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Distinct mechanisms in maintaining calvaria and long bone mass in adult mouse
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Objective: Among diverse anabolic mechanical stimuli, which bones may experience, we have previously reported in vitro that signals downstream of stretching were processed in osteocytes resulting in bone formation while those downstream of low intensity, high frequency pulsed ultrasound (LIPUS) were processed in osteoblasts resulting in differentiation. It has been reported by others that disuse osteoporosis by bed rest affects long bones but not skull bones. To evaluate distinct mechanisms in the different responses, we isolated osteoblasts and osteocytes and analyzed mechanotransduction pathways.

Methods: Osteogenic cells were isolated from 16-week-old C57BL/6j mouse lower leg and calvarial bone chips by sequential treatments with collagenase and EGTA: osteoblasts from repeated collagenase digestion and osteocytes, after EGTA treatment. Cells were exposed to mechanical stimuli after one-week culture either by stretching in a FlexCell strain unit (osteocytes) or by exposing to LIPUS (osteoblasts).

Results: In both isolated long bone osteoblasts and osteocytes, mechanical stimulation resulted in upregulated message levels of component molecules in mechanotransduction pathways such as Wnt1 and 3a, FZD, and COX-2. By stretching, upregulation of DMP-1 and downregulation of SOST/sclerostin, two reported mechanosensitive osteocyte markers, reproduced the response in loaded long bone. On the other hand, mouse long bone osteoblasts responded to LIPUS with elevated levels of DMP-1 and SOST/sclerostin, suggesting that LIPUS accelerated differentiation of osteoblasts to osteocytes. The above mentioned machinery molecules in mechanotransduction as well as SOST/sclerostin, a Wnt/β-catenin-pathway inhibitor, behaved similarly in the stimulated calvarial cells. Basal expression levels in osteoblasts, however, are generally much higher in calvaria than in the long bone.

Conclusions: Our results suggested that maintaining long bone mass in adult mouse requires mechanical stimuli but that calvarial bone relies on some other pathway(s) as a default mechanism.

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Sequential remodeling and loss of epithelial basement membrane type IV collagen α chains in the intraepithelial neoplasia (CIN) and squamous cell carcinoma of the uterine cervix
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Objective: The destruction of the basement membrane (BM) is the first step in cancer cell invasion and metastasis. Type IV collagen is a major component of the BM and is composed of six genetically distinct α(IV) chains: α1(IV) to α6(IV). The loss of α5/α6(IV) chains from the epithelial BM at the early stage of cancer cell invasion has been reported in several types of cancer (1-3). However, the sequential remodeling or loss of α(IV) chains in the BM of intraepithelial neoplasia (CIN) and squamous cell carcinoma (SCC) of the uterine cervix remains to be unknown.

Method: The expression of α(IV) chains were immunohistochemically examined in 60 cases of biopsy and resected samples with CIN and SCC of the uterine cervix.

Results: In CIN 1-2 (mild to moderate dysplasia), both α1/α2(IV) and α5/α6(IV) chains were linearly expressed in the BM of the squamous epithelium. However, in CIN 3 (severe dysplasia/carcinoma in situ), sequential remodeling in the BM of the squamous epithelium were observed that severe dysplasia expressed both α1/α2(IV) and α5/α6(IV) chains in the BM, and that carcinoma in situ expressed only α1/α2(IV) chains in the BM. Interestingly, these transitional zone of the sequentially remodeled BM from α1/α2(IV) and α5/α6(IV) to α1/α2(IV) chains was confirmed in the BM of CIN3 lesion.

Conclusions: The sequential remodeling of type IV collagen α chains of the BM of the uterine cervical cancer seems to be closely related to cancer development preceded by cancer cell invasion.

Reference