Abstract. This study assessed the use of composite autogenous bone and deproteinized bovine bone (DBB) for repairing alveolar cleft compared with autogenous bone alone in terms of clinical outcomes and patient morbidity. 30 patients with a mean age of 10.2 ± 1.7 years were randomly divided into two groups. Group I used autogenous cancellous bone graft harvested from the anterior iliac crests by the conventional trapdoor approach. Group II used a composite of DBB and autogenous cancellous bone harvested by a trephine bone collector; the proportion of 1:1 by volume was used. The bone graft quantities of both groups decreased with time. Their average changes were not statistically different over 24 months after grafting. The canines of both groups could spontaneously or orthodontically erupt through the grafting areas. Patients in group II recovered from uncomfortable walking significantly faster than those in group I (p < 0.05) and their duration of hospital stay was significantly shorter than those in group I (p < 0.05). The average operation time, intra-operative blood loss and postoperative pain were less in group II than in group I (p > 0.05).

Keywords: alveolar cleft; autogenous; deproteinized bovine bone; graft; hydroxyapatite; iliac crest.
moderate to severe postoperative pain, gait disturbance and extended hospital stays.

The use of xenograft or alloplastic materials, instead of autogenous bone, has been widely accepted for many years. These materials include tricalcium phosphate, hydroxyapatite, deproteinized bovine bone (DBB) and synthetic polymers. DBB can be prepared by chemical treatment, low temperature sintering and high temperature sintering. Bio-Oss (Geistlich Pharma AG, Wolhusen, Switzerland), one of the most commonly used DBB in periodontal and implant surgery,[6,7,11,17,26] was prepared by chemical treatment. The cost is high, particularly if a large amount of the commercial bone graft is required. The National Metal and Materials Technology Center of Thailand (MTEC) has developed DBB as a grafting material, obtained by sintering bovine bone at 1200 °C to eliminate proteins and lipids, which are antigenic organic matter. It acts as a scaffold consisting of interconnecting pore systems; the pore size is 200–500 μm and the available particle sizes are 0.25–1 mm. In vivo, it showed osteoconductive behaviour that enhanced bone formation in rabbit calvariums. The average new bone formation for DBB alone in a 10 mm² defect in 12 weeks was 25 ± 12% compared with 35 ± 5% for an autogenous bone chip (p > 0.05).[21]

Several studies[6,10,17] have tried to increase the grafting success rate by combining autogenous bone with DBB in various ratios. The proportions of 1:1 and 1:2 were found to obtain maximum new bone formation when compared with 1:4. The latest in vivo study[22] demonstrated that 8 weeks after grafting with autogenous bone combined with DBB (MTEC), the proportions of 1:1 and 1:2 gained more new bone formation (21 ± 6% and 23 ± 6%, respectively) than 1:4 (10 ± 2%), DBB alone (14 ± 3%), or the critical size defect (11 ± 9%), but less than the autogenous bone group (30 ± 17%).

The aim of the present study was to evaluate the clinical and radiographic results of alveolar cleft bone grafting using the composite of autogenous bone from the iliac crest and DBB and to comparing these results with autogenous bone alone over a 24-month follow-up period.

Materials and methods

The study was conducted from November 2004 to June 2007 at the Dental Hospital, Prince of Songkla University in accordance with the regulations of the Faculty board ethics committee. ASA class I patients, aged 9–12 years, with residual alveolar clefts were included in the study. Patients who had bleeding disorders, bone and metabolic diseases, and were not available for 2-year follow-up were excluded from the study. 30 patients who had unilaterial alveolar clefts and 8 had bilateral alveolar clefts. Secondary alveolar bone grafting was performed by two oral surgeons using the same surgical technique. The patients were randomly divided into two groups and were unaware of the technique used. In group I, the cancellous bone graft was harvested from the anterior iliac crests by the conventional trap door technique. In group II, the cancellous bone was harvested from the anterior iliac crests using a trephine bone collector (Medicon, Tuttlingen, Germany; diameter 8.0 mm) and mixed with DBB (MTEC, Pathumthani, Thailand), with a particle size of 0.25 mm in the ratio of 1:1 by volume (Fig. 1). The bone grafts for both groups were compressed in 5 ml syringes and the volumes were measured prior to filling the alveolar cleft defects. In both groups, the alveolar cleft sites were grafted and closed using the gingival advancement flap technique. Analgesics (acetaminophen and meperidine) and antibiotics (intravenous cephalosporin) were prescribed according to the standard protocol.

Clinical assessment

The recordings for intra-operative assessments included duration of the operation (h), bone graft volume (ml) taken from the donor sites, and estimated blood loss (ml) calculated from the total fluid volume in the suction bottle minus the irrigating fluid plus the blood volume in blood-soaked four-by-four gauzes used in the operation. Postoperative assessments included: duration of hospital stay (day); time taken to walk again, with and without assistance (h); and postoperative pain level (using a 10 cm visual ana-
Table 1. Summary of clinical parameters.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group I (Autogenous bone alone (mean ± SD))</th>
<th>Group II (Autogenous bone + DBB 1:1 (mean ± SD))</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of unilateral cleft patients</td>
<td>11</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>Number of bilateral cleft patients (number of cleft sites)</td>
<td>3 (6)</td>
<td>3 (6)</td>
<td></td>
</tr>
<tr>
<td>Total cleft sites</td>
<td>17</td>
<td>16</td>
<td></td>
</tr>
<tr>
<td>Bone graft volume (ml)</td>
<td>2.53 ± 1.07</td>
<td>1.22 ± 0.20*</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Intra-operative blood loss (ml)</td>
<td>150 ± 46.30</td>
<td>122.5 ± 35.45</td>
<td>0.239</td>
</tr>
<tr>
<td>Duration of the operation (h)</td>
<td>2.68 ± 0.61</td>
<td>2.29 ± 0.91</td>
<td>0.172</td>
</tr>
<tr>
<td>Time taken to walk with assistance (h)</td>
<td>37.88 ± 11.07</td>
<td>25.5 ± 6.46*</td>
<td>0.006</td>
</tr>
<tr>
<td>Time taken to walk without assistance (h)</td>
<td>67.07 ± 13.86</td>
<td>46.63 ± 13.82*</td>
<td>0.003</td>
</tr>
<tr>
<td>Duration of hospital stay (day)</td>
<td>5.4 ± 1.12</td>
<td>4.23 ± 0.8*</td>
<td>0.006</td>
</tr>
</tbody>
</table>

* Unpaired t-tests, significant difference from group I at p < 0.05.

**Statistical analysis**

The data were analysed using SPSS version 13 (SPSS Inc., Chicago, IL, USA). Descriptive statistics and unpaired t-tests were used to compare the clinical results and bone graft quantities between the two groups, at each time interval, during the follow-up periods. One-way analysis of variance was applied to detect the change in bone quantities for each group during the follow-up periods. Multiple comparisons, using Tukey’s HSD test, were made where variances were homogeneous, otherwise Dunnett T3 was performed. The Mann–Whitney U-test was applied to detect postoperative pain differences between the two groups at each time interval. The Kruskal–Wallis test and Dunn’s multiple comparison test were used to assess the change in postoperative pain for each group during the 7 days after surgery. A p-value < 0.05 was considered significant.

**Results**

All patients tolerated the operation well without anaesthetic complications. The data for all clinical parameters are given in Table 1. One patient in group I and two patients in group II were excluded from the study due to lack of compliance and loss to follow up. The mean autogenous bone graft volume used in group II could be reduced by adding an equal volume of DBB. There was no significant difference in the operation time and intra-operative blood loss between the two groups. The patients in group II recovered from walking uncomfortably statistically faster than those in group I. The duration of the hospital stay was significantly shorter in group II than in group I.

The postoperative pain in both groups significantly reduced within 3 days after surgery. The overall pain score was less in group II than in group I, but the results were not statistically significant (Fig. 2).

Regarding complications at the donor site, paresthesia of the skin around the incision lines occurred in one case and pain when walking was reported in three cases in group I. No complications were detected in group II. At the recipient site, wound infection occurred in one patient in each group. Wound dehiscence was

![Fig. 2. Postoperative pain; *p < 0.05 significant compared with day 1 for groups I and II, respectively. Group I, autogenous bone alone; group II, autogenous bone + DBB 1:1.](image-url)
detected in one patient in group I and in three in group II. This might be due to excessive bone graft volume packed into the cleft sites, the tension of wound closure and the patient’s oral hygiene. They were healed eventually following wound debridement and antibiotics.

Canine tooth buds presented in 10 cleft sites in group I and 12 cleft sites in group II. Within the 24-month follow-up period, spontaneous eruption of canines had occurred; 5 teeth in group I (50%) and 5 teeth in group II (42%). The tooth eruption had been assisted by orthodontic force in three teeth in group I (30%) and two teeth in group II (17%). The spontaneous eruption of canines was demonstrated by serial occlusal radiographs (Fig. 3). The status of the teeth moved by orthodontic appliances is shown in Fig. 4.

The average bone graft densities and heights of both groups were not statistically different at each time interval ($p > 0.05$). The densities of both groups significantly reduced within 6 months after grafting, then seemed to be stable until month 24. The density in group I and group II reduced 31% and 27%, respectively, by 24 months after grafting (Fig. 5). The bone graft heights gradually decreased with time, 24% in both groups by 24 months post-surgery (Fig. 6).

**Discussion**

Satisfactory clinical outcomes were obtained from the bone trephination technique. A tiny incision line, and minimal muscle and periosteum detachment minimized the postoperative pain. As a result, patients recovered from walking uncomfortably faster. The operation time, intraoperative blood loss and duration of hospital stay were reduced when compared with the conventional trap door technique.

According to the authors’ protocol of alveolar cleft bone grafting, the conventional trap door approach was used as a standard technique for harvesting the bone graft because of the large amount of cancellous bone harvested. Bone graft harvesting by trephine bone collecting is less invasive and considered the standard technique in many centres, but for Thai patients, the bone graft volume achieved using this technique is much less than the volume from the trap door approach and inadequate for filling one cleft site defect. When the composite graft was used, the quantity of autogenous bone needed was approximately half less than using autogenous bone alone, therefore the trephination technique was more suitable than a trap door approach.

Regarding the non-resorbable property of hydroxyapatite that may retard the bone remodelling process$^{3,8}$, the new bone regenerating within the pores of DBB might be affected by stress shielding and did not undergo mechanical loading, which acted as a trigger for remodelling$^{1}$. In contrast, combining with autogenous bone seems to improve the graft success rate$^{8}$. The mixture of autogenous cancellous bone and hydroxyapatite contained viable osteoblasts and osteoprogenitor cells, essential for the mechanisms of osteogenesis and osteoinduction, and the xenogenic scaffold for osteoconduction. The authors used the 1:1 ratio of autogenous bone to DBB, rather than 1:2, to limit the excessive volume of the xenograft.

Feinberg et al.$^{2,3}$ revealed that hydroxyapatite particles delayed tooth eruption in animal models. Several studies$^{4,19,20,25}$ demonstrated that allogenic and xenogenic bone grafts, including DBB, had no inhibitory effect on tooth eruption or orthodontic tooth movement in vivo. A case report demonstrated the successful tooth movement, by orthodontic force, through the alveolar cleft region 6 months after grafting with a mix of decalcified freeze-dried bone allograft and a granular bioactive glass graft material in the ratio 1:1$^{30}$. This study confirmed that there was no interference of the canine eruption.

**Fig. 3.** Occlusal radiographs of two patients in group II showing progression of canine eruption (arrows) through the grafted area (a) 1 day postoperative, (b) 6 months postoperative and (c) 12 months postoperative.

**Fig. 4.** The canine (arrow) could be moved by orthodontic force.
process in patients for spontaneous or orthodontically assisted eruptions.

Several studies have recommended computed tomography (CT) for evaluating bone graft quantities because of the clear advantages in reproducibility and the three-dimensional images. The disadvantages of CT are the higher radiation exposure and the cost. This study simplified the method for evaluating bone graft quantities by using intraoral radiographs. Plain radiographs only demonstrate details in two dimensions, but are economical and produce less radiation than CT. Using computer software, the boundary of the grafted cleft sites can be delineated and the density of bone grafts in these areas can be determined. To detect changes in bone graft quantities at each time interval, the outline of the bone graft area from the first postoperative radiograph is superimposed on subsequent radiographs for each patient in the same position. For this purpose a constant position has to be achieved. A simple individual custom-made film holder was made for each patient to reproduce the spatial position between the radiographic beam and the film. The aluminium step wedge attached to the films provided precision for the digital image analysis of bone density using the calibration technique. The wedge has been widely used to calibrate the grey level of the radiographic image for subtracted interpretation. This method can reduce the error of different radiation exposures and film processing. The method for measuring the bone graft height was modified from Long et al. and Rosenstein et al. The heights were measured and demonstrated as a percentage of bone coverage of the tooth roots adjacent to the cleft site rather than in millimetres, to prevent radiographic shortening and elongation of the reference points.

The radiographic results demonstrate that the bone graft density of both groups rapidly decreased within the 6 months after grafting, then became stable. This implies that a rapid remodelling process occurred immediately after grafting and maturation of the cortical structure was complete within 6 months. This pattern was similar to that of other studies, which suggested that the pattern of bone graft remodelling would be complete within 6–12 months. Autogenous bone graft or a composite graft of autogenous bone and DBB showed the same pattern of bone remodelling, which could come mainly from the effect of autogenous bone remodelling.

Several factors influence the resorption of the bone graft including functioning of the presented tooth or the eruption of the tooth adjacent to the graft, tension of the mucoperiosteal flaps covering the graft and origin of the graft. The effect of the functioning tooth on bone resorption is controversial. Hosma et al. recommended that the graft site should be restored by a functioning tooth to prevent further bone resorption. Long et al. suggested that the pattern of canine eruption did not affect the success of bone grafting. The present study found that the decrease of the bone graft height related to canine eruption and the starting site of orthodontic tooth movement. Tooth eruption and functional tooth movement might affect bone graft resorption. The volume of bone graft in both groups could be maintained during the period of spontaneous tooth eruption and orthodontically assisted tooth eruption.

In conclusion, the protocol of combining DBB with autogenous bone graft from iliac crests in the ratio 1:1 was introduced for repairing alveolar cleft defects. Its efficacy was comparable with that of autogenous bone alone in terms of bone remodelling and tooth eruption, either spontaneous or orthodontically assisted. This technique significantly reduced the amount of autogenous bone required from the crest, patient morbidity and hospitalization, so it could be considered as an alternative technique for treatment of alveolar cleft.

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Competing interests
Not declared.

Ethical approval
0521.1/1.03/0397. This research has been approved by the Ethics Committee, Faculty of Dentistry, Prince of Songkla University, Songkhla, Thailand.

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