Effects of a Growth Modulation Device on the Immature Goat Spine

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Abstract: *Background Context*: A variety of spinal instrumentations has been explored to treat the early-onset scoliosis in either children or animal model. A growth modulation device (GMD) with the conception of tilting in the coronal and sagittal plane while allowing a longitudinal sliding was developed. We hypothesize our GMD will lead to higher compressive forces induced by the GMD between intervertebral regions lead to changes of vertebral endochondral ossification.

Purpose: We presented a new GMD, which was evaluated by observing morphological changes of the spine within and adjacent to the instrumented segments, having a radiographic review, and performing a histological analysis. In addition, we analyzed the growth rate and pattern of the asymmetric spine.

Study Design: A perspective in-vivo study of a novel growth modulation device (GMD).

Methods: The four skeletally immature goats were approached via a standard thoracotomy from T6 to T10. The right side of the vertebral body was exposed and instrumented with the GMD. One goat was sacrificed at 1, 2, 3, and 6 months following spinal surgery, respectively. Radiographs and histomorphometry were performed.

Results: The average Cobb angle (T2-T12) increased from 3º to 10º. The length (T6-T10) increased from 8.9cm to 12.6cm. Bony and cartilage density was greater on the instrumented side. The heights of physis were reduced (20%-33%) on the anterior aspect of the instrumented side, but less on the posterior (12%-21%). Growth spurts are greatly reduced within the instrumented segments and growth remains constant over the animal's life, but on the vertebrae far from the instrumentation the growth increases linearly, and the growth rate is higher.

Conclusions: The GMD yields a controllable impact on the bony and cartilage density, and endochondral ossification height for a growth modulation.

Keywords: Growth modulation, fusionless device, growth plate, immature goat spin.

1. INTRODUCTION

Early onset scoliosis presents a unique treatment challenge due to its progressive nature. While bracing has effectiveness in preventing further progression in a subset of patients, spinal fusion is the only current method reversing and preventing curve progression for large curves. However, spinal fusion has significant drawbacks including spinal and thoracic cavity growth restriction. Further understanding of normal growth and the effects of scoliosis and spinal fusion have led to alternative growth modulation strategies. Growth modulation devices (GMD), in contrast to fusion, are used to harness a patient's scoliotic growth and redirect it to halt or reverse the deformity.

Numerous growth modulation strategies are currently being employed and investigated. Compression based implant methods achieve correction via the Hueter-Volkmann principle, which states that bony expansion of the physis is slowed by mechanical compression and is increased by distraction. One compression method, vertebral stapling, was first described nearly 60 years ago [1], and its ability to limit and modulate curve progression is promising for both animal and clinical models [2-5]. More recently, a compression technique involving vertebral bone anchors with a flexible ligament tether implanted along the convexity of the curve have been used. Animal studies have shown that tethers are able to successfully induce both scoliosis and kyphosis [6-8] and have the ability to correct coronal deformities [9-10].

There are also a few studies that have examined the histologic changes induced in vertebra when asymmetric compressive forces are applied. Newton determined that there were changes in proteoglycan synthesis, collagen type distribution, and disc thickness in tethered vertebral discs when compared to discs without a compressive force applied [11]. It has been shown that sustained compression forces cause a reduction in chondrocyte numbers [12], volumes [13-14], and physeal thinning [13-16]. Compressive forces may also lead to an alteration of chondrocyte column alignments [13, 15, 17]. For this study, we introduce a new GMD, evaluate its effects on goat spine growth by histological and radiographic means, and compare the heights of the chondrocyte zones, disc structure, and bone and cartilage density between the instrumented and uninstrumented side of the vertebral body.

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2. MATERIALS AND METHODS

2.1. Subjects

Four 2- month-old goats used in this study. A goat model was chosen due to its lordoscoliotic spine, neural elements being located away from the tether, and comparable dimensions to a juvenile child [18].

2.2. GMD

Our GMD was developed prior to the start of this study. It consists of a vertebral body screw, a locking screw and cap, and an adjustable plate to apply a compressive force between the screw heads (Figure **1a**). Many of the previous GMDs use rigid or flexible tethers to correct scoliosis [11-13], but this may be a limitation to treat adolescent idiopathic scoliosis (AIS), as they may limit the growing potential of the subjects or cause overall growth arrest. In addition, our screws are also able to slide along the plates to allow for spinal rotation and growth along the longitudinal axis. At the same time, our adjustable GMD will allow for a constant compressive force applied to the spinal process, while also allowing for normal longitudinal and rotational spinal growth in pediatric patients.

2.3. Surgical Protocol

Protocol for the study was approved by the Institutional Animal Care and Use Committee, Medical College of Wisconsin. Animals were anesthetized and intubated and then a right-sided thoracotomy was performed between the 6^{th} and 7th ribs was performed. The lung was retracted, and the pleura and segmental vessels were cauterized over 5 consecutive vertebrae (T6-T10). During implantation, care was taken to avoid damage to the intervertebral discs and epiphyseal growth plates (Figure **1a**). The appropriate levels and screw position were confirmed with intraoperative fluoroscopy (Figure **1b**). One goat was sacrificed at 1 month, 2 months, 3 months, and 6 months following spinal surgery. Spines were immediately harvested following goat sacrifice. A macroscopic evaluation of the instrumented spine segments was performed looking for dislodgement or failure of the GMD, bony abnormality, debris, or spinal segment fusion.

2.4. Radiographic Analysis

Dorsoventral and lateral radiographs of the thoracic spine were taken on a monthly basis (General Electric, Waukesha, WI, USA). The coronal and sagittal deformity was measured from T2 to T12 using standard Cobb angle techniques. In addition the length between T6 and T10 was measured. The average bony density of the vertebrae on both instrumented and uninstrumented side was determined by radiography.

2.5. Histologic Analysis

The implants were removed from the spines after the harvest and visual inspection. The specimens were prepared for subsequent analysis. Histologic analysis was performed in the apical spinal segments (2 adjacent vertebrae and the first intervening disc) using standard hematoxylin and eosin staining of a microdermal section.

Using an Olympus microscope (Olympus America, Inc., Center Valley, PA) and imaging software (MicroSuite™, Olympus America, Inc., Center Valley, PA), Microscopic analysis was performed on the harvested and prepared sections. Initially, the

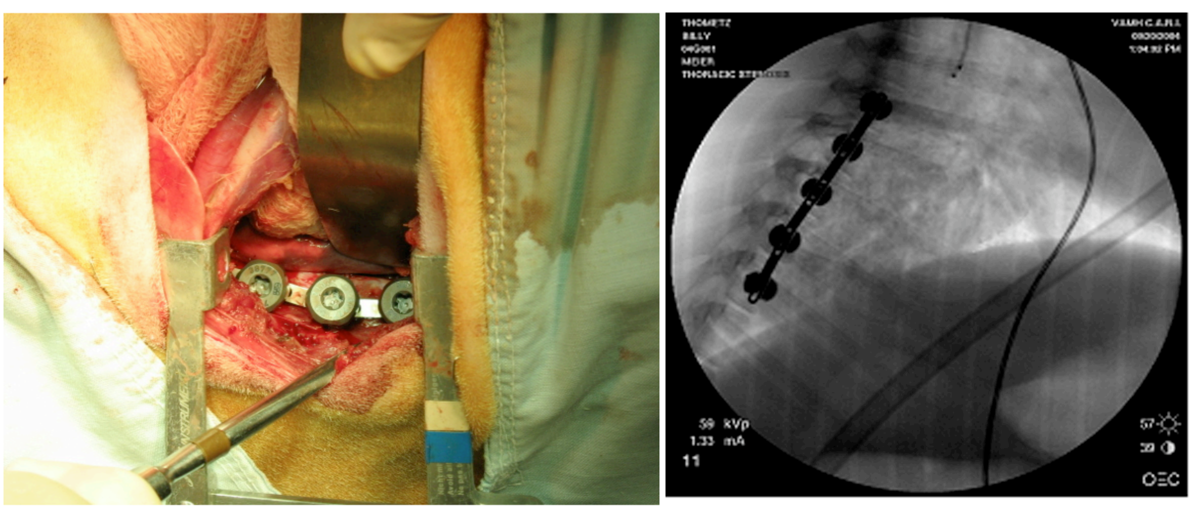

Figure 1: a. The vertebral bodies from the 6th to the 10th thoracic vertebra on the right side were exposed and instrumented with the GMD. **b**. Intraoperative fluoroscopy confirmed satisfactory position of the GMD on the right side of the vertebral body.

microscopic anatomy of the vertebral disc and physis was examined for pathologic lesions or degenerative changes. The heights of the chondrocyte zones, consisting of the reserve, proliferative, and hypertrophic area in both the anterolateral and posterolateral aspects of the physis on both the instrumented and uninstrumented side were measured. Using the Window-based software we easily identified the region of interests of the physis (top, middle, bottom), its densities of the instrumented side and uninstrumented side were measured with a Grey scale level [19].

Statistical Analysis

Because of small sample size of this study, a descriptive analysis (SPSS. Inc. Chicago, USA and Math Works. Inc. Massachusetts, USA) was performed to describe changes of Cobb angle before the GMD and 1,2,3, 6 months following GMD. Radiographic bone density, histological cartilage density, and histopathology assessment were delineated and compared between the instrumented and uninstrumented side on either the vertebral body or the physis.

RESULTS

Macroscopic evaluation of the instrumented spine segments showed no dislodgement or failure of the GMD, no obvious bone abnormalities, no debris, and no spinal segment fusion.

The average Cobb angle in the thoracic spine increased over time from 3º to 10º, with a maximum of 12 º (Figure **2**). There was no crankshaft findings in the X-Rays. The length between the middle point of T6 upper endplate and T10 lower endplate steadily increased from 8.9 cm to 12.6 cm over the 6 month period. The average bone density on the instrumented side of the vertebrae was greater than the un-instrumented side between T6 and T10 over the months (Figure **3**).

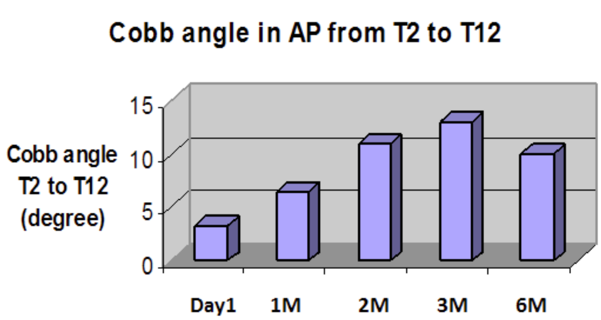

Figure 2. Changes in the average Cobb angle in thoracic spine from day 1 to 6 months following the anterior spinal surgery with the GMD.

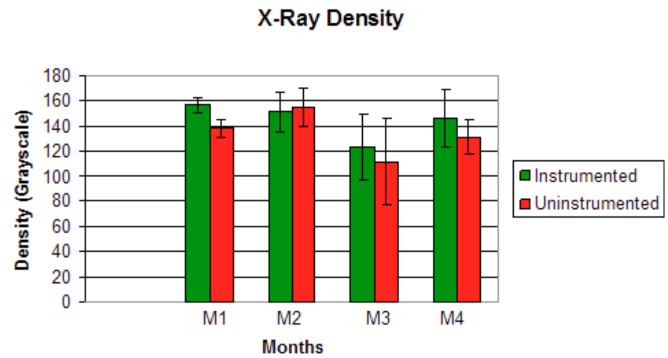

Figure 3: A slight increase (mean and SD) in bony density on the instrumented side as compared to uninstrumented side at 1, 2, 3, and 4 months post-surgery.

Microscopically, all growth plate zones were present. No specific pathological lesions were noted in any growth plate. On average, the heights of the three endochondral zones were decreased by 9.4% on the instrumented side of the anterior vertebral physis. (Figure **4**, Table **1**). Particularly, the proliferative and hypertrophic zones had the highest decrease in percentage (20 and 23% respectively). A similar effect occurred to a lesser extent on the posterior aspect of the physis (1.3%). There were no discrete histological changes within the disc between the two sides. The average cartilage density on the instrumented side of the vertebrae was also greater between T6 and T10 at the 6 month post-surgery mark (Figure **5**).

DISCUSSION

Our experiment utilized a novel growth modulation device (GMD) in a goat model to induce a scoliotic curve in an otherwise normal spine. The purpose of the study was to analyze the effects of our GMD on goat spine models through recording radiographic and histologic changes that were induced in the vertebral physis. Specifically, the heights of the chondrocyte zones, disc structure, and bone and cartilage density between the instrumented and un-instrumented side of the vertebral body were compared. This is a preliminary study of our design, and the efficacy of our design will be further evaluated by conducting a finite element analysis of our GMD (FEA), analyzing additional growth parameters, and comparing our GMD to some of the previously used GMDs.

In our preliminary study the instrumented spine segments showed no dislodgement or failure of the GMD, no obvious bone abnormalities, no debris, and no spinal segment fusion. Prior to this study, a previous study demonstrated the improvement of using tethers on goat models compared to vertebral body staples [9]. The goats with the bone-anchor-tether construct

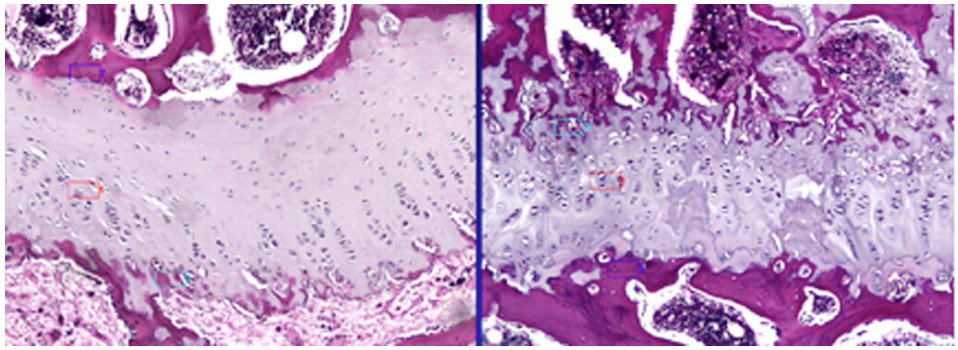

Figure 4: Comparison of three chondrocyte zones on the instrumented side (right) and uninstrumented side (left) in the anterior vertebral physis of T6: decreased height of the proliferative and hypertrophic zones on the right (HE, X 10).

Table 1: Comparison of Heights of Three Chondrocyte Zones in the Growth Plate between Instrumented and Contralateral Side (μm)

Anterior aspect of physis	Instrumented Mean Value	Uninstrumented Mean Value	% change
Reserve	173.8	144.3	12.5%
Proliferative	160.4	199.5	-19.6
Hypertrophic	132.4	171.3	-22.7%
Total	466.6	515.1	-9.4%

demonstrated a greater prevention of curve progression and greater pullout strength than the vertebral body staple construct both initially and at study completion. This increased pull out strength is thought to be one of the theoretical benefits of using a bone-anchor tether implant over a vertebral body staple [9].

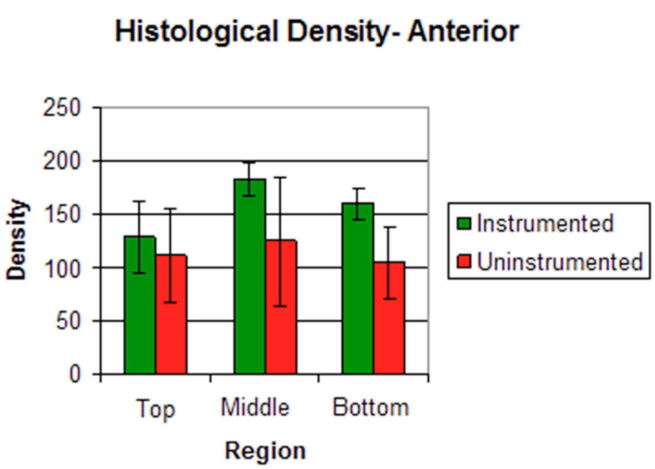

Figure 5: Comparison of cartilage density on the instrumented side with uninstrumented side on the anterior part of the vertebral physis (divided by top, middle, and bottom layer) using a histomorphometry analysis (Gray scale) 6 months postsurgery.

There have been several studies examining the radiographic, clinical, and biomechanical effects of asymmetric loads applied to the vertebral body physis. Scoliosis progression is governed by the Hueter-Volkmann principle. This states that bony expansion of a physis is slowed by mechanical compression and increased by distraction. This principle has been verified in numerous studies of long bone angular deformity correction in growing children [20-22]. The development of scoliotic curve is a self promoting process: spinal curvature leads to non-uniform compressive loads applied to the vertebra which in turn causes asymmetric growth on the compressed side leading to further progression of the curve. Rat-tail studies have supported this by showing that when angular compressive forces are applied to the tail a wedge shaped vertebra can be induced similar to those seen in scoliotic curves [6,7]. Studies in calves and sheep have shown that when tethers were placed to modulate coronal or sagittal growth both scoliosis and kyphosis were successfully induced [6,7].

Our study supports the ability of asymmetric compression having the ability to induce curve formation and progression and cause physeal changes. Radiographic analysis of our spinal models demonstrated an increase of the Cobb angle in the thoracic spine from 3º to 11º, followed by a decrease to 9º. The sudden decrease of the Cobb angle may have occurred due to the fact that only one subject's Cobb angle was not sacrificed before the 6 month mark. Other time frames were an average of a few subjects. Average bony density and cartilage density on the instrumented side of the vertebrae were greater than on the un-instrumented side between T6 and T10. This demonstrates the Heuter-Volkmann principle, increased loads on the instrumented sides lead to decreased growth inducing curve formation. The length between T6 and T10 increasing from 8.9 cm to 12.6 cm over 6 months. This demonstrates the modulation, rather than full arrest of growth.

Few studies have examined the histologic changes induced by asymmetric loads on the vertebral body physis. It is known that longitudinal growth of the physis starts with division of the chondrocytes in the proliferative zone, followed by increase in volumetric size of the chondrocytes in the hypertrophic zone, and ends with apoptosis of the chondrocytes leading to mineralization in the zone of provisional calcification [23,24]. It is thought that the number of new chondrocytes created in the proliferative zone and the magnitude of chondrocyte volume increase in the hypertrophic zone are the key factors that correlate with longitudinal growth of the growth plate [25].

It has been shown that compression leads to changes in the rate of physeal growth [25-28]. Increased compressive loading causes a reduction in chondrocyte proliferation [13,14], growth plate thinning in the proliferative [15] and hypertrophic [13-16] zones, and alters the alignment of chondrocyte columns [13,15,17]. Histologic analysis of our specimens confirmed these previous studies findings. Microscopically, all growth plate zones were present and no pathological lesions were noted in any growth plate. On average, the heights of the three chondrocyte zones were decreased approximately 9.4% between the instrumented side of the anterolateral vertebral physis, compared to the un-instrumented side. This was most evident for the proliferative and hypertrophic zones, with a 20 and 23% change respectively. A similar effect occurred to a lesser extent on the posterolateral aspect of the physis (1.3%). There were no discrete histological changes within the disc on instrumented compared to un-instrumented side.

This study analyzed a growth-modulation device that applied a gradually augmented compressive force on an immature goat spine. Using this device the growth of the spine could be modulated without producing fusion. Further histologic analysis demonstrated that the growth modulation device reduced the heights of the endochondral ossification zones and increased bone and cartilage density on the instrumented side of the vertebral body. This was done without producing any discrete pathologic changes within the disc. This study provides an insight into the biophysiologic response of the immature spine to a fusionless device over a short period of time.

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