis accounting for an anticipated 20% attrition rate, the minimum required sample sizes were calculated to be 47 patients (KOA group) and 39 controls to achieve adequate statistical power. The final sample sizes met these requirements. Sample size calculations were performed using the following formula:

$$n = \frac{2(Z_{\alpha/2} + Z_{\beta})^2 \times \sigma^2}{\delta^2}$$

Inclusion criteria were as follows: diagnosis of KOA according to standardized diagnostic guidelines [16]; presence of clinical symptoms for at least three months; disease classification based on the Kellgren-Lawrence (KL) grading scale [17]; no prior treatment for KOA; X-ray evaluations jointly performed by radiologists and orthopedic surgeons; normal cognitive and communication abilities without neurological impairments; and complete medical records.

Exclusion criteria included: bone tumors; prior joint trauma or rheumatoid arthritis; infectious diseases; previous knee surgeries; significant knee deformities (varus > 20° or valgus > 10°); limited range of motion (flexion < 90° or extension limitation > 5°); comorbidities such as hypertension, diabetes, or coronary heart disease; recent pharmacological treatments that could affect study outcomes; malignancies or severe organ dysfunction; and pregnancy or lactation.

Assessment of disease severity

Cartilage damage was evaluated using arthroscopy combined with the Outerbridge classification and the International Cartilage Repair Society (ICRS) grading system [18].

(1) Grade I: Mild softening or swelling of the cartilage surface, occasionally with minor blistering. (2) Grade II: Surface irregularities or superficial ulcers and fibrosis with a diameter < 1 cm. (3) Grade III: Deep ulcers > 1 cm in diameter without exposure of subchondral bone. (4) Grade IV: Full-thickness cartilage loss with exposed subchondral bone.

The KL grading system was applied as follows: (1) Grade 0: No radiographic abnormalities. (2) Grade I: No joint space narrowing, with detectable osteophytes or minor bone spurs. (3) Grade II: Possible joint space narrowing with mild osteophyte formation. (4) Grade III: Marked joint space narrowing, sclerotic changes, and

moderate osteophyte formation. (5) Grade IV: Severe joint space narrowing with significant sclerosis, joint deformity, and extensive osteophyte formation.

KOA severity classification [19]: (1) Mild: Grade I cartilage damage and KL grades 0-I. (2) Moderate: Grade II-III cartilage damage and KL grade II. (3) Severe: Grade IV cartilage damage and KL grades III-IV.

Detection methods

Serum levels of STAT3, VEGF, IL-2, and IL-4 were measured using enzyme-linked immunosorbent assay (ELISA; Wenzhou KeMiao Biological Technology Co., Ltd.; kit numbers KM092272, KMEHu010115, KMEHu012479, KMEHu011730). Fasting venous blood samples (2 mL) were collected from all participants; KOA patients provided additional samples after eight weeks of treatment. Samples were centrifuged to obtain serum, which was then transferred to 96-well microplates pre-coated with rabbit polyclonal antibodies against human STAT3, VEGF, IL-2, and IL-4. Plates were incubated for 1 hour at 37°C, washed three times with buffer, and incubated with enzyme-conjugated secondary antibodies (0.1 mL per well) at 37°C for another hour. After three further washes, 0.1 mL of freshly prepared TMB substrate solution was added to each well and incubated for 15 minutes at 37°C. The reaction was terminated by adding 2 mol/L sulfuric acid, and absorbance was measured at 450 nm using a microplate reader. Standard curves were constructed using recombinant human proteins. Each sample was analyzed in triplicate, and mean values were used for analysis.

Treatment protocol

KOA patients received a standard treatment regimen comprising celecoxib and glucosamine sulfate. Celecoxib (Beijing Kangruina Biotechnology Co., Ltd.; Batch No. LB1351) was administered orally at 200 mg once daily. Glucosamine sulfate capsules (Beijing Yita Biotechnology Co., Ltd.; Batch No. YT62582) were prescribed at two capsules, taken three times daily. Both medications were continued for eight weeks without interruption.

Statistical analysis

Continuous data were tested for homogeneity of variance using Bartlett's test and for normality using the Kolmogorov-Smirnov test. Data meeting these assumptions were presented as mean ± standard error of the mean (SEM), and group comparisons were conducted using independent samples t-tests. Categorical variables were expressed as percentages and analyzed using chi-square (χ^2) tests. Diagnostic performance was assessed via receiver operating characteristic (ROC) curve analysis, with area under the curve (AUC) values calculated. Optimal cut-off values were determined using the Youden index, which maximizes the sum of sensitivity and specificity. Spearman's correlation analysis was used to examine relationships between serum biomarker levels and KOA severity. All analyses were performed using GraphPad Prism 7.0, with $P < 0.05$ considered statistically significant.

Results

Comparison of baseline characteristics of participants

The baseline characteristics of participants, including gender, age, body mass index (BMI), family medical history, alcohol consumption, and smoking status, showed no significant differences between the KOA and control groups (all $P > 0.05$). Among KOA patients, disease severity was categorized as mild in 52 cases, moderate in 40 cases, and severe in 23 cases (**Table 1**).

Comparison of serum STAT3, VEGF, IL-2, and IL-4 levels

Serum levels of STAT3, VEGF, IL-2, and IL-4 were significantly higher in KOA patients than in healthy controls (**Figure 1**). Specifically, STAT3 concentrations averaged 1.33 ± 0.62 ng/L in KOA patients compared to 0.93 ± 0.26 ng/L in controls. VEGF levels were 90.77 ± 31.52 pg/mL in the KOA group versus 66.84 ± 18.81 pg/mL in controls. IL-2 levels were 48.32 ± 18.68 pg/mL in patients and 35.8 ± 13.14 pg/mL in controls. IL-4 levels reached 21.67 ± 9.44 pg/mL in KOA patients, significantly exceeding the 12.62 ± 3.31 pg/mL observed in healthy individuals. All differences were statistically significant (all $P < 0.001$).

Comparison of serum STAT3, VEGF, IL-2, and IL-4 levels before and after treatment in KOA patients

To evaluate therapeutic efficacy, changes in serum STAT3, VEGF, IL-2, and IL-4 levels were

Table 1. Comparison of baseline characteristics

Indicators	Research group (n = 115)	Control group (n = 50)	χ^2/t	*P*
Gender			1.364	0.243
Male	44 (38.26)	24 (48.00)		
Female	71 (61.74)	26 (52.00)		
Age (years)	61.00 ± 8.19	60.78 ± 7.12	0.165	0.869
BMI (kg/m^2)	22.18 ± 2.67	22.04 ± 2.62	0.311	0.756
Family medical history	11 (9.57)	3 (6.00)	0.570	0.450
Alcohol consumption history	29 (25.22)	12 (24.00)	0.028	0.868
Smoking history	15 (13.04)	8 (16.00)	0.254	0.614
Disease severity			-	-
Mild	52 (45.22)	-		
Moderate	40 (34.78)	-		
Severe	23 (20.00)	-		

Research group: KOA patients; Control group: Healthy controls.

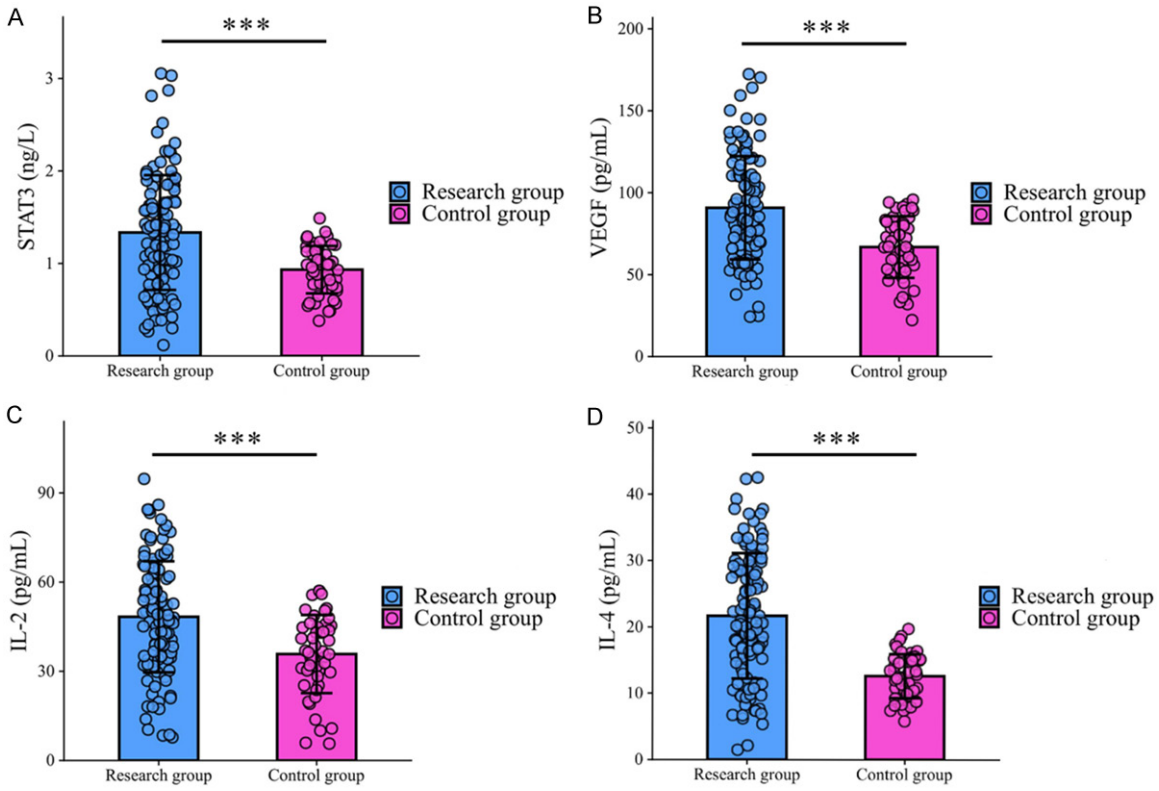

Figure 1. Comparison of serum levels of STAT3, VEGF, IL-2, and IL-4 between groups. A. Comparison of serum STAT3 levels. B. Comparison of serum VEGF levels. C. Comparison of serum IL-2 levels. D. Comparison of serum IL-4 levels. Notes: STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4. ***P < 0.001. Research group: KOA patients; Control group: Healthy controls.

assessed following treatment. Post-treatment, STAT3 and VEGF concentrations showed significant reductions, with VEGF exhibiting a particularly notable decrease. Similarly, IL-2 and IL-4 levels were significantly lower after treatment compared to pretreatment values (all P < 0.05; **Figure 2**).

Diagnostic performance of serum STAT3, VEGF, IL-2, and IL-4 for KOA detection

ROC curve analysis was conducted to assess the diagnostic performance of these biomarkers for KOA. Results were as follows: (1) STAT3: AUC of 0.710 (95% CI: 0.634-0.786, P < 0.001),

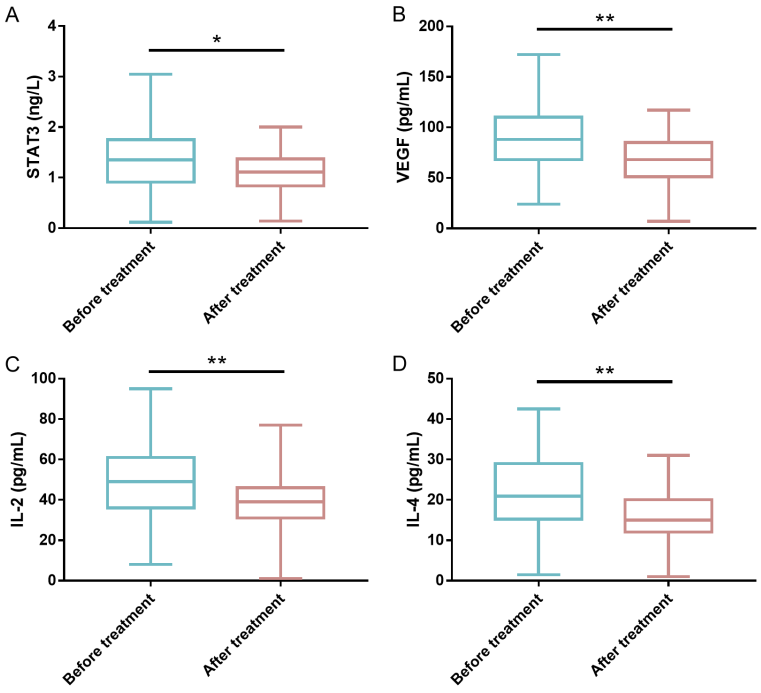

Figure 2. Comparison of serum STAT3, VEGF, IL-2, and IL-4 levels before and after treatment in KOA patients. A. Changes in serum STAT3 levels before and after treatment. B. Changes in serum VEGF levels before and after treatment. C. Changes in serum IL-2 levels before and after treatment. D. Changes in serum IL-4 levels before and after treatment. Notes: KOA, knee osteoarthritis; STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4. *$P < 0.05$, **$P < 0.01$.

sensitivity 98.00%, specificity 50.43%, cutoff 1.35 ng/L. (2) VEGF: AUC of 0.726 (95% CI: 0.651-0.802, P < 0.001), sensitivity 94.00%, specificity 45.22%, cutoff 91.50 pg/mL. (3) IL-2: AUC of 0.704 (95% CI: 0.624-0.783, P < 0.001), sensitivity 82.00%, specificity 56.52%, cutoff 46.50 pg/mL. (4) IL-4: AUC of 0.797 (95% CI: 0.731-0.863, P < 0.001), sensitivity 90.00%, specificity 70.43%, cutoff 16.56 pg/mL. (5) Combined biomarkers: AUC increased to 0.913 (95% CI: 0.872-0.955, P < 0.001), sensitivity 98.00%, specificity 80.87%, cutoff 0.27.

These findings are illustrated in **Figure 3** and detailed in **Table 2**.

Comparison of serum STAT3, VEGF, IL-2, and IL-4 levels among KOA severity groups

Serum biomarker levels were compared between mild and moderate-to-severe KOA groups, revealing significant differences: (1) STAT3: 1.07 ± 0.52 ng/L (mild) vs. 1.56 ± 0.61 ng/L (moderate-to-severe). (2) VEGF: 76.25 ± 23.06 pg/mL (mild) vs. 102.75 ± 32.67 pg/mL (moderate-to-severe). (3) IL-2: 39.31 ± 16.40 pg/mL (mild) vs. 55.76 ± 17.20 pg/mL (moderate-to-severe). (4) IL-4: 17.01 ± 9.11 pg/mL (mild) vs. 25.52 ± 7.89 pg/mL (moderate-to-severe).

All four biomarkers were significantly lower in mild cases compared to moderate-to-severe cases (all P < 0.001; **Figure 4**).

Correlations between serum biomarkers and KOA severity

To explore associations with disease severity, KOA severity was numerically coded (1 = mild, 2 = moderate, 3 = severe) for Spearman's correlation analysis. Significant positive correlations were identified between serum biomarker levels and KOA severity (all P < 0.001; **Figure 5**): STAT3 (r = 0.436), VEGF (r = 0.467), IL-2 (r = 0.497), and IL-4 (r = 0.509).

Comparison of diagnostic performance of serum STAT3, VEGF, IL-2, and IL-4 for KOA severity assessment

ROC analysis was conducted to evaluate the diagnostic utility of these biomarkers for assessing KOA severity: STAT3: AUC 0.723 (95% CI: 0.630-0.815, P < 0.001), sensitivity 60.32%, specificity 75.00%, cutoff 1.42 ng/L. VEGF: AUC 0.742 (95% CI: 0.649-0.836, P < 0.001), sensitivity 60.32%, specificity 92.31%, cutoff 101.50 pg/mL. IL-2: AUC 0.769 (95% CI: 0.682-0.857, P < 0.001), sensitivity 60.32%, specificity 82.69%, cutoff 50.50 pg/mL. IL-4: AUC 0.757 (95% CI: 0.667-0.847, P < 0.001), sensitivity 74.60%, specificity 71.15%, cutoff 20.30 pg/mL. Combined biomarkers: AUC 0.899 (95% CI: 0.845-0.952, P < 0.001), sensitivity 74.60%, specificity 90.38%, cutoff 0.69. These results are shown in **Figure 6** and detailed in **Table 3**.

Discussion

This study revealed significantly elevated serum levels of STAT3, VEGF, IL-2, and IL-4 in KOA patients compared to healthy controls, sug-

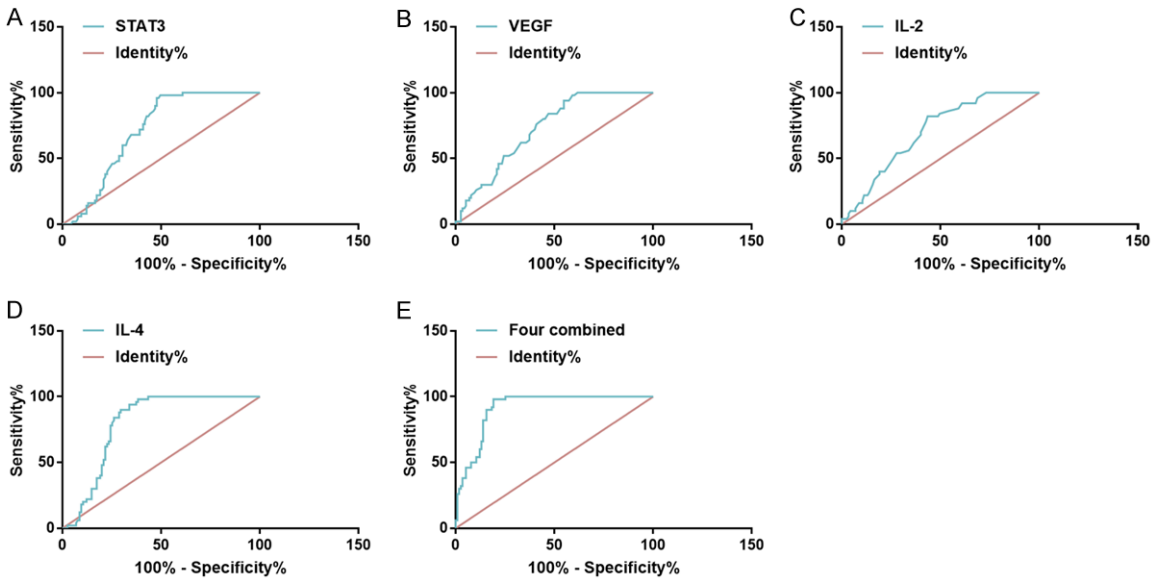

Figure 3. Diagnostic performance of serum STAT3, VEGF, IL-2, and IL-4 in KOA detection. A. ROC curve analysis for serum STAT3 in KOA detection. B. ROC curve analysis for serum VEGF in KOA detection. C. ROC curve analysis for serum IL-2 in KOA detection. D. ROC curve analysis for serum IL-4 in KOA detection. E. ROC curve analysis for the combined four biomarkers in KOA detection. Notes: STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4; KOA, knee osteoarthritis; ROC, receiver operating characteristic.

Table 2. ROC curve data for serum STAT3, VEGF, IL-2, and IL-4 in KOA diagnosis

Indicators	AUC	95% CI	*P* value	Specificity	Sensitivity	Cutoff
STAT3 (ng/L)	0.710	0.634-0.786	< 0.001	50.43%	98.00%	1.35 ng/L
VEGF (pg/mL)	0.726	0.651-0.802	< 0.001	45.22%	94.00%	91.50 pg/mL
IL-2 (pg/mL)	0.704	0.624-0.783	< 0.001	56.52%	82.00%	46.50 pg/mL
IL-4 (pg/mL)	0.797	0.731-0.863	< 0.001	70.43%	90.00%	16.56 pg/mL
Combined diagnosis	0.913	0.872-0.955	< 0.001	80.87%	98.00%	

Notes: ROC, receiver operating characteristic; STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4; KOA, knee osteoarthritis; AUC, area under the curve; CI, confidence interval.

gesting their potential involvement in the pathological processes underlying KOA onset and progression. Moreover, the marked reductions in these biomarkers following treatment imply their possible utility in evaluating therapeutic efficacy. STAT3 has been extensively studied in skeletal diseases, including osteoporosis and osteoarthritis, where it regulates key pathological processes such as inflammation, cartilage degradation, osteoclast activation, osteoblast differentiation, and macrophage polarization [20]. According to a systematic review by Chen et al. [21], targeting the Janus kinase 2 (JAK2)/STAT3 pathway, either directly or indirectly, may offer therapeutic benefits for KOA, though further clinical trials are needed for validation. Additionally, the STAT3/hypoxia-inducible factor 1 (HIF-1)/VEGF signaling axis has been associated with pain relief, reduced joint swelling, and anti-inflammatory effects, with its inhibition demonstrating protective effects against both rheumatoid arthritis and KOA [22].

IL-2 is known to play a dual role in inflammatory diseases, facilitating both immune activation and resolution [23]. Similarly, IL-4 functions as a regulatory cytokine in inflammatory arthritis and has been implicated in the inflammatory processes of rheumatoid arthritis [24]. In the present study, diagnostic evaluations of these biomarkers yielded AUC values ranging from 0.700 to 0.800, with sensitivities between 82.00% and 98.00%, and specificities between 45.22% and 70.43%. Notably, combining all

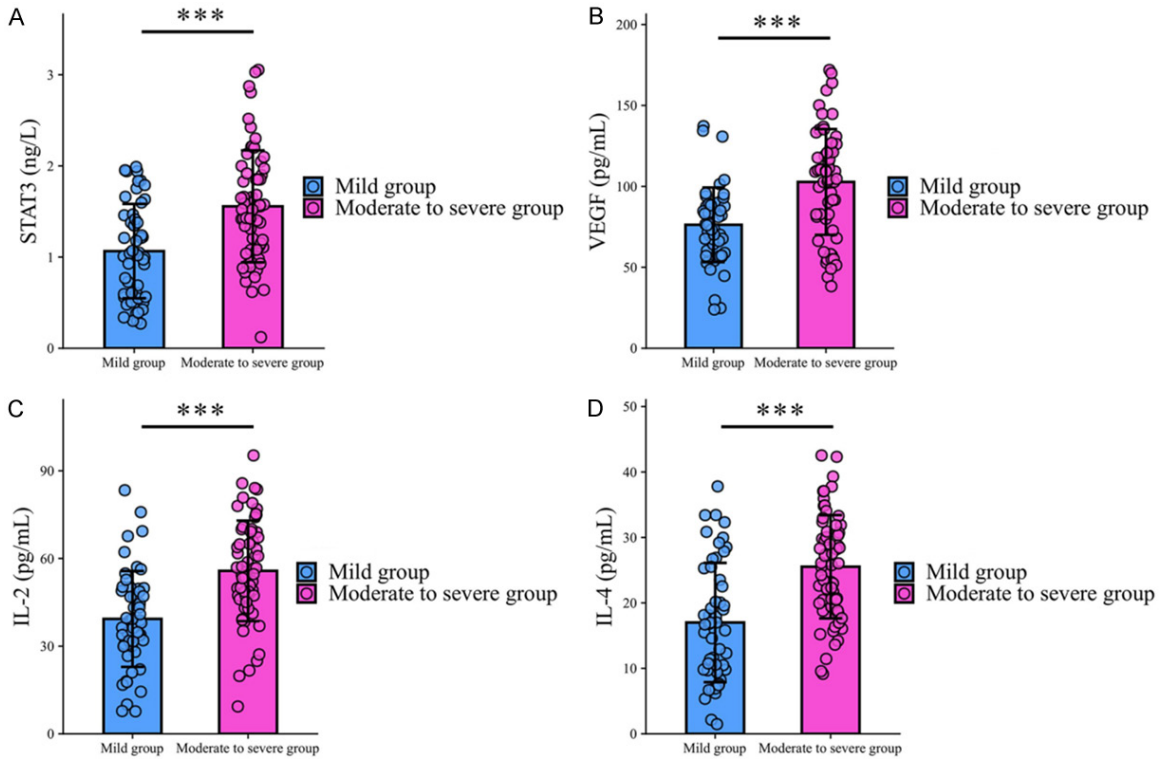

Figure 4. Serum levels of STAT3, VEGF, IL-2, and IL-4 in KOA severity groups. A. Comparison of serum STAT3 levels between mild and moderate-to-severe groups. B. Comparison of serum VEGF levels between mild and moderate-to-severe groups. C. Comparison of serum IL-2 levels between mild and moderate-to-severe groups. D. Comparison of serum IL-4 levels between mild and moderate-to-severe groups. Notes: STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4; KOA, knee osteoarthritis. ***P < 0.001.

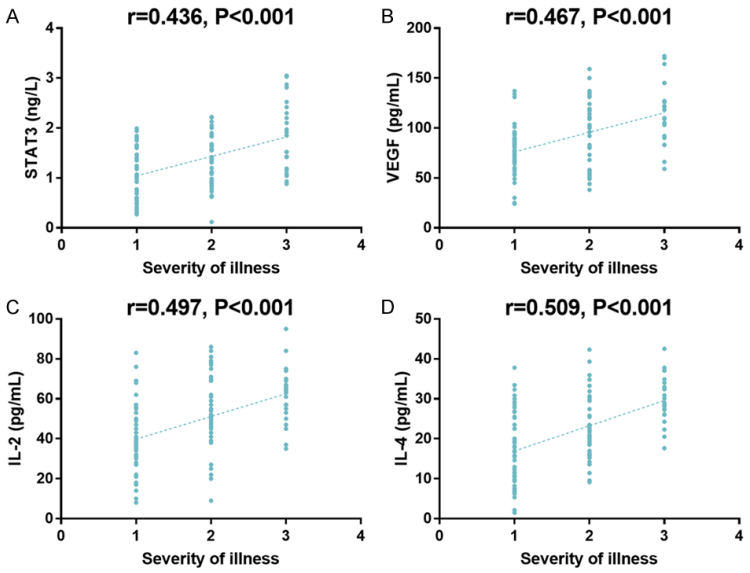

Figure 5. Correlations between serum STAT3, VEGF, IL-2, IL-4, and KOA severity. A. Correlations between serum STAT3 levels and KOA severity. B. Correlations between serum VEGF levels and KOA severity. C. Correlations between serum IL-2 levels and KOA severity. D. Correlations between serum IL-4 levels and KOA severity. Notes: STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4; KOA, knee osteoarthritis.

four biomarkers substantially improved diagnostic accuracy, achieving an AUC of 0.913 with specificity and sensitivity of 80.87% and 98.00%, respectively, and an optimal cutoff value of 0.27. Previous studies have similarly shown that IL-4 combined with IL-6 can predict prosthetic joint infections with an AUC of 0.962 [25]. Despite these promising results, the diagnostic utility of these biomarkers in KOA remains underexplored, necessitating further validation to establish their clinical applicability.

This study also demonstrated significantly higher serum levels of STAT3, VEGF, IL-2, and IL-4 in patients with moderate-to-severe KOA compared to those with mild disease, suggesting their involvement in disease

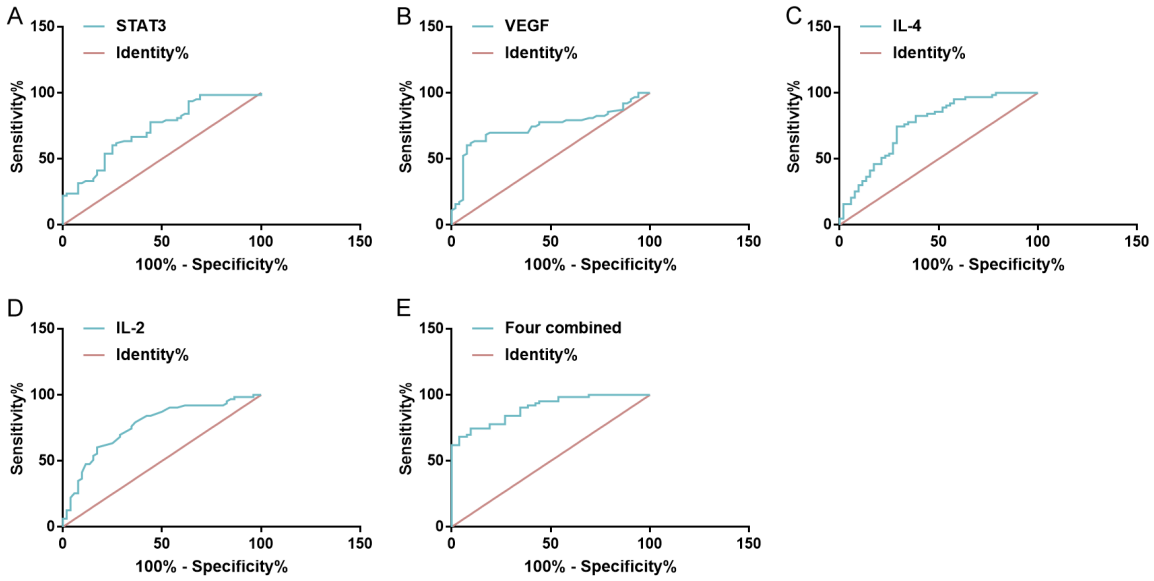

Figure 6. Diagnostic performance of serum STAT3, VEGF, IL-2, and IL-4 for KOA severity assessment. A. ROC curve for serum STAT3 in evaluating KOA severity. B. ROC curve for serum VEGF in evaluating KOA severity. C. ROC curve for serum IL-2 in evaluating KOA severity. D. ROC curve for serum IL-4 in evaluating KOA severity. E. ROC curve of the four parameters for KOA severity diagnosis. Notes: ROC, receiver operating characteristic; STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4; KOA, knee osteoarthritis.

Table 3. ROC curve data for serum STAT3, VEGF, IL-2, and IL-4 in evaluating KOA severity

Indicators	AUC	95% CI	*P* value	Specificity	Sensitivity	Cutoff
STAT3 (ng/L)	0.723	0.630-0.815	< 0.001	75.00%	60.32%	1.42 ng/L
VEGF (pg/mL)	0.742	0.649-0.836	< 0.001	92.31%	60.32%	101.50 pg/mL
IL-2 (pg/mL)	0.769	0.682-0.857	< 0.001	82.69%	60.32%	50.50 pg/mL
IL-4 (pg/mL)	0.757	0.667-0.847	< 0.001	71.15%	74.60%	20.30 pg/mL
Combined evaluation	0.899	0.845-0.952	< 0.001	90.38%	74.60%	

Notes: ROC, receiver operating characteristic; STAT3, signal transducer and activator of transcription 3; VEGF, vascular endothelial growth factor; IL-2/4, interleukin-2/4; KOA, knee osteoarthritis; AUC, area under the curve; CI, confidence interval.

progression. Furthermore, moderate positive correlations were identified between biomarker levels and KOA severity, indicating their potential in predicting disease progression. Wahba et al. [26] reported that the STAT3 rs1053005 variant is closely associated with KOA susceptibility and severity. Similarly, Wang et al. [27] observed that synovial VEGF levels increased with cartilage damage severity, highlighting its role as a progression marker. Supporting this, Saetan et al. [28] found positive correlations between VEGF levels in plasma and synovial fluid and radiographic KOA severity. Additionally, Vangsness Jr et al. [29] reported that elevated cytokine levels, including IL-2, were associated with KOA progression, consistent with our findings.

Further analysis showed that AUC values for predicting KOA severity using these biomarkers ranged from 0.720 to 0.770. Among them, VEGF exhibited the highest specificity (92.31%), while IL-4 showed the highest sensitivity (74.60%). When all four biomarkers were combined, predictive performance improved significantly, with an AUC of 0.899, specificity of 90.38%, sensitivity of 74.60%, and an optimal cutoff value of 0.69.

This study has several limitations. First, the lack of follow-up data prevents assessment of the prognostic value of serum STAT3, VEGF, IL-2, and IL-4 levels, warranting future longitudinal studies to establish their predictive potential for disease outcomes. Second, the underly-

ing regulatory mechanisms of these biomarkers in KOA pathogenesis were not investigated through in vitro or in vivo models; further mechanistic studies could elucidate disease pathways and identify novel therapeutic targets. Third, the study did not compare these biomarkers against a gold standard diagnostic method, limiting the assessment of their consistency and practical utility. Incorporating such comparisons in future studies would enhance the comprehensiveness of diagnostic workflows.

In conclusion, KOA is associated with abnormally elevated serum levels of STAT3, VEGF, IL-2, and IL-4, which may serve as auxiliary indicators for treatment efficacy, diagnosis, and severity assessment. A combined biomarker panel enhances diagnostic sensitivity and predictive accuracy. These findings provide robust indicators for the clinical evaluation and monitoring of KOA and lay the theoretical groundwork for the development of new therapeutic targets.

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Disclosure of conflict of interest

None.

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