**Abstract:** The effect of the geometric structure of artificial biomaterials on skull regeneration remains unclear. In a previous study, we succeeded in developing honeycomb $\beta$-tricalcium phosphate ($\beta$-TCP), which has through-and-through holes and is able to provide the optimum bone microenvironment for bone tissue regeneration. We demonstrated that $\beta$-TCP with 300-$\mu$m hole diameters induced vigorous bone formation. In the present study, we investigated how differences in hole directions of honeycomb $\beta$-TCP (horizontal or vertical holes) influence bone tissue regeneration in skull defects. Honeycomb $\beta$-TCP with vertical and horizontal holes was loaded with BMP-2 using Matrigel and Collagen gel as carriers, and transplanted into skull bone defect model rats. The results showed that in each four groups (Collagen alone group, Matrigel alone group, Collagen + BMP group and Matrigel + BMP-2), vigorous bone formation was observed on the vertical $\beta$-TCP compared with horizontal $\beta$-TCP. The osteogenic area was larger in the Matrigel groups (with and without BMP-2) than in the Collagen group (with and without BMP-2) in both vertical $\beta$-TCP and horizontal $\beta$-TCP. However, when BMP-2 was added, the bone formation area was not significantly different between the Collagen group and the Matrigel group in the vertical $\beta$-TCP. Histological finding showed that, in vertical honeycomb $\beta$-TCP, new bone formation extended to the upper part of the holes and was observed from the dura side to the periosteum side as added to the inner walls of the holes. Therefore, we can control efficient bone formation by creating a bone microenvironment provided by vertical honeycomb $\beta$-TCP. Vertical honeycomb $\beta$-TCP has the potential to be an excellent biomaterial for bone tissue regeneration in skull defects and is expected to have clinical applications.

**Keywords:** honeycomb $\beta$-TCP; bone tissue regeneration; bone microenvironment; Vertical and Horizontal holes; geometrical structure