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ORIGINAL ARTICLE

Observational Study

Changes of serum inflammatory factors and miR-145 expression in patients with osteoarthritis before and after treatment and their clinical value

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Abstract

BACKGROUND

Osteoarthritis is a chronic degenerative disease with an incidence of 50% in people over 65 years old and 80% in people over 80 years old worldwide. It is the second leading reason of loss of working capacity after cardiovascular diseases and severely affects the society and families. Therefore, finding biological markers related to the diagnosis and treatment of osteoarthritis is of great significance in clinical practice.

AIM

To observe the changes and clinical value of serum inflammatory factors and miR-145 expression in patients with osteoarthritis before and after treatment.

METHODS

Eighty-three patients with knee osteoarthritis (observation group) who were admitted to our hospital from April 2013 to June 2015, and 60 healthy people (control group) during the same period were selected. After 4 wk of treatment, the levels of miR-145, tumor necrosis factor (TNF)-α, interleukin (IL)-6, and IL-10 were compared between the control group and the observation group before treatment. The correlation of miR-145, TNF-α, IL-6, and IL-10 levels with visual analogue scale (VAS), Lysholm, and Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) scores was assessed by Pearson correlation analysis. The correlation of the expression of miR-145, TNF-α, IL-6, and IL-10 with Kellgren-Lawrence (K-L) grades was assessed by Spearman correlation analysis. The critical levels of miR-145, TNF-α, IL-6, and IL-10 in distinguishing different K-L grades were determined by receiver operating characteristic (ROC) curve analysis.

RESULTS

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The expression level of miR-145 in the observation group was significantly higher than that in the control group before treatment ($P < 0.05$). After treatment, the expression level of miR-145 in the observation group was significantly lower than that before treatment ($P < 0.05$). The levels of TNF-α and IL-6 in the observation group were significantly higher than those in the control group ($P < 0.05$), and the level of IL-10 was significantly lower than that in the control group ($P < 0.05$). After treatment, the levels of TNF-α and IL-6 in the observation group were significantly lower than those before treatment ($P < 0.05$), and IL-I0 level was significantly higher than that before treatment ($P < 0.05$). VAS and WOMAC scores were both positively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and negatively correlated with IL-10 ($P < 0.05$), while Lysholm scores were negatively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and positively correlated with IL-10 ($P < 0.05$). K-L grades were positively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and negatively correlated with IL-10 ($P < 0.05$). The area under the ROC curve (AUC) and specificity of TNF-α in differentiating K-L grades I-II were the highest, which were 0.785 and 97.45%, respectively, and miR145 had the highest sensitivity of 94.59%; the AUC and sensitivity of IL-6 in differentiating K-L grades II-III were the highest, which were 0.766 and 97.30%, respectively, and TNF-α had the highest specificity of 86.68%.

CONCLUSION
MiR-145 and inflammatory factors have certain diagnostic value in osteoarthritis, and they are expected to become potential indicators for the diagnosis and evaluation of osteoarthritis in the future.

Key words: Osteoarthritis; Treatment; Inflammatory factor; miR-145

Core tip: At present, the clinical evaluation of osteoarthritis relies on VAS score, Lysholm score, and WOMAC score and other scales. These scales have a common deficiency, *i.e.*, they are affected by a certain degree of subjective factors. Many clinical workers use Kellgren-Lawrence (K-L) grading to evaluate the therapeutic effect of patients with osteoarthritis, but the K-L grading depends on imaging, the effect of radiation on the human body and the medical expenses are high, and it takes a long time. This study explored the clinical value of miR-145 and inflammatory factors in peripheral blood, and explored the application of serological analysis in the diagnosis and treatment of osteoarthritis, so as to provide a reference for the diagnosis and treatment of osteoarthritis.

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INTRODUCTION

Osteoarthritis is a chronic degenerative disease characterized by cartilage erosion and lesions in the subchondral bone and other joint tissues, mostly in the hip and knee joints[1,2]. According to epidemiological statistics of osteoarthritis, the incidence of osteoarthritis in people over 65 years old can reach 50%, and the incidence of osteoarthritis in people over 80 years old is as high as 80%[3]. Osteoarthritis is the second most important cause of patients' loss of ability after cardiovascular diseases, seriously affecting the society and families[4,5].

Ultrasound is widely used in the diagnosis and efficacy evaluation of various joint diseases, which has good repeatability and is closely related to the Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) score. However, its operation requires experienced and skilled imaging doctors and high-resolution ultrasound equipment[6-8]. Therefore, finding osteoarthritis-related biomarkers has

become a research focus in recent years.

MicroRNAs (miRNAs) are a class of non-coding small RNAs belonging to miRNA-induced silencing complexes[9]. In previous studies, it was found that the occurrence and development of osteoarthritis are accompanied by changes in various miRNAs. Santini *et al*[10] reported that miR-149 is down-regulated in osteoarthritis, which is associated with increased expression of pro-inflammatory cytokines, such as tumor necrosis factor (TNF)-α, interleukin (IL)-1β, and IL-6. Philipot *et al*[11] found that miR-24 negatively regulates the p16INK4a/MMP1 axis in osteoarthritis, which is involved in the senescence of inflammatory chondrocytes and matrix remodeling related to terminal differentiation. In recent years, it has been reported that miR-145 can regulate the function of cartilage cells and promote cartilage recovery[12]. In a related report on miRNA screening in osteoarthritis, miR-145 was found to increase in osteoarthritis[13]. These studies suggest that miR-145 plays an important role in the development of osteoarthritis. However, there are few reports on the value of miR-145 in the clinical application of osteoarthritis.

This study aimed to observe the changes and clinical value of serum inflammatory factors and miR-145 expression in patients with osteoarthritis before and after treatment, and provide a theoretical and experimental basis for clinical treatment of hip osteoarthritis.

MATERIALS AND METHODS

Materials

Eighty-three patients with knee osteoarthritis (observation group; aged 42-70 years old) admitted to our hospital from April 2013 to June 2015, and 60 healthy people (control group; aged 42-70 years old) during the same period were selected. The inclusion criterion was as follows: (1) All patients in the observation group met the 2007 osteoarthritis diagnostic guidelines[14], and knee osteoarthritis was treated according to the OARSI guidelines[15]; and (2) All patients in the observation group underwent X-ray radiology diagnosis and performed Kellgren-Lawrence (K-L) radiation grading. The exclusion criteria were as follows: (1) K-L grade IV in the observation group; (2) Patients with cataclasis, other systemic inflammatory syndrome, severe endocrine dysfunction, pregnant women, cardiovascular, liver and kidney diseases, neurological or psychiatric history, chronic pain syndrome, language communication difficulties, joint bone tumors, various cancer bone metastases, recent acute stroke, autoimmune diseases, or other inflammatory arthritis; and (3) The control group had obvious clinical disease characteristics, pregnant women, psychiatric history, and language communication disorders. This study was approved by the Hospital Ethics Committee. All patients and their families signed an informed consent form.

Observation indexes

Before treatment and after 4 wk of treatment, the levels of miR-145, TNF-α, IL-6, and IL-I0 were compared between the control group and observation group. The correlation of miR-145, TNF-α, IL-6, and IL-10 levels with visual analogue scale (VAS), Lysholm, and WOMAC scores was assessed by Pearson correlation analysis. The correlation of the expression of miR-145, TNF-α, IL-6, and IL-10 with K-L grades was assessed by Spearman correlation analysis. The critical levels of miR-145, TNF-α, IL-6, and IL-10 in distinguishing different K-L grades were determined by receiver operating characteristic (ROC) curve analysis.

Quantitative real-time PCR (qRT-PCR)

Peripheral blood was collected from all patients before and after treatment, and serum was taken after centrifugation. Total RNA in serum was extracted with TRIzol (Guangzhou Labgene Biotechnology Co., Ltd.) following the manufacturer's protocol. A micro-ultraviolet spectrophotometer DanoProp1000 (Thmorgan Biotechnology Co., Ltd.) was used to analyze the concentration and purity of the extracted RNA, and 3% agarose gel electrophoresis (gel electrophoresis kit purchased from Shanghai JingkeChemical Technology Co., Ltd.) was used to analyze the integrity of RNA. The A260/A280 value between 1.8 and 2.1 was considered to meet the experimental requirements. After RNA extraction was completed, qRT-PCR reaction was carried out. Reverse transcription was performed in a 20-μL system consisting of 4 μL of 5 × TransScript All in One SuperMix for PCR, 2 μg of total RNA, and appropriate amount of ribonuclease free distilled water. The reaction conditions were 25 °C for 10 min, 42 °C for 30 min, and inactivation of reverse transcriptase at 85°C for 5 s. After reverse transcription, PCR amplification was carried out in a 50 μL amplification system

consisting of 2 μL of cDNA template, 25 μL of 2 × TransTaq HiFi PCR SuperMix II, 1 μL of upstream primer and downstream primer, and appropriate amount of double distilled water. The cycling parameters were pre-denaturation at 95°C for 3 min, 40 cycles of 94 °C for 2 min, 94 °C for 30 s, 55 °C for 30 s, 72°C 1-2 kb/min, and extension for 5 min at 72 °C. U6 was used as the internal reference of the reaction, and the results were analyzed by the 2^{-Ct} method. TransScript Two-Step RT-PCRSuperMix was purchased from Beijing TransGen Biotech, with the catalog number of AT401-0l, and the primer sequence was designed and synthesized by Hepeng (Shanghai) Biotechnology Co., Ltd. (Table 1).

Enzyme-linked immunosorbent assay (ELISA)

Peripheral blood was collected from all patients before and after treatment, and serum was taken after centrifugation. The levels of TNF-α, IL-6, and IL-10 were detected by ELISA using kits purchased from Abcam (ab181421, ab178013, and ab46601) following the kit instructions.

Statistical analysis

SPSS 22.0 software (Asia Analytics Formerly SPSS, China) was used in this study for statistical analyses. Measurement data are expressed as the mean ± SD, and count data are expressed as percentages. Count data were compared by χ^2 tests, and measurement data were compared by the independent sample *t*-test between two groups. The correlation of miR-145, TNF-α, IL-6, and IL-10 levels with VAS, Lysholm, and WOMAC scores was assessed by Pearson correlation analysis. The correlation of the expression of miR-145, TNF-α, IL-6, and IL-10 with K-L grades was assessed by Spearman correlation analysis. The critical levels of miR-145, TNF-α, IL-6, and IL-10 in distinguishing different K-L grades were determined by ROC curve analysis. Statistical significance was indicated by $P < 0.05$.

RESULTS

General data

There were 60 patients in the control group, including 33 males (55.00%) and 27 females (45.00%), with a mean age of 54.76 ± 13.64 years old and a mean body mass index (BMI) of 23.53 ± 3.06 kg/m^2. There were 83 patients in the observation group, including 52 males (62.65%) and 31 females (37.35%), with a mean age of 57.48 ± 12.49 years old and a mean BMI of 24.77 ± 4.14 kg/m^2. There was no significant difference in sex ratio, age, or BMI between the two groups ($P > 0.05$). Other basic data of the observation group are shown in Table 2.

Difference of miR-145 expression in peripheral blood between the two groups

The expression level of miR-145 in the observation group was significantly higher than that in the control group before treatment ($P < 0.05$) (Figure 1A). The expression level of miR-145 in the observation group after treatment was significantly lower than that before treatment ($P < 0.05$) (Figure 1B).

Difference analysis of inflammatory factors in peripheral blood between the two groups

The levels of TNF-α and IL-6 in the observation group were significantly higher than those in the control group ($P < 0.05$), and the level of IL-10 was significantly lower than that in the control group ($P < 0.05$) (Figure 2A). The levels of TNF-α and IL-6 in the observation group after treatment were significantly lower than those before treatment ($P < 0.05$), and IL-10 level was significantly higher than that before treatment ($P < 0.05$) (Figure 2B).

Correlation of miR-145 and inflammatory factors with VAS scores

VAS scores were positively correlated with miR-145, TNF-α, and IL-6, and negatively correlated with IL-10 ($P < 0.05$) (Figure 3).

Correlation of miR-145 and inflammatory factors with Lysholm scores

Lysholm scores were negatively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and positively correlated with IL-10 ($P < 0.05$) (Figure 4).

Correlation of miR-145 and inflammatory factors with WOMAC scores

WOMAC scores were positively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and negatively correlated with IL-10 ($P < 0.05$) (Figure 5).

Correlation of miR-145 and inflammatory factors with K-L grades

Table 1 Sequence of primers

	Forward primer	Reverse primer
MiR-145	5'-GTCCAGTTTTCCCAGGAATCCCT- 3'	5'-GCTGTCAACATACGCTACGTAACG- 3'
U6	5'-GCGCGTCGTGAAGCGTTC- 3'	5'-GTGCAGGGTCCGAGGT- 3'

L grades were positively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and negatively correlated with IL-10 ($P < 0.05$) (Figure 6).

ROC analysis of miR-145 and inflammatory factors in differentiating K-L grades

The area under the ROC area (AUC), critical level, sensitivity, and specificity of miR-145 in distinguishing K-L grades I and II were 0.762, 1.356, 94.59%, and 51.61%, respectively, and 0.587, 1.475, 32.43%, and 86.67% for grades II and III (Figure 7A). The AUC, critical level, sensitivity, and specificity of TNF-α in differentiating K-L grades I and II were 0.785, 142.00 ng/L, 59. 46%, and 97.45%, respectively, and 0.631, 167.80 ng/L, 48.65%, and 86.68% for grades II and III (Figure 7B). The AUC, critical level, sensitivity, and specificity of IL-6 in distinguishing K-L grades I and II were 0.758, 13.06 μg/L, 62. 16%, and 90.32%, respectively, and 0.766, 19.62 μg/L, 97.30%, and 46.67% for grades II and III (Figure 7C). The AUC, critical level, sensitivity, and specificity of IL-10 in differentiating K-L grades I and II were 0.767, 64.37 pg/mL, 67.57%, and 90.32%, respectively, and 0.663, 51.43 pg/mL, 75.68%, and 60.00% for grades II and III (Figure 7D) (Table 3).

DISCUSSION

At present, the clinical evaluation of osteoarthritis relies on VAS score, Lysholm score, and WOMAC score and other scales[16-18]. These scales have a common deficiency, *i.e.*, they are affected by a certain degree of subjective factors. Many clinical workers use K-L grading to evaluate the therapeutic effect of patients with osteoarthritis[19], but the K-L grading depends on imaging, the effect of radiation on the human body and the medical expenses are high, and it takes a long time[20]. This study explored the clinical value of miR-145 and inflammatory factors in peripheral blood, and explored the application of serological analysis in the diagnosis and treatment of osteoarthritis, so as to provide a reference for the diagnosis and treatment of osteoarthritis.

We first analyzed the changes of miR-145 and inflammatory factor levels in peripheral blood of healthy people and patients with knee osteoarthritis before and after treatment. It is clear that miR-145 is elevated in patients with knee osteoarthritis. And after treatment, the levels of inflammatory factors are consistent with previous reports[21,22]. TNF-α and IL-6 levels in peripheral blood of patients with knee osteoarthritis increased, while IL-10 levels decreased. After treatment, the levels decreased and increased accordingly. At present, there are few clinical studies on the correlation between miR-145 and osteoarthritis. From our analysis, miR-145, TNF-α, IL-6, and IL-10 had a linear correlation with VAS, Lysholm, and WOMAC scores, suggesting that these serological factors are closely related to the clinical efficacy of osteoarthritis. But the degree of correlation is mostly moderate or low, suggesting that there are still shortcomings in the use of miR-145 and inflammatory factors to evaluate the therapeutic effect. The pathogenesis of osteoarthritis is complex, especially primary osteoarthritis[23]. At present, the cause of osteoarthritis is not clear, and it may be related to the age span of our subjects and the long course of osteoarthritis[24]. Moreover, we have not further analyzed the efficacy of miR-145 and inflammatory factors in primary osteoarthritis and secondary osteoarthritis, because the specific reasons need to be further analyzed. In recent years, many scholars have verified the mechanism of action of miR-145 in osteoarthritis, such as the inhibitory effect of miR-145 targeting TNFRSF11B on proliferation and fibrosis of human osteoarthritis cartilage cells[25]. MiR-145 inhibits cartilage matrix degradation driven by TNF-α in osteoarthritis by directly inhibiting mitogen-activated protein kinase 4[26]. And researchers reported that down-regulation of miR-145 expression can promote apoptosis of cartilage cells in osteoarthritis[27], and miR-145 knockout can also inhibit the proliferation/differentiation of osteoclasts, induce osteoclast apoptosis, limit bone excessive absorption, and finally reduce osteoarthritis by up-regulating the expression of osteopontin[28,29]. These reports also suggested the complex role of miR-145 in osteoarthritis. Therefore, if miR-145 is used to evaluate the clinical efficacy of osteoarthritis, the time point of blood collection needs to be further clarified. We will design more rigorous and detailed experimental steps in the future research.

Table 2 General information

	Control group (*n* = 60)	Observation group (*n* = 83)	χ^2/t	*P*-value
Sex, *n* (%)			0.846	0.358
Male	33 (55.00)	52 (62.65)		
Female	27 (45.00)	31 (37.35)		
Age (yr)	54.76 ± 13.64	57.48 ± 12.49	1.236	0.218
BMI (kg/m^2)	23.53 ± 3.06	24.77 ± 4.14	1.964	0.052
Course of disease (yr)		3.25 ± 1.37		
K-L grade, *n* (%)				
I		31 (37.35)		
II		37 (44.58)		
II		15 (18.07)		
VAS score				
Before treatment		5.49 ± 1.27		
After treatment		2.53 ± 088[a]		
Lysholm score				
Before treatment		58.45 ± 10.62		
After treatment		75.26 ± 9.46[a]		
WOMAC score				
Before treatment		50.46 ± 9.14		
After treatment		18.86 ± 8.38[a]		
Disease site				
Left knee		40 (48.19)		
Right knee		31 (37.35)		
Both knees		12 (14.46)		

[a]$P < 0.05$ *vs* before treatment in the same group.

K-L grade is an important index for the diagnosis and grading of osteoarthritis. We analyzed the relationship of miR-145, TNF-α, IL-6, and IL-10 with K-L grades. K-L grades were positively correlated with miR-145, TNF-α, and IL-6, and negatively correlated with IL-10. ROC analysis showed that miR-145, TNF-a, IL-6, and IL-10 can better distinguish K-L grades I and II and grades II and III, and all the AUCs exceeded 0.5, although they had the highest discrimination value for K-L grades I and II. The above results also suggested that miR-145, TNF-α, IL-6, and IL-10 are closely related to the imaging findings in patients with osteoarthritis. In previous reports, the relationship between IL-6 and K-L grades was analyzed. Shimura *et al*[30] reported that there was a negative correlation between the levels of IL-6 in synovium and K-L grades in patients with knee osteoarthritis, which is completely contrary to our results. Shimura *et al*[31] reported that there was no significant difference in serum IL-6 levels between early stage (K-L grade 2) and advanced stage (K-L grades 3 and 4) in patients with knee osteoarthritis, but some studies have reported that IL-6 levels in peripheral blood increase with the severity of knee osteoarthritis[32]. They have not further analyzed or speculated on this phenomenon, so we can only speculate that this is related to the difference in the subjects of the study and the origin of the specimens. Therefore, more research is needed to further confirm this result.

In summary, miR-145 and inflammatory factors have certain diagnostic value in osteoarthritis, and they are expected to become potential indicators for future diagnosis and evaluation of osteoarthritis.

Table 3 Receiver operating characteristic curve analysis of miR-145 and inflammatory factors in differentiating Kellgren-Lawrence grades

	miR-145	TNF-α	IL-6	IL-10
I-II				
AUC	0.762	0.785	0.758	0.767
Critical level	1.356	142.00 ng/L	13.06 μg/L	64.37 pg/mL
Sensitivity	94.59%	59.46%	62.16%	67.57%
Specificity	51.61%	97.45%	90.32%	90.32%
II-III				
AUC	0.587	0.631	0.766	0.663
Critical level	1.475	167.80 ng/L	19.62 μg/L	51.43 pg/mL
Sensitivity	32.43%	48.65%	97.30%	75.68%
Specificity	86.67%	86.68%	46.67%	60.00%

AUC: Area under the curve; TNF: Tumor necrosis factor; IL: Interleukin.

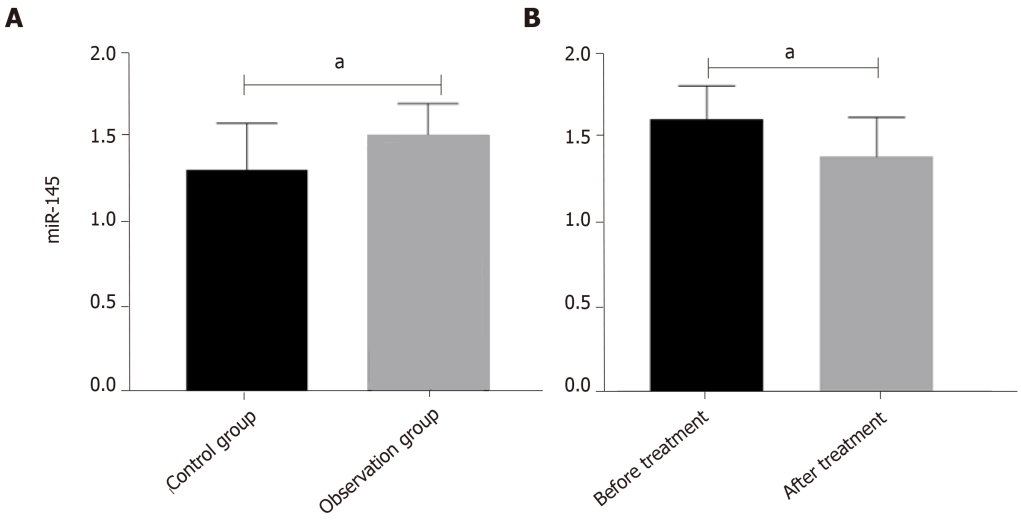

Figure 1 Expression levels of miR-145 in the peripheral blood of the two groups. $^{a}P < 0.05$.

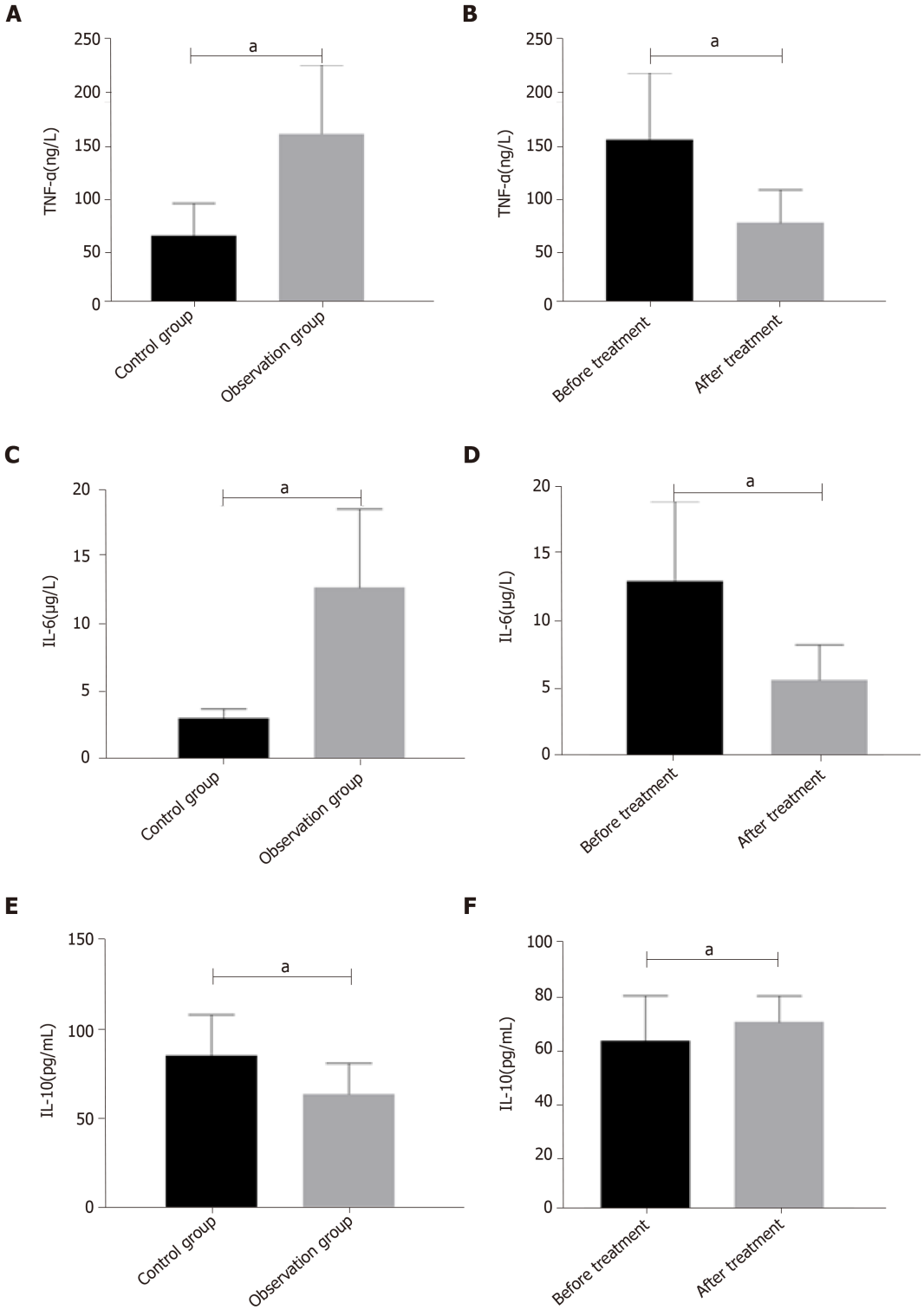

Figure 2 Levels of inflammatory factors in peripheral blood between the two groups. [a]*P* < 0.05.

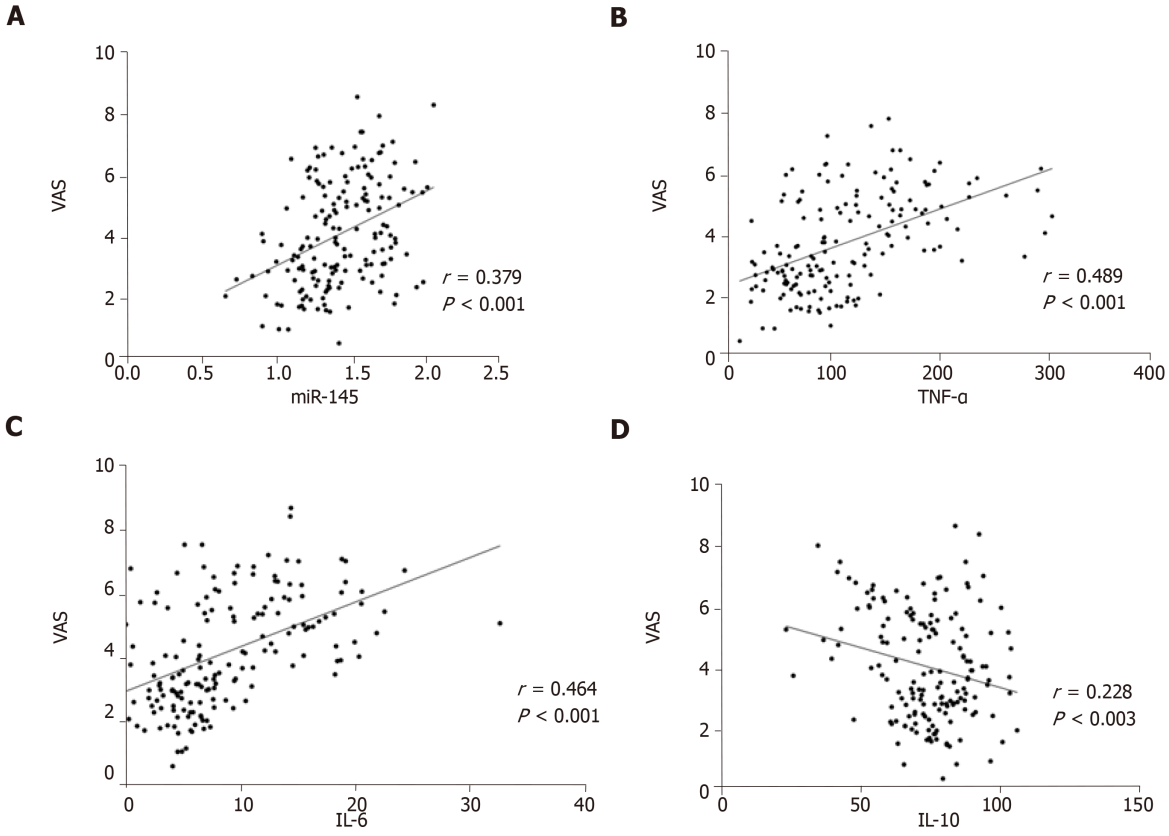

Figure 3 Correlation of miR-145 and inflammatory factors with VAS scores. VAS scores were positively correlated with miR-145, TNF-α, and IL-6, and negatively correlated with IL-10 ($P < 0.05$). TNF: Tumor necrosis factor; IL: Interleukin.

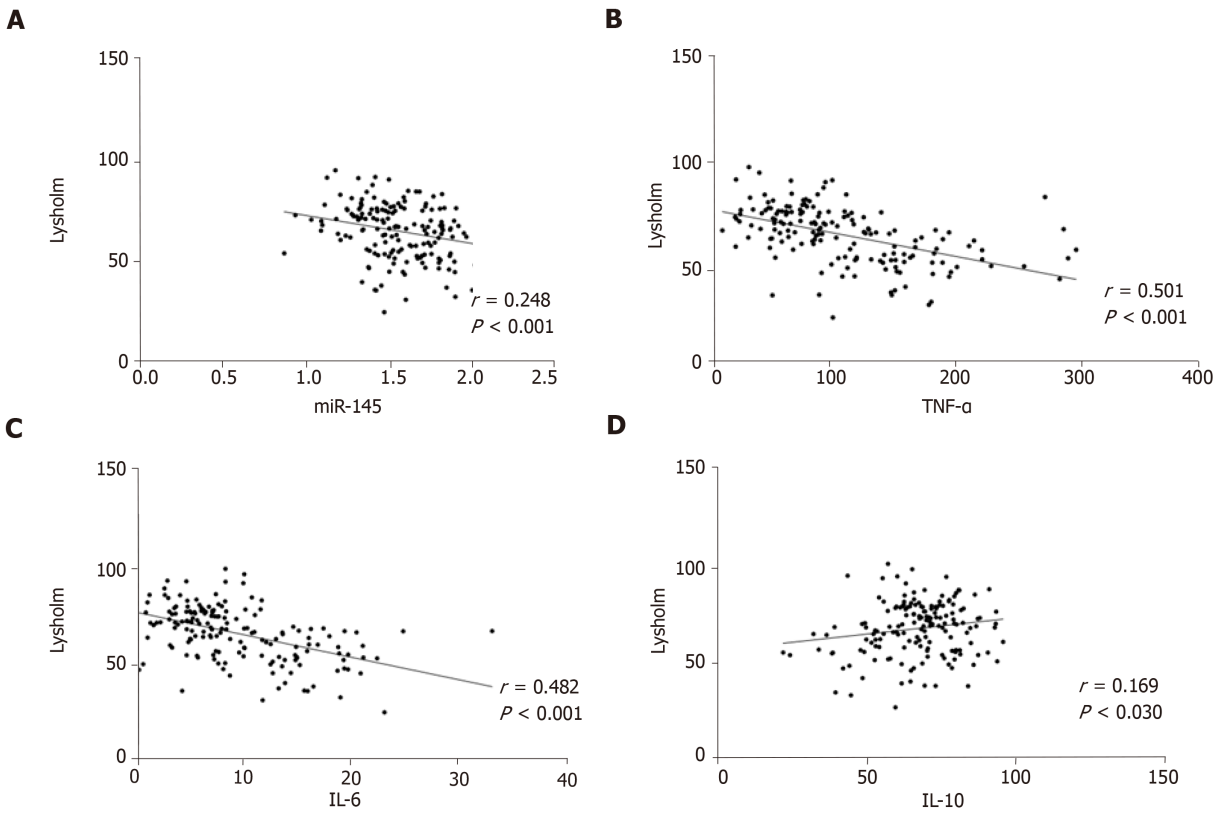

Figure 4 Relationship of miR-145 and inflammatory factors with Lysholm scores. Lysholm scores were negatively correlated with miR-145, TNF-α, and IL-6, and positively correlated with IL-10 ($P < 0.05$). TNF: Tumor necrosis factor; IL: Interleukin.

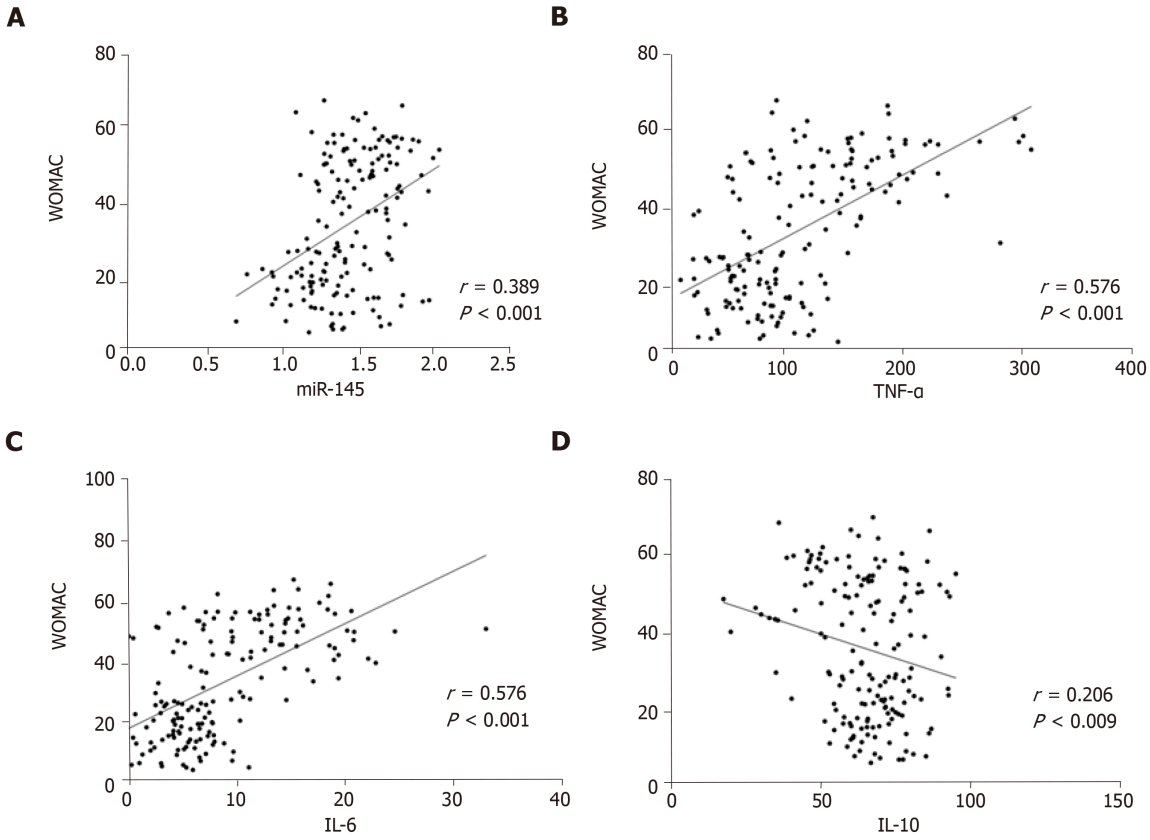

Figure 5 Correlation of miR-145 and inflammatory factors with WOMAC scores. WOMAC scores were positively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and negatively correlated with IL-10 ($P < 0.05$). TNF: Tumor necrosis factor; IL: Interleukin.

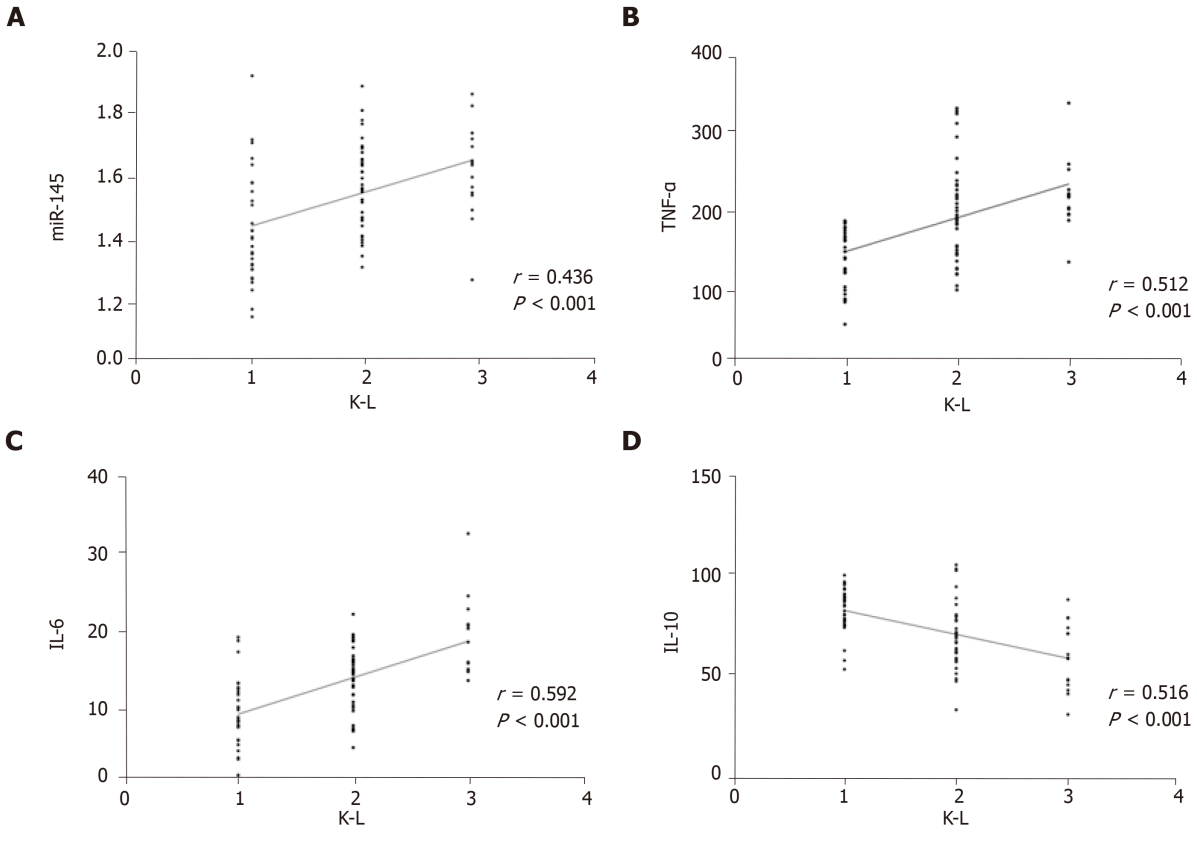

Figure 6 Correlation of miR-145 and inflammatory factors with Kellgren-Lawrence grades. Kellgren-Lawrence grades were positively correlated with miR-145, TNF-α, and IL-6 ($P < 0.05$), and negatively correlated with IL-10 ($P < 0.05$). TNF: Tumor necrosis factor; IL: Interleukin.

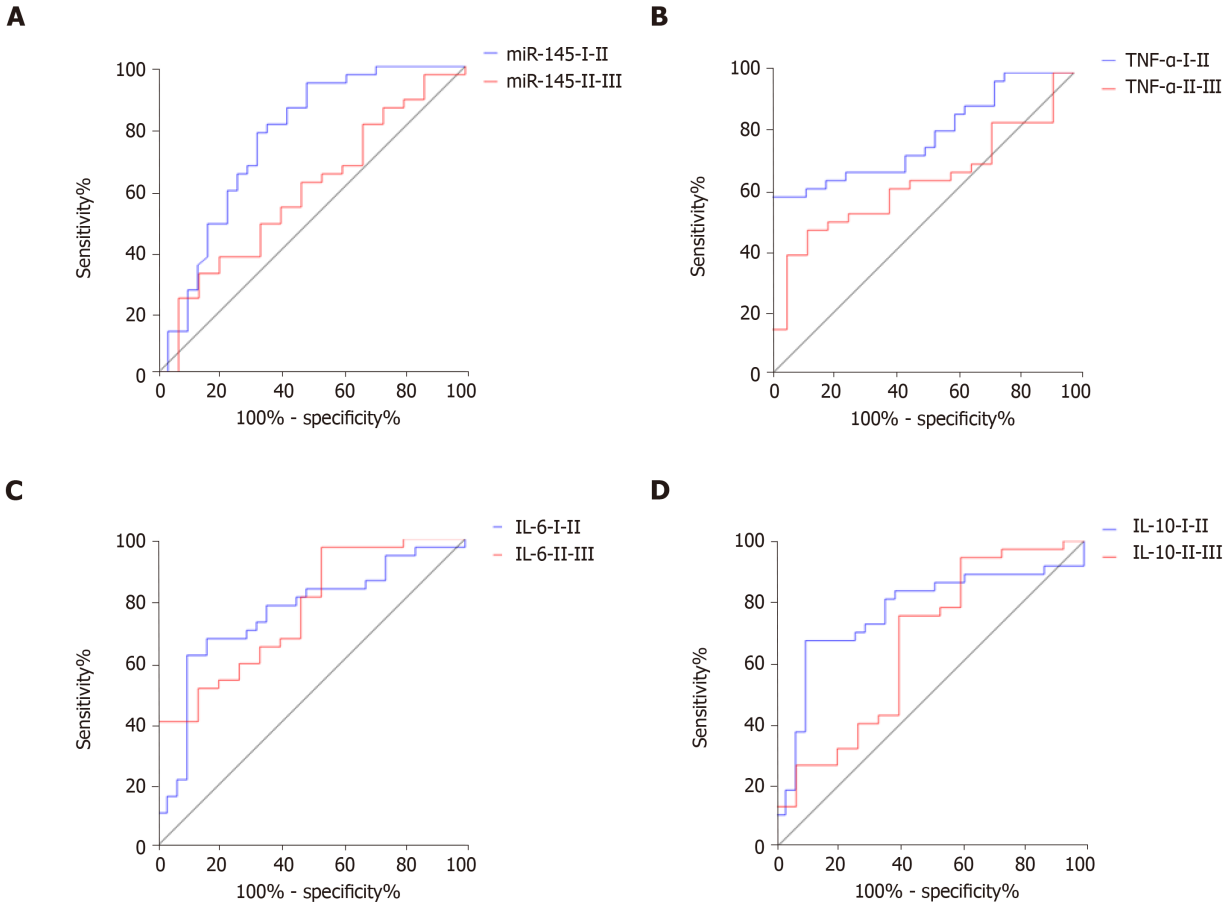

Figure 7 Receiver operating characteristic curve analysis of miR-145 and inflammatory factors in differentiating Kellgren-Lawrence grades.

ARTICLE HIGHLIGHTS

Research background

Osteoarthritis is the most common musculoskeletal disease in the world, which is characterized by articular cartilage degeneration, subchondral sclerosis, and narrowing of joint space. It is the sixth leading cause of disability worldwide and is expected to rise to the fourth by 2020, causing serious impact on the society and families. Therefore, finding biological markers related to the diagnosis and treatment of osteoarthritis is of great significance in clinical practice.

Research motivation

At present, the clinical efficacy evaluation for osteoarthritis mostly depends on VAS, Lysholm, and WOMAC scores, all of which are easily affected by subjective factors. Kellgren-Lawrence (K-L) classification is usually employed to assess the therapeutic effect of osteoarthritis patients as well, but it has the disadvantages of radiation, high cost, and being time-consuming. This study explored the application of serological analysis in diagnosis and treatment of osteoarthritis by analyzing the diagnostic and therapeutic value of miR-145 and inflammatory factors in peripheral blood of patients, so as to provide a reference for clinical diagnosis and treatment of osteoarthritis.

Research objectives

The study aimed to explore the application of serological analysis in the diagnosis and treatment of osteoarthritis by analyzing the diagnostic and therapeutic value of miR-145 and inflammatory factors in peripheral blood of patients, so as to provide a reference for clinical diagnosis and treatment of osteoarthritis.

Research methods

Eighty-three patients with knee osteoarthritis admitted to our hospital from April 2013 to June 2015 and 60 healthy subjects were enrolled in our study. The expression of miR-145 in the peripheral blood in the two groups was detected by qRT-PCR, and the changes of miR-145, tumor necrosis factor (TNF)-α, interleukin (IL)-6, IL-10, as well as VAS, Lysholm, and WOMAC scores after 4 weeks of treatment were compared between the two groups. Furthermore, Pearson correlation analysis, Spearman correlation analysis, and ROC curve analysis were used to analyze the relationship of miR-145, TNF-α, IL-6, and IL-10 levels with VAS, Lysholm, and

WOMAC scores to verify the application value of miR-145 and inflammatory factors in the diagnosis and treatment of osteoarthritis.

Research results

This study found that the expression of miR-145 and inflammatory factors increased in patients with knee osteoarthritis, with corresponding decrease and increase after treatment. VAS, Lysholm, and WOMAC scores were significantly improved after treatment, suggesting that the treatment was effective and the reduction of miR-145 and inflammatory factors after treatment might be related to the improvement of the patient's condition. Pearson correlation analysis showed that VAS, Lysholm, and WOMAC scores were either positively or negatively correlated with miR-145 and inflammatory factors, which verified our previous speculation. Moreover, the changes of miR-145 and inflammatory factors were related to the improvement of the patient's condition, indicating that miR-145 and inflammatory factors might be the potential efficacy predictors for osteoarthritis. This study also revealed that miR-145 and inflammatory factors were closely related to K-L classification, which suggested that miR-145 and inflammatory factors are valuable in determining the severity of osteoarthritis, and might be potential biological markers for osteoarthritis diagnosis in the future.

Research conclusions

The levels of miR-145 and inflammatory factors (TNF-α, IL-6, and IL-10) in peripheral blood of patients with osteoarthritis are significantly higher than those in healthy individuals, and increase with the severity of the disease, suggesting that miR-145 and inflammatory factors might be objective indicators to assess the progression of osteoarthritis. After treatment, the levels of miR-145 and inflammatory factors change significantly in patients with osteoarthritis, and are closely related to therapeutic effect-related indicators (VAS, Lysholm, and WOMAC scores). Therefore, miR-145 and inflammatory factors have potential value for evaluating the therapeutic effect on osteoarthritis. This study demonstrates that miR-145 and inflammatory factors are valuable in the diagnosis and treatment of osteoarthritis, which are expected to become potential indicators for future diagnosis and efficacy evaluation.

Research perspectives

This study reveals the potential role of miR-145 and inflammatory factors in the diagnosis and treatment of osteoarthritis. For the better use of miR-145 and inflammatory factors in clinical practice, the blood sampling time point needs further confirmation. Besides, whether miR-145 can become a target for osteoarthritis treatment needs further analysis. The mechanism of action of miR-145 on the occurrence and development of osteoarthritis needs to be verified by *in vitro* cell experiments and *in vivo* animal experiments to provide an experimental basis for identifying new targets for osteoarthritis treatment.

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