Computed Tomography Imaging of Femoral Bone Defects in Rats Treated with AD DIEE Biophytomodulators

OROS Daniela1*, Liviu OANA1, Robert PURDOIU1, Cristian CRECAN1, Ancu DINCA2

1) Faculty of Veterinary Medicine, USAMV Cluj-Napoca
2) The Biosynergic Research and Study Centre “Dinca Ancu”, Bucharest

*Corresponding author, e-mail: orosdaniela@yahoo.com

Abstract. The role of AD DIEE Biophytomodulators in helping bone healing is still unclear, but biochemical and histological investigations showed that healing in bone defects in rats is enhanced when using these devices. This study aimed to investigate the effect of AD-DIEE biophytomodulators on healing of bone defects in rats, by computed tomography imaging of the newly formed bone callus. Femoral bone defects were surgically induced in Wistar rats, males of the same age, clinically healthy. Surgeries were performed under general anaesthesia. The animals were divided into two groups, one of which was treated with AD-DIEE biophytomodulators. Postoperative CT scans were performed using the apparatus NewTom 3G. The images obtained were analyzed by measuring the size of bone defects and the newly formed bone callus. In the treated animals, bone defects were smaller at 3 and 6 weeks postoperatively, compared to those found in the untreated group. The bone defects in the treated animals are shrinking at a higher rate compared to the control group. The images obtained with CT scans are consistent with the previous preclinical studies regarding the action of AD DIEE biophytomodulators upon bone healing.

Keywords: biophytomodulator, bone, callus, CT scan.

INTRODUCTION

Volume and density of bone callus are extremely important elements in assessing the healing of a bone defect. These values provide important clues on the recovery of mechanical function of the affected bone. (Klaus et all, 2009). Oana et al. (2010) proved that AD DIEE Biophytomodulators have a beneficial influence on the healing of bone defects in sheep, and in 2012 the same group demonstrated, through histopathological investigations, the positive effect these Biophytomodulators have on bone healing in rats. This study aimed to investigate the effect of AD-DIEE Biophytomodulators on healing of bone defects in rats, by computed tomography imaging of the newly formed bone callus.

MATERIALS AND METHODS

The experimental design was done in the Faculty of Veterinary Medicine, Cluj-Napoca, in the department of Surgical Techniques. As in the study of 2012 (Oros et al., 2012), the animals used in this study were 10 white Wistar rats, males, 1 year old, clinically healthy, average weight of 250 g. The bone defects were done using a technique described by Oana et al. (2011). The animals underwent general anesthesia, xylazine 8mg/kg + ketamine 40 mg/kg. The femoral shaft was exposed surgically, after the incision of the skin and the dilacerations of the thigh muscles. Bone defects were made using a dental drill of 1.7 mm diameter, adapted to a dental micro motor at a speed of 3500 rpm and irrigated with saline to avoid
overheating. The drilling was done through the whole thickness of the compact, reaching the medullary canal, obtaining a bone defect of 1.7 mm in diameter. The skin was sutured, using 4-0 surgical silk. The animals were randomly divided into two groups, control and test group.

The control group (M group) received no medical treatment. The animals from the test group (BF group) were treated with AD DIEE Biophytomodulators. Three Biophytomodulators were attached to the bottom of the cages, in a triangular shape.

To obtain the images, rats underwent computed tomography (CT), 1, 3, 6 weeks postoperatively (Fig. 3.4), using NewTom 3G CT machine at a private clinic in Cluj-Napoca. Each time, the animals were subjected to general anesthesia (same protocol from the bone defect surgery). For statistical calculation, the determination of the index "p" was performed using the program Microsoft Office Excel 2007, using two-tailed t-test.

Statistical interpretation of data and the value of "p" were carried out according to the following scheme: p ≤ 0.05 - statistically significant results, p ≤ 0.01 - distinct statistically significant results, p ≤ 0.001 - statistically very significant results.
RESULTS AND DISCUSSIONS

CT images obtained were analyzed and interpreted using NNTViewer program that comes with measuring instruments.
One week postoperatively, the size of the bone defects remains at the initial size, 1.7 mm and initial phenomena of callus formation can be seen in both groups of animals.

Tabel 1

Size (mm) of bone defects, by section, measured in NNTViewer

<table>
<thead>
<tr>
<th>Time</th>
<th>Animal Number</th>
<th>Group</th>
<th>3 weeks</th>
<th>6 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>M</td>
<td>BF</td>
<td>M</td>
</tr>
<tr>
<td>1</td>
<td>1</td>
<td>1.1</td>
<td>0.9</td>
<td>0.8</td>
</tr>
<tr>
<td>2</td>
<td>1.5</td>
<td>0.8</td>
<td>0.8</td>
<td>0.6</td>
</tr>
<tr>
<td>3</td>
<td>1.3</td>
<td>1</td>
<td>0.6</td>
<td>0.7</td>
</tr>
<tr>
<td>4</td>
<td>1</td>
<td>1.1</td>
<td>0.7</td>
<td>0.5</td>
</tr>
<tr>
<td>5</td>
<td>1.3</td>
<td>1.3</td>
<td>1.1</td>
<td>1</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Time</th>
<th>Mean</th>
<th>Median</th>
<th>Standard deviation</th>
<th>Variance</th>
<th>Standard error</th>
<th>Confidence interval</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1.22</td>
<td>1.3</td>
<td>0.216</td>
<td>0.047</td>
<td>0.190</td>
<td>1.22 ±0.019</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td></td>
<td>1.06</td>
<td>1.1</td>
<td>0.181</td>
<td>0.033</td>
<td>0.159</td>
<td>1.06±0.159</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td></td>
<td>0.82</td>
<td>0.8</td>
<td>0.192</td>
<td>0.037</td>
<td>0.168</td>
<td>0.82±0.168</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.72</td>
<td>0.7</td>
<td>0.187</td>
<td>0.037</td>
<td>0.164</td>
<td>0.72±0.164</td>
<td></td>
</tr>
</tbody>
</table>

Fig. 13. Variation of the mean size (and confidence interval) of the bone defects in the 2 groups (M and BF) at 1, 3 and 6 weeks post operatively

At 3 weeks postoperatively the newly formed callus appears, as previously demonstrated by the histological study (Oros et al, 2012), the average size of the defect decreased to 1.22 mm in the control group and 1.06 mm in the treated animals. Newly formed callus is filling the bone defect, apparently having a greater density in the BF group.
At 6 weeks postoperatively, the bone defect is filled almost completely with the newly formed callus, significantly reducing the size of defects in both groups of animals, up to an average of 0.82 mm in control and 0.72 mm in the animals treated with AD DIEE Biophytomodulators. Compared to histological investigation, computed imaging has a major advantage, namely it is a non-invasive method that allows dynamic evaluation of healing processes on the same patient.

CONCLUSIONS

Imaging tests give an overview of bone healing phenomena, with the possibility of subsequent evaluation in the same patient, as opposed to histological investigation. Computed tomography is an essential tool in the investigation of bone healing.

Linear measurements show a greater average decrease in diameter of the defect in animals treated with AD DIEE Biophytomodulators, compared to controls, but the differences between the treated group and control are not supported statistically.

These data suggest that AD DIEE Biophytomodulators may be a complementary therapy in bone repair dynamics. The involvement of AD DIEE Biophytomodulators in bone healing mechanism remains to be clarified.

REFERENCES