

# Efek penggunaan bahan tandur tulang Demineralized Freeze Dried Bone Xenograft (DFDBX) dan membran perikardium pada penyembuhan defek tulang tikus Sprague Dawley : Evaluasi radiologik, histopatologik, dan imunohistokimia = The effect of using Demineralized Freeze Dried Bone Xenograft (DFDBX) and pericardium membrane on the treatment of bone calvarial defect in Sprague Dawley rats : A radiological, histopathological, and immunohistochemical evaluation

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## Abstrak

### [<b>ABSTRAK</b><br>

Latar Belakang: Penggunaan biomaterial berupa bahan tandur tulang dan membran untuk prosedur Guided Bone Regeneration (GBR) sangat diperlukan di bidang bedah maksilofasial dan, untuk mengatasi defek tulang yang dapat terjadi oleh berbagai sebab. Penelitian ini bertujuan untuk mengetahui efek pemakaian bahan tandur tulang DFDBX dengan membran perikardium (MPK) bovine pada defek tulang kalvaria tikus.

Bahan dan Metode: Studi eksperimental ini menggunakan 45 ekor tikus Sprague Dawley sebagai hewan coba dibagi dalam 3 kelompok secara acak. Ciritical size defect sebesar diameter 5 mm dibuat pada tulang kalvaria seluruh hewan coba. Kelompok I merupakan kelompok kontrol, tidak diberikan perlakuan dan defek dibiarkan sembuh dengan sendirinya, kelompok II yang diberi DFDBX, dan pada kelompok III defek diisi dengan DFDBX dan ditutup dengan MPK (DFDBX+MPK). Setelah 1,4 dan 8 minggu dilakukan pengorbanan pada kelompok hewan coba, dilanjutkan dengan evaluasi secara radiologik, histopatologik untuk reaksi radang, pertumbuhan tulang dan pemeriksaan imunohistokimia dengan osteokalsin. Data dianalisis secara statistik dengan menggunakan uji ANOVA.

Hasil: Penilaian radiografik diperoleh perbedaan bermakna pada rerata densitas area defek minggu ke 8 antara kelompok kontrol dengan DFDBX+MPK ( $p<0,001$ ) dan antara kelompok DFDBX dengan DFDBX+MPK ( $p=0,03$ ).

Pertumbuhan tulang baru pada minggu ke 8 tertinggi adalah pada kelompok DFDBX+MPK dengan perbedaan bermakna dengan kelompok kontrol ( $p=0,016$ ) dan dengan kelompok DFDBX nilai  $p=0,048$ . Ekspresi osteokalsin minggu ke-8 menunjukkan perbedaan bermakna antara kelompok kontrol dengan kelompok DFDBX ( $p<0,001$ ) maupun dengan kelompok DFDBX+MPK ( $p=0,0013$ ), namun tidak terdapat perbedaan bermakna antara kelompok DFDBX dengan kelompok MPK ( $p=1,000$ ).

Kesimpulan: Penggunaan DFDBX dengan kombinasi MPK terbukti secara radiologik, histopatologik dan imunohistokimia dapat meningkatkan regenerasi tulang pada defek tulang kalvaria.

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### <b>ABSTRACT</b><br>

Background: Reconstruction of cranial and maxillofacial defects is a challenging task. The standard method has included bone grafting and using membrane in guided bone regeneration procedure. Using biomaterial such as bone grafting and membrane for Guided Bone Regeneration (GBR) procedures is an essential issue in maxillofacial and dental reconstruction surgery to overcome bone defects caused by various etiologies.

Our study was aimed to identify the effect of using Demineralized Freeze-Dried Bone Xenograft (DFDBX) with (or without) bovine pericardium membrane (PCM) on the treatment of rats calvarial bone defects. **Materials and Method:** The experimental study used 45 Sprague-Dawley rats as the experimental animals, which were categorized randomly into three groups, i.e. the control group, DFDBX group, and DFDBX+PCM group. The 5-mm-critical-sized calvarial defects were created in all experimental animals. The first group was a control group, which did not receive any treatment with self-limiting defects; while subjects in the second group received DFDBX (DFDBX group) and in the third group, the defects were filled with DFDBX and PCM (DFDBX + PCM group). Animals were sacrificed at the 1st, 4th, and 8th weeks following the surgery. Subsequently, an evaluation was carried out using radiological analysis, histopathological assay to observe inflammatory reaction and bone growth, as well as immunohistochemical analysis of osteocalcin. Data were analyzed statistically using ANOVA test. The specimens were embedded in paraffin, serially cut, and stained with hematoxylin and eosin for analysis under light microscope. The inflammation reaction, new bone formation, and the rest of DFDBX and PCM were histomorphometrically evaluated. Immunohistochemical analysis of osteocalcin expression was performed.

**Results:** Radiological analysis demonstrated a significant difference of mean bone density in the defect area at the 8th week between subjects in the control group and those in DFDBX+PCM group ( $p < 0.001$ ), as well as between subjects in the DFDBX group and those in DFDBX+PCM group ( $p = 0.03$ ). The highest rate of bone healing at the 8th week was found in DFDBX+PCM group, which showed significant difference compared to the control group ( $p=0.016$ ) and to DFDBX group ( $p=0.048$ ). There was a significant difference of osteocalcin expression between the control group and DFDBX group ( $p < 0.001$ ), as well as between the control group and DFDBX + PCM group ( $p=0.0013$ ). However, there was no significant difference between the DFDBX group and the DFDBX+PCM group ( $p = 1.000$ ).

**Conclusion:** Our radiological, histopathological and immunohistochemical evaluation has demonstrated that DFDBX combined with PCM increases bone regeneration in the treatment of bone calvarial defect.

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The standard method has been bone grafting and using membrane in guided bone regeneration procedure.

The aim of this study was to analyze the effect of Demineralized Freeze Dried Bone Xenograft (DFDBX) with (or without) bovine pericardium membrane (PCM) on bone regeneration, in surgically created critical-size defects in rat calvaria, radiographically, histopathologically and immunohistochemically.

**Material and Methods:** Surgical critical-size bone defects were created in 45 animals that randomly divided into three groups: control group, DFDBX group, and DFDBX+PCM group. Animals were sacrificed at 1, 4 and 8 weeks post surgery.

Radiological analysis was done. The specimens were embedded in paraffin, serially cut, and stained with hematoxylin and eosin for analysis under light microscope. The inflammation reaction, new bone formation, and the rest of DFDBX and PCM were histomorphometrically evaluated. Immunohistochemical analysis of osteocalcin expression was performed.

**Result:** DFDBX and DFDBX+PCM groups demonstrated superior bone healing compared with control group. Group DFDBX+PCM showed more advanced healing at 8 weeks post surgery and show the highest density radiographically as compared with the

other group DFDBX and control. Immunohistochemistry revealed the presence of osteocalcin in osteoblast and matrix extracellular and show significant differences were noted between DFDBX and DFDBX+PCM to control groups.

Conclusion : Application of DFDBX combined with bovine PCM gave the best result in bone regeneration of critical size defects in rat calvaria. , Background :Reconstruction of cranial and maxillofacial defects is a challenging task.

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