INTRODUCTION: Growth hormone (GH) is a systemically circulating hormone that has a direct and an indirect stimulatory effect on tissue, the latter mediated by insulin like growth factor-I (IGF-I) (1). Previous studies have demonstrated that recombinant porcine Growth Hormone (r-pGH) accelerates regenerate consolidation in distraction osteogenesis and promotes healing of segmental bone defects in Yucatan micropigs (2,3). However conflicting results exists concerning the effect of GH in fracture healing in different animal models (4,5).

Thus, the objective of this study was determine whether r-pGH accelerates healing in standardized osteotomy model of the tibia in Yucatan micropigs.

METHODS: 24 mature Yucatan micropigs were ranked for age and weight and equally distributed into two treatment groups. The animals in the study group received a daily subcutaneous injection of recombinant porcine growth hormone (100 µg/kg bodyweight) starting the day of surgery, while the animals in the control group received sodium-chloride as a placebo. Under general anesthesia, a standardized osteotomy was created surgically in the middyaphysis of the right tibia. The tibiae were stabilized using stainless steel plates. All procedures were carried out under ethical permission of the animal rights protection authorities. Serum IGF-I analysis and radiographs were performed every fourth day.

Four weeks postoperatively the animals were sacrificed. The tibiae were harvested and subjected to a material testing machine in a torsional modus. Torsional stiffness and maximum torsional moment were determined from the load displacement curves. After biomechanical testing, the osteotomy zone and 2 cm of the adjacent cortical bone were divided into 3 mm thick sagittal sections using a precision grinding saw and embedded in methylmethacrylate. 6 µm serial slices were produced using a hard-cutting microtome. The sections were stained with von Kossa / Safranine-O stain. The slices were digitized with a 3-Chip CCD color camera and processed using the LEICA Quantimet image analysis work station. 100 fields were evaluated to determine the bone density (BD) of the osteotomy zone. The bone density (BD) of the callus was also calculated: $\text{BD (mg/cm}^3) = \text{callus area} / \text{callus length}$.

For statistical analysis of the treatment groups, the Mann Whitney U test was implemented to compare the biomechanical and histomorphometric data for both groups at a significance level of $p<0.05$.

RESULTS: The mean serum level of IGF-I increased to 4-fold of preoperative basal level in the treatment group and remained unchanged in the control group. Radiographically the animals treated with r-pGH demonstrated a more advanced healing of the osteotomy.

Maximum torsional moment was 92% higher in the GH-treated group (17.1 ± 4.6 Nm) than in the control group (8.9 ± 4.9 Nm) ($p = 0.001$), and torsional stiffness was 66% higher in the treatment group (2.48 ± 0.91 Nm/°) than in the control group (1.49 ± 0.76 Nm/°) ($p<0.05$). Compared with the intact contralateral tibia, the tibia in the GH group and the control group reached 135 ± 53% and 84 ± 49% of torsional stiffness, respectively ($p < 0.05$), and 68 ± 24% and 29 ± 14% of maximum torsional moment ($p=0.05$) (Figure 1).

The histomorphometric-measurements revealed an advance for the CA (GH: 127.6 ± 38.9 mm²; placebo: 75.9 ± 50.7 mm²; $p < 0.005$) as well as for the BA (GH: 89.3 ± 25.8 mm²; placebo: 55.9 ± 38.5 mm²; $p < 0.001$) for the GH-treated animals in comparison to the control animals (Figure 2). The BD was similar in both groups (GH: 70.6 ± 8.4%; placebo: 74.0 ± 6.24%; $p = 0.26$).

DISCUSSION: The histomorphometrical results of the study revealed that systemic application of GH leads to an accelerated new bone and callus formation in this osteotomy model. The bone density (BD) was similar in both groups indicating that the structure of the callus tissue in both groups is comparable. This is in contrast to other studies using a rat model, where the callus formed during GH treatment was abnormal with an extemely loose structure (6).

The advanced callus formation resulted in a higher biomechanical stability of the bone after 4 weeks healing. This could be observed also radiographically where a earlier and faster callus formation was visible in the GH-treated animals compared to the placebo animals. The increase of IGF-I in the GH-treated group indicates in intact GH-IGF-I axis. This is an contrast to another study where the IGF-I levels were unchanged in response to GH-administration and no effect of GH was visible (4).

In conclusion, these results confirm our previous findings and suggest that systemic administration of homologous growth hormone accelerates new bone formation in fracture healing. Therefore further studies for the use of GH as a clinical tool for improved fracture healing should be considered.

REFERENCES: