The relationship between the spine–pelvis–lower extremity sagittal parameters and disease–related molecules in serum and synovial fluid in patients with knee osteoarthritis

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Objective: To study the relationship between spine-pelvis-lower extremity sagittal parameters and disease-related molecules in serum and synovial fluid in patients with knee osteoarthritis.

Methods: A total of 65 patients diagnosed with knee osteoarthritis in Dongguan Tangxia Hospital between May 2013 and December 2016 were collected as OA group, and 70 patients with meniscus and cruciate ligament injury undergoing repair surgery during the same period were collected as the control group. Spine-pelvis-lower extremity sagittal parameters were measured by erect whole spine lateral radiographs. The serum was collected and inflammatory factor contents were measured. The synovial fluid was collected to determine inflammatory factor contents and apoptosis molecule expression. Results: Affected-side PFA and SFA levels of OA group were significantly lower than those of healthy side while FI and ST levels were significantly higher than those of healthy side; serum TNF-α, IL-1β, IL-18, MMP1 and MMP3 contents of OA group were significantly higher than those of control group, negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels; TNF-α, IL-1β, IL-18, MMP1, MMP3, Fas, FasL, Caveolin-1, NO, CHOP and GRP78 contents in synovial fluid of OA group were significantly higher than those of control group, negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels. Conclusion: Sagittal parameters of spine-pelvis-lower extremity can be used to evaluate the inflammatory response and apoptosis in patients with knee osteoarthritis.

1. Introduction

Osteoarthritis (OA) is an important disease that affects the health of the elderly, which trends to occur in the knee joint, and characterized by pain and dysfunction in the joints. Articular cartilage degeneration, abnormal new bone formation and surrounding osteophyte formation are the most basic pathological local characteristics of patients with knee OA, and the inflammatory reaction and apoptosis are closely related to the pathological changes of articular cartilage. Knee joint, pelvis and spine are the important anatomical structures that maintain body balance together, and during the development of OA, knee joint disease can only affect local anatomical shape of the joint, but can also cause pelvic and spinal shape change, and lead to chronic low back pain, lumbar spondylolisthesis and other secondary changes[1,2]. In the following study, spine-pelvis-lower extremity sagittal parameters were determined to reflect the anatomical features of knee joint, pelvis, and spine in patients with knee OA, and the correlation of spine-pelvis-lower extremity sagittal parameters with inflammatory reaction and apoptosis molecules in serum and synovial fluid was studied.

2. Subjects and methods

2.1 Research subject information

A total of 65 patients diagnosed with knee osteoarthritis in Dongguan Tangxia Hospital between May 2013 and December 2016 were collected as OA group of the research, all patients were
in accordance with the diagnosis for primary knee osteoarthritis, the affected-side knee joint was without obvious deformity and with activity $>100^\circ$, and the contralateral knee joint was normal; patients associated with rheumatoid arthritis, traumatic arthritis, gouty arthritis and infectious arthritis were excluded. 70 patients undergoing repair surgery for meniscus and cruciate ligament injury in our hospital during the same period were collected as the control group of the research, and they were without the history of osteoarthritis. OA group included 29 male cases and 36 female cases that were 58-71 years old; control group included 33 male cases and 37 female cases that were 52-66 years old. There was no significant difference in general information between the two groups of patients ($P>0.05$).

2.2 Imageological examination and parameter determination methods

Both groups of patients received erect whole spine lateral radiographs to get images, and then the spine-pelvis-lower extremity sagittal parameters spinal tilt (ST), pelvis, pelvic femoral angle (PFA), sacrum femoral angle (SFA) and femoral anteversion (FI) were measured.

2.3 Inflammatory factor content detection methods

5ml of peripheral venous blood was collected from the two groups of patients before the operation and centrifuged to separate serum; suitable amount of synovial fluid was collected from OA group during arthrocentesis, suitable amount of synovial fluid was collected from the control group during surgery to repair the meniscus or cruciate ligaments, and enzyme-linked immunosorbent assay kit was used to determine TNF-$\alpha$, IL-1$\beta$, IL-18, MMP1 and MMP3 levels in serum and synovial fluid.

2.4 Apoptosis molecule expression detection methods

Suitable amount of synovial fluid was collected from OA group during arthrocentesis, suitable amount of synovial fluid was collected from the control group during surgery to repair the meniscus or cruciate ligaments, RNA was extracted, then fluorescence quantitative PCR kit was used to amplify Fas, FasL, Caveolin-1, NO, CHOP and GRP78, and the obtained amplification curve was referred to calculate Fas, FasL, Caveolin-1, NO, CHOP and GRP78 mRNA expression.

2.5 Statistical methods

SPSS 19.0 software was used to input inflammatory factor contents and apoptosis molecule expression, differences between groups were by t test and $P<0.05$ indicated statistical significance in differences.

3. Results

3.1 Spine–pelvis–lower extremity sagittal parameters

Analysis of affected-side and healthy-side spine-pelvis-lower extremity sagittal parameters PFA, SFA, FI and ST levels of OA group was as follows: affected-side PFA and SFA levels of OA group were significantly lower than those of healthy side while FI and ST levels were significantly higher than those of healthy side. The differences were statistically significant in the affected-side and healthy-side spine-pelvis-lower extremity sagittal parameters PFA, SFA, FI and ST levels of OA group ($P<0.05$), shown in Table 1.

3.2 Serum inflammatory factor contents of two groups of patients

Analysis of serum inflammatory factors TNF-$\alpha$ (ng/L), IL-1$\beta$ (ng/L), IL-18 (ng/L), MMP1 ($\mu$g/L) and MMP3 ($\mu$g/L) contents between two groups of patients was as follows: serum TNF-$\alpha$, IL-1$\beta$, IL-18, MMP1 and MMP3 contents of OA group were significantly higher than those of control group. Differences in serum TNF-$\alpha$, IL-1$\beta$, IL-18, MMP1 and MMP3 contents were statistically significant between two groups of patients ($P<0.05$), shown in Table 2. Pearson test showed that serum TNF-$\alpha$, IL-1$\beta$, IL-18, MMP1 and MMP3 contents were negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels.

Table 1.

<table>
<thead>
<tr>
<th>OA group</th>
<th>n</th>
<th>PFA</th>
<th>SFA</th>
<th>FI</th>
<th>ST</th>
</tr>
</thead>
<tbody>
<tr>
<td>Affected-side</td>
<td>65</td>
<td>3.15±0.62</td>
<td>42.91±6.21</td>
<td>11.84±1.95</td>
<td>93.68±11.27</td>
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<tr>
<td>Healthy-side</td>
<td>65</td>
<td>8.72±0.93</td>
<td>53.54±7.48</td>
<td>4.26±0.67</td>
<td>85.79±9.37</td>
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<tr>
<td>$T$</td>
<td></td>
<td>14.827</td>
<td>7.498</td>
<td>12.672</td>
<td>8.712</td>
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<td>$P$</td>
<td>&lt;0.05</td>
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</tbody>
</table>

Table 2.

<table>
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<tr>
<th>Groups</th>
<th>n</th>
<th>TNF-$\alpha$</th>
<th>IL-1$\beta$</th>
<th>IL-18</th>
<th>MMP1</th>
<th>MMP3</th>
</tr>
</thead>
<tbody>
<tr>
<td>OA group</td>
<td>65</td>
<td>15.58±1.85</td>
<td>11.59±1.62</td>
<td>9.35±1.15</td>
<td>5.29±0.94</td>
<td>3.96±0.51</td>
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<tr>
<td>Control group</td>
<td>70</td>
<td>6.59±0.93</td>
<td>4.42±0.56</td>
<td>3.02±0.51</td>
<td>2.41±0.35</td>
<td>1.58±0.22</td>
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<tr>
<td>$P$</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
<td>&lt;0.05</td>
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</tbody>
</table>
3.3 Inflammatory factor contents in synovial fluid of two groups of patients

Analysis of inflammatory factors TNF-α (ng/L), IL-1β (ng/L), IL-18 (ng/L), MMP1 (μg/L) and MMP3 (μg/L) contents in synovial fluid between two groups of patients was as follows: TNF-α, IL-1β, IL-18, MMP1 and MMP3 contents in synovial fluid of OA group were significantly higher than those of control group. Differences in TNF-α, IL-1β, IL-18, MMP1 and MMP3 contents in synovial fluid were statistically significant between two groups of patients (P<0.05), shown in Table 3. Pearson test showed that TNF-α, IL-1β, IL-18, MMP1 and MMP3 contents in synovial fluid were negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels.

3.4 Apoptosis molecule expression in synovial fluid of two groups of patients

Analysis of apoptosis molecules Fas, FasL, Caveolin-1, NO, CHOP and GRP78 mRNA expression in synovial fluid between two groups of patients was as follows: Fas, FasL, Caveolin-1, NO, CHOP and GRP78 expression in synovial fluid of OA group were significantly higher than those of control group. Differences in Fas, FasL, Caveolin-1, NO, CHOP and GRP78 mRNA expression in synovial fluid were statistically significant between two groups of patients (P<0.05), shown in Table 4. Pearson test showed that Fas, FasL, Caveolin-1, NO, CHOP and GRP78 expression were negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels.

4. Discussion

The incidence of knee OA is high in the elderly, and local lesions in the knee joint will not only affect the structure and function of the knee joint, but will also affect the shape of the hip and spine because of the damage of lower extremity alignment and balance. In clinical practice, the imaging examination can obtain the spine-pelvis-lower extremity image, and measuring the sagittal parameters PFA, SFA, FI and ST can assess morphological change of the spine, pelvis and lower limb[3]. The FI can directly reflect the local morphological changes caused by knee lesions, the ST and SFA can reflect the degree of lumbar lordosis, and the PFA can reflect the degree of hip flexion[4]. In the study, the analysis of the changes of spine-pelvis-lower extremity sagittal parameters PFA, SFA, FI and ST in patients with OA showed that PFA and SFA levels of OA group were significantly lower than those of control group while FI and ST levels were significantly higher than those of control group. This means that the knee joint lesions in patients with knee OA can affect the morphology of hip and spine and cause hip flexion and spine antversion, and spine-pelvis-lower extremity sagittal parameters can evaluate OA severity from the imaging perspective. The occurrence and development of OA are associated with articular cartilage inflammation, apoptosis and other biological processes, and in order to further clarify whether the spine-pelvis-lower extremity sagittal parameters were associated with the development and changes of OA, the correlation of above imaging parameters with articular cartilage inflammation and cell apoptosis was analyzed in the study. The activation of inflammatory response is the basic pathological characteristic within OA lesions, and the abnormal synthesis and secretion of TNF-α, IL-1β, IL-18, MMP1, MMP3 and a variety of other inflammatory factors will cause synovial inflammation and cartilage inflammation, and the abnormal synthesis and secretion of TNF-α, IL-1β, IL-18, MMP1, MMP3 and a variety of other inflammatory factors will cause synovial inflammation and cartilage inflammation. TNF-α is the pro-inflammatory medium playing an important role in OA progression, which on the one hand, promote the release of collagenase and the activation of protease release by prostaglandin to cause the destruction of articular cartilage, and on the other hand, can activate the oxidative stress response of the articular cartilage to result in apoptosis[8,9]. IL-1β and IL-18 are the members of the IL-1 family, and the abnormally secreted IL-1β and IL-18 in articular cartilage can not only cause inflammation cascade activation, but can also be combined with the receptors IL-1R and IL-18R to increase the secretion of protease MMP1 and MMP3, and then result in the destruction of articular cartilage[10-12]. In the study, the analysis of the content of above inflammatory factors in serum and synovial fluid of OA patients showed that TNF-α, IL-1β, IL-18, MMP1 and MMP3 contents

Table 3.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>TNF-α (ng/L)</th>
<th>IL-1β (ng/L)</th>
<th>IL-18 (ng/L)</th>
<th>MMP1 (μg/L)</th>
<th>MMP3 (μg/L)</th>
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<tbody>
<tr>
<td>OA group</td>
<td>65</td>
<td>46.28±7.14</td>
<td>25.82±3.85</td>
<td>15.28±2.03</td>
<td>0.39±0.06</td>
<td>0.24±0.05</td>
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<td>Control group</td>
<td>70</td>
<td>21.29±3.58</td>
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<td>6.54±0.89</td>
<td>0.21±0.04</td>
<td>0.11±0.02</td>
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<tr>
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Table 4.

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>Fas</th>
<th>FasL</th>
<th>Caveolin-1</th>
<th>NO</th>
<th>CHOP</th>
<th>GRP78</th>
</tr>
</thead>
<tbody>
<tr>
<td>OA group</td>
<td>65</td>
<td>3.29±0.52</td>
<td>2.58±0.41</td>
<td>1.93±0.22</td>
<td>2.32±0.45</td>
<td>2.11±0.37</td>
<td>2.94±0.56</td>
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<tr>
<td>Control group</td>
<td>70</td>
<td>1.02±0.18</td>
<td>1.06±0.13</td>
<td>1.01±0.15</td>
<td>0.98±0.14</td>
<td>1.08±0.20</td>
<td>1.03±0.15</td>
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<tr>
<td>P</td>
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</tr>
</tbody>
</table>
in serum and synovial fluid of OA group were significantly higher than those of control group. This indicates that the abnormally activated inflammatory response in local joints is closely related to the occurrence and development of OA. Further analysis of the relationship between imaging parameters and inflammatory factor levels showed that TNF-α, IL-1β, IL-18, MMP1 and MMP3 contents in serum and synovial fluid were negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels. This shows that the change in the spine-pelvis-lower extremity sagittal parameters is associated with abnormal activation of the inflammatory response of the joint cartilage.

The pathological essence of OA is articular cartilage degeneration, and excessive articular cartilage cell apoptosis under the action of a variety of inflammatory mediators will cause cartilage tissue destruction and collagen ingredient loss[13,14]. The apoptosis of articular cartilage cells is not only regulated by traditional death receptor apoptosis pathways, but is also closely related to the new signaling molecules such as Caveolin-1 and NO. Fas/FasL are the key molecules regulating apoptosis through death receptor pathway, tumor necrosis factor receptor superfamily member Fas can be combined with ligand FasL to launch the activation of a variety of downstream caspase molecules, and then cause chondrocyte apoptosis through caspase cascade activation reaction[15]. Caveolin-1 is the membrane molecule playing an important role in the process of caveola structure formation and maintenance, and it can interact with ceramide to antagonize PI3K activity, inhibit the biological effect of PI3K/AKT pathway, increase the sensitivity of cells to apoptosis signal, and promote the occurrence of apoptosis[16]. NO is a kind of gas signal molecule, and the excessively generated NO in cartilage tissue can increase the expression of endoplasmic reticulum stress molecules CHOP and GRP78, and then result in the chondrocytes apoptosis through endoplasmic reticulum stress[17]. In the study, analysis of the expression of above apoptotic molecules in OA synovial fluid showed that Fas, FasL, Caveolin-1, NO, CHOP and GRP78 expression in synovial fluid of OA group were significantly higher than those of control group. This indicates that the abnormally highly expressed apoptotic molecules in the synovial fluid are closely related to the occurrence and development of OA. Further analysis of the relationship between imaging parameters and cell apoptosis molecule expression showed that Fas, FasL, Caveolin-1, NO, CHOP and GRP78 expression were negatively correlated with PFA and SFA levels, and positively correlated with FI and ST levels. This suggests that the change in the spine-pelvis-lower extremity sagittal parameters is related to the abnormal apoptosis of the articular cartilage.

To sum up, it is believed that spine-pelvis-lower extremity sagittal parameters significantly change in patients with knee OA and are correlated with inflammatory factor content and apoptosis molecule expression, and spine-pelvis-lower extremity sagittal parameters can be used to assess the inflammation and apoptosis in the course of patients with OA.

References