452
VITAMIN D ENHANCED THE OSTEOGENIC RESPONSE OF ADOLESCENT IDIOPATHIC SCOLIOSIS PRIMARY OSTEOBLASTS TO VIBRATION

Jiajun Zhang[a,b,c], Wayne Yuk-wai Lee [a,b,c], Huanxiong Chen [a,b,c], Louis Wing-hoi Cheung [a,b], Bobby Kin-wah Ng [a,b,c], Jack Chun-yiu Cheng [a,b,c], Tsz Ping Lam [a,b,c]
[a]*Department of Orthopaedics and Traumatology, The Chinese University of Hong Kong, Shatin, NT, Hong Kong*
[b]*Joint Scoliosis Research Center of the Chinese University of Hong Kong and Nanjing University, Hong Kong*
[c]*SH Ho Scoliosis Research Laboratory, The Chinese University of Hong Kong, Shatin, NT, Hong Kong*

Introduction: Adolescent idiopathic scoliosis (AIS) is a complex rotational spinal deformity affecting children during their pubertal growth spurt. Unclear etiopathogenesis restricts current treatment regimes which are targeting the complication of spinal deformity. AIS is associated with systemic low bone mineral density (BMD). In 2005, we reported that low BMD was a significant prognostic factor for curve progression (OR = 2.3). These observations led us to investigate if bone mineralization is a novel therapeutic target for the clinical management of AIS. Our randomized controlled trial (RCT) showed anabolic bone effects of whole body vibration (WBV) in AIS girls which was more effective at sufficient serum Vit-D level. With the well-established primary osteoblasts culture, we aimed to investigate the role of Vit-D in modulating the effect of WBV on osteoblasts activity.
Subjects and method: In this in vitro case-control study, primary osteoblasts were isolated from iliac crest bone biopsies harvested from AIS patients (n = 10) and non-AIS control (n = 5) subjects intra-operatively. Osteoblasts were treated with 0, 10 nM and 100nM of 1, 25(OH)2Vit-D3 (Vit-D3) with or without concurrent vibration treatment (0.3g, 35 Hz for 20 minutes daily). Cell metabolic activity was determined by Alamar blue. Gene expression of representative osteogenic markers was detected by qPCR. Mineralization ability was determined by alkaline phosphatase (ALP) staining and Alizarin red staining. Semi-quantitation was done for independent *t* test analysis.
Results: Despite similar metabolic activity, AIS osteoblasts exhibited lower intrinsic ALP activity and calcium nodules formation. Vit-D treatment did not render additive effect on calcium nodules formation in control group under vibration. On the contrary, 100nM Vit-D positively enhanced the osteogenic effect of vibration in AIS osteoblasts. At transcription level, Vit-D promoted the vibration-induced upregulation of *Spp1*, *Bglap* and *ALP*, but not *Col1*, in AIS osteoblasts in a concentration-dependent manner. Vit-D co-treatment also promoted the mRNA level of *VDR* and *LRP5* which might partly underlie the positive effect of Vit-D on the anabolic effect of vibration.
Discussion and conclusion: The more sensitive osteogenic response of AIS osteoblasts to concurrent treatment of Vit-D and vibration might be part of the underlying mechanisms of the modulating effect of Vit-D level on anabolic effect of vibration therapy in previous clinical trial. The present study was limited by osteoblasts model which cannot represent the entire mechanosensation system in bone. With a newly established 3D osteocytes model, we are looking at the effects of Vit-D on vibration in AIS primary osteocytes culture.
Funding/support: This study was supported by CUHK Direct Grant (2012.1.034).
http://dx.doi.org/10.1016/j.jot.2016.06.128

413
SURGICAL CORRECTION OF DESTROPHIC NEUROFIBROMATOSIS TYPE 1 SCOLIOSIS—THE CORRELATION OF SURGICAL STRATEGY AND RADIOGRAPHICS

Qiang Jie, Xiong Zhao, Qingda Lu, Xiaolei Zhao, Yishan Ma, Long Sun, Wei Lei
Xijing Hospital, the Fourth Military Medical University, China

Objective: Dystrophic spinal curve in neurofibromatosis type 1 (NF-1) is a complex problem. How to correct the curve and stabilize the spine with the most appropriate and safe method without causing neurologic injury? The purpose of the present study was to evaluate the correlation of surgical strategy, radiological findings and clinical outcome after correction of dystrophic spinal curves in NF-1 based on pre-operation precise scheme.
Methods: This retrospective study consisted of 32 NF-1 child patients with different spinal deformities treated between 2001 and 2013 in our department. Preoperative X-ray, 3D-CT and MRI were performed to evaluate the deformities of dystrophic scoliosis accurately. All patients were treated with posterior instrumented fusion or combined with anterior release by using hybrid fixation. According to the anatomical development situation of each patient's pedicles, laminars and the transverse processes, we chose different fixations including screws, hooks and wires. The radiographics and clinical outcomes of surgical correction were evaluated post-operatively.
Results: The average pre-operative scoliosis Cobb angle was 63° (range 45°—110°) corrected to an average of 24° (range 5°—42°) post-operatively. The average pre-operative kyphosis was 52° (range -10°—84°) corrected to an average of 26° (range 0°—36°) post-operatively. The apical vertebral body rotation was corrected by an average of 56%.
Conclusions: Pre-operative radiographics findings and precise surgical plans and careful manipulation are crucial to keep neurological intact, and through which the satisfactory clinical effects can be achieved in NF-1 scoliosis.
http://dx.doi.org/10.1016/j.jot.2016.06.129

478
A HIGH-CALCIUM, HIGH-PHOSPHORUS, HIGH-LACTOSE DIET RESCUES THE INTERVERTEBRAL DISC DEGENERATION PHENOTYPE INDUCED BY THE DEFICIENCY OF 1,25(OH)2D3

Hao Xu [a], Daofang Ding [a], Dengshun Miao [b], David Goltzman [c], Qi Shi [a], Qianqian Liang [a], Yongjun Wang [a]
[a]*Longhua Hospital, Shanghai University of Traditional Chinese Medicine, Shanghai, China*
[b]*The State Key Laboratory of Reproductive Medicine, The Research Center for Bone and Stem Cells, Department of Anatomy, Histology and Embryology, Nanjing Medical University, Nanjing, China*
[c]*Calcium Laboratory, Royal Victoria Hospital, McGill University Health Centre, Montreal, Quebec, Canada*

Background: Intervertebral disc degeneration (IVDD) is an important source of neck or low back pain, a common cause of musculoskeletal disability worldwide, and imposes high economic burdens. Age, obesity, height, smoking history, occupation, and lumbosacral structure are reportedly the main risk factors of lumbar IVDD, but underlying biological mechanisms remain unclear. In the current study, we aim to investigate the role and possible mechanisms of 1,25(OH)2D3 deficiency in the development of IVDD by applying 1α (OH)ase$^{-/-}$ mice and their wild-type litter-mates on a regular or rescue diet from weaning until 7 months of age.
Methods and Results: 1α (OH)ase$^{-/-}$ mice were generated through breeding of heterozygous mice. The genotype of the mice was confirmed by PCR using mouse tail samples. Wild-type littermates were used as control animals in all experiments. The use of animals in this study was approved by the Institutional Animal Care and Use Committee of Shanghai University of traditional Chinese medicine. Ten pairs of 1-month-old matched 1α(OH)ase$^{-/-}$ and wild-type littermates were used in this study. After weaning, 5 pairs of them were fed with rescue diet (applied by SLAC Shanghai laboratory animals limited liability company) containing 2% calcium, 1.25% phosphorus, and 20% lactose until they were 6 months old. Mice were sacrificed at the age of 7 months for examination by histology, IHC, qPCR and micro-CT. Histology revealed growth plate thickness increase in 1α(OH)ase$^{-/-}$ mice with indistinct tide mark between endplate and the fibrous loop. Q-PCR showed IL-1, IL-6, MMP-3, MMP13, CATHSK, Adampts 5 expression increased in the 1α(OH)ase$^{-/-}$ mice group. IHC staining revealed collagen type II expression decreased, collagen type X increased, and Nitege expression increased in the 1α(OH)ase$^{-/-}$ mice. However the 1α(OH)ase$^{-/-}$ mice feed with high calcium, high phosphorus, high lactose diet (rescue food, RF) group showed the almost the same phenotype with wild type.
Discussion and Conclusion: We used a 1,25(OH)2D3 deficiency mouse model and identified a novel etiology of intervertebral disc degeneration, which could be rescued by a high-calcium, high-phosphorus, high-lactose diet. First, hypertrophy chondrocyte number increased and arranged disorder in the growth plate of CYP27B1 KO mouse. Second, tide mark showed indistinctly, fibrous loop ossification, and extracellular matrix loss in the endplate of CYP27B1 KO mouse intervertebral disc. Third, catabolic factors as IL-1, IL-6, MMP3, MMP13, Cathepsin K, and Adampts 5 expression increased in the CYP27B1 KO mouse intervertebral disc. Fourth, COL II expression increased, COLX and NITEGE expression decreased in the CYP27B1 KO mouse intervertebral disc. Fifth, osteoporosis showed in the 4th vertebrae of CYP27B1 KO mouse. Lack of nutrition could be the reason for the degeneration of intervertebral disc. Since nutrition transportation to the intervertebral disc could be blocking by the thick hypertrophic growth plate. The reasons for the thick hypertrophic growth plate may be as followed. One is that 1,25(OH)2D3 deficiency may induce apoptosis blocking in the growth plate of the CYP27B1 KO mouse, resulting hypertrophic chondrocyte accumulation. The other may be that hypertrophic growth plate can be also resulted from force alteration by osteoporosis in the vertebrae. However, a high-calcium, high-phosphorus, high-lactose diet can not only inhibit intervertebral disc degeneration and vertebral osteoporosis. Therefore, high-calcium, high-phosphorus, high-lactose diet could be a potential therapy to treat with 1,25(OH)2D3 deficiency patients to prevent intervertebral disc degeneration.
http://dx.doi.org/10.1016/j.jot.2016.06.130