QUANTIFICATION OF RAT KNEE CONTACT PROPERTIES USING *IN VIVO* MICRO-COMPUTED TOMOGRAPHY AT FLEXION-TO-EXTENSION POSITIONS

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**SUMMARY**

This paper describes a method for the evaluation of contact properties of rat knee at flexion-to-extension positions using *in vivo* micro-Computed Tomography (µCT). Twelve knees of healthy rats were scanned and divided into two age groups: group A: 8 weeks old (w.o.) (n=6), group B: 12 w.o. (n=6). Each rat is placed under anesthesia and positioned supine with lower members in three positions: bended, physiological, extended. A minimal distance algorithm was employed to produce 3D distance maps of each meshed compartment. Bone contact areas with contact threshold 0.6 mm were calculated. Root-mean-square curvatures of the contact surface were also computed. The results reveal most modifications at the bended position and depend on the ageing process.

**INTRODUCTION**

Osteoarthritis (OA) is a widespread disabling pathology which involves high socio-economic impacts. At present, many studies have been focused on the assessment of joint inflammation and damage in inflammatory arthritis [1]. But, little works concern the functionality of the movement and the degradation of the dynamic properties due to the osteoarthritis. Our current study is a preliminary work on healthy rats with the purpose to characterize the knee joint contact properties such as contact areas and curvatures at several flexion-to-extension positions using quantitative *in vivo* 3D micro-computed tomography (µCT). µCT images were obtained at a resolution of 35 μm x 35 μm x 35 μm. Four compartments (medial and lateral femoral bone, medial and lateral tibial bone) of each knees were extracted using segmentation module of Simpleware software.

**METHODS**

Healthy rats (n=12 knees) were scanned *in vivo* using high-resolution µCT (55kVp, 169μA). They were divided into 2 groups as follows: group A: 8 weeks old (w.o.) (n=6), group B: 12 w.o. (n=6). Animals were placed under anesthesia with isofluorine and positioned supine with lower members in three positions: bended, physiological, extended. Each joint was scanned at an isotropic resolution of 35 μm x 35 μm x 35 μm and images were coded in 8 bits gray levels. In order to measure flexion angle and to increase the field of view, another series was also acquired with low resolution (sagittal in-plane resolution: 70 μm x 70 μm, slice thickness: 35 μm). This modality of imaging was chosen because of its ability to detect the osseous parts [2-3]. Simpleware 4.0 was used to segment and visualize the bones of the knee from micro-CT scans. For the knee, the Segmentation module of ScanIP was used to label femur and tibia. A density threshold higher than 100 was set as “bone”; the threshold was kept constant throughout the study. Then, four labels were reconstructed using the Create mask module: lateral and medial compartments for femur and tibia. They are manually corrected to disjoint the compartments: 3D smooth surface meshes (one mesh for each compartment) were generated by employing a marching cube algorithm [4] and exported in STL format. The next step consisted in computing minimal distances between all points belonging to the two 3D meshes, in order to determine joint contact areas maps (contact threshold being defined at 0.6 mm) [5, 6]. According to the 3D meshes, root-mean-square (rms) curvatures of the contact surface were also computed [7].

For comparison between means at two positions, nonparametric tests (Kruskal-Wallis test) were performed due to the low number of cases. Two tailed P values of 0.2 or less were considered statistically significant. Intra-reader reliability on the results was estimated using coefficients of variation: one 3D model was built from the CT-scans ten times over a two weeks period with the same protocol described above.

**RESULTS AND DISCUSSION**

The angle between femoral axis and tibial axis for the bended, physiological and extended positions was measured respectively at around 18°, 46° and 165° (Figure 1).
In the case of rats of 8 w.o. (Figure 2), the change from bended to physiological position is responsible of a significant (p=0.2) difference in the medial and lateral femoral contact areas. At bended position, the effect is higher at lateral side and lower at medial side.

For rats of 12 w.o., the effects are not the same (Figure 3): a significant difference when comparing to extended position is highlighted at lateral side in the femoral and tibial contact sites (p ≤ 0.04). These changes may be due to the ageing process responsible of a different growth in the femoral and tibial compartments.

These results correlate with the findings of Yamanaka et al [8] on human living knees with OA which demonstrate that the best position to follow the changes in the joint is 15° knee flexion.

When computing the rms curvatures for rats of 8 w.o. (Table 1), a significant increase (p<0.1) at femoral and tibial contact sites is noticed at bended position. Similar effects are observed for rats of 12 w.o. (Table 2). This can be related to the flattening of the contact surface at bended position.

**Table 1:** Mean values ± SD of rms curvature (mm-1) at contact sites (femur, tibia) for rats at 8 w.o. for the bended, extended and physiological positions.

<table>
<thead>
<tr>
<th></th>
<th>Bended</th>
<th>Extended</th>
<th>Physiological</th>
</tr>
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<tbody>
<tr>
<td>Femur</td>
<td>13.5±7.8</td>
<td>7.6±5.7</td>
<td>9.3±6.5</td>
</tr>
<tr>
<td>Tibia</td>
<td>4.6±5.5</td>
<td>2.2±2.1</td>
<td>3.6±2.1</td>
</tr>
</tbody>
</table>

**Table 2:** Mean values ± SD of rms curvature (mm-1) at contact sites (femur, tibia) for rats at 12 w.o. for the bended, extended and physiological positions.

<table>
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<tr>
<th></th>
<th>Bended</th>
<th>Extended</th>
<th>Physiological</th>
</tr>
</thead>
<tbody>
<tr>
<td>Femur</td>
<td>7.2±1.5</td>
<td>5.9±3.7</td>
<td>6.3±1.6</td>
</tr>
<tr>
<td>Tibia</td>
<td>3.4±3.3</td>
<td>1.8±1.7</td>
<td>2.6±1.7</td>
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</tbody>
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The test of reproducibility leads to a mean coefficient of variation of 5 % for the contact area and 15 % for the rms curvature. This indicates that the rms curvature is more sensible to the noise in the surface model.

**CONCLUSIONS**

A quantitative analysis of geometrical articular contact properties (areas and curvatures) for three flexion-to-extension positions was obtained from in vivo rat knee micro-CT imaging. Actual results on healthy rats reveal most modifications at the bended position. This study suggests that our methodology could also be employed in pathological context to follow and quantify the impact of OA diseases on movement.

**ACKNOWLEDGEMENTS**

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**REFERENCES**