Zygapophyseal Joint Adhesions After Induced Hypomobility

Gregory D. Cramer, DC, PhD,[a] Charles N.R. Henderson, DC, PhD,[b] Joshua W. Little, DC,[c] Clover Daley, BS,[d] and Thomas J. Grieve, DC[e]

Abstract

Objective: Adhesions (ADH) have been previously identified in many hypomobile joints, but not in the zygapophyseal (Z) joints of the spine. The objective of this study was to determine if connective tissue ADH developed in lumbar Z joints after induced intervertebral hypomobility (segmental fixation).

Methods: Using an established rat model, 3 contiguous segments (L4, L5, L6) were fixed with specially engineered, surgically implanted, vertebral fixation devices. Z joints of experimental rats (17 rats, 64 Z joints) with 4, 8, 12, or 16 weeks of induced hypomobility were compared with Z joints of age-matched control rats (23 rats, 86 Z joints). Tissue was prepared for brightfield microscopy, examined, and photomicrographed. A standardized grading system identified small, medium, and large ADH and the average numbers of each per joint were calculated.

Results: Connective tissue ADH were characterized and their location within Z joints described. Small and medium ADH were found in rats from all study groups. However, large ADH were found only in rats with 8, 12, or 16 weeks of experimentally induced intervertebral hypomobility. Significant differences among study groups were found for small ($P < .003$), medium ($P < .000$), and large ($P < .000$) ADH. The average number of medium and large ADH per joint increased with the length of experimentally induced hypomobility in rats with 8 and 16 weeks of induced hypomobility.

Conclusions: We conclude that hypomobility results in time-dependent ADH development within the Z joints. Such ADH development may have relevance to spinal manipulation, which could theoretically break up Z joint intra-articular ADHs. (J Manipulative Physiol Ther 2010;33:508-518)

Key Indexing Terms: *Zygapophyseal Joint; Tissue Adhesions; Chiropractic; Manipulation, Spinal*

A theoretical model of putative biomechanical/anatomical beneficial effects of spinal manipulation (Fig 1) begins with the theory that the zygapophyseal (Z) joints become hypomobile for a variety of reasons (eg, sedentary lifestyle, repetitive occupation-related activities, etc; Fig 1, Step 1).[1-4] The hypomobility can result in the development of intra-articular adhesions (ADH) and degenerative changes in the Z joints (Fig 1, Step 2).[3] Spinal adjusting is thought to gap the Z joints (Fig 1, Step 3) and break up ADH (Fig 1, Step 4), which may slow the degenerative processes in the hypomobile joints (Fig 1, Step 5).[1,4-6] Other mechanisms of spinal manipulation are also being investigated (eg, neurological).[7-9] The various models (mechanisms) are not mutually exclusive.

The biomechanical/anatomical model of spinal manipulation (Fig 1) is supported by previous studies. For example, spinal manipulation (adjusting) gaps the Z joints in healthy human subjects[4,5] (Fig 1, Step 3), and gapping is

[a] Professor and Dean of Research, Department of Research, National University of Health Sciences, Lombard, Ill.
[b] Associate Professor, Palmer Center for Chiropractic Research, Davenport, Iowa.
[c] Graduate Student, Department of Surgery, Center for Anatomical Science and Education, St. Louis University School of Medicine, St. Louis, Mo.
[d] Research Assistant, Palmer Center for Chiropractic Research, Davenport, Iowa.
[e] Research Faculty, Department of Research, National University of Health Sciences, Lombard, Ill.

Submit requests for reprints to: Gregory D. Cramer, DC, PhD, Professor and Dean of Research, National University of Health Sciences, 200 East Roosevelt Road, Lombard, IL 60510 (e-mail: *gcramer@nuhs.edu*).

Paper submitted October 30, 2009; in revised form June 7, 2010; accepted June 8, 2010.
0161-4754/$36.00

doi:10.1016/j.jmpt.2010.08.002

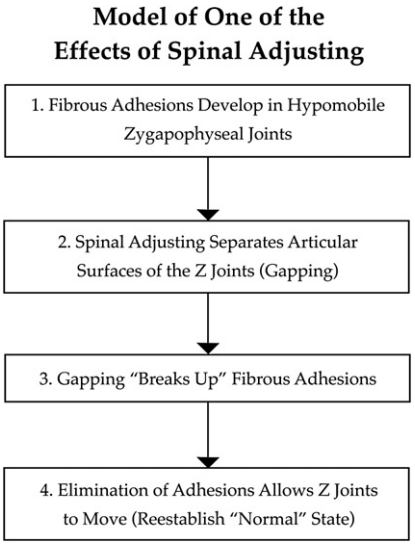

Fig 1. *Flowchart showing a model of putative beneficial anatomical/biomechanical effects of spinal manipulation. Putative neurological or other effects (eg, immunological effects) are not included in this flowchart. (Adapted with permission from Cramer et al[3] Journal of Manipulative and Physiological Therapeutics 2004;27:141-54.)*

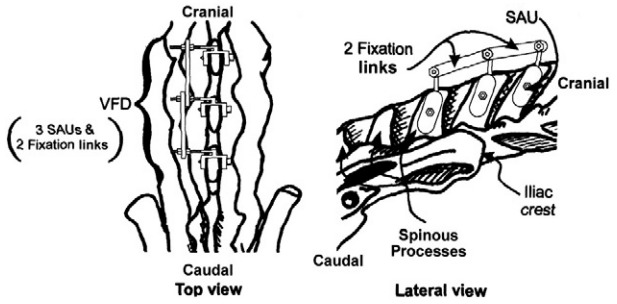

Fig 2. *External linking system (vertebral fixation device = VFD in illustration on the left) used in this study. Left illustration is a dorsal (top) view and right illustration is a right lateral view. The saddle-shaped SAUs fit over the lower three lumbar spinous processes. (Reprinted with permission from Cramer et al[3] Journal of Manipulative and Physiological Therapeutics 2004;27:141-54.)*

currently being assessed in subjects with acute and recurrent low back pain.[10] In addition, induced intervertebral hypomobility in the rat produces time-dependent spinal morphological changes (more changes with increased duration of hypomobility) such as Z joint articular surface degeneration (after 4 weeks of hypomobility) and osteophyte formation (after 8 weeks of hypomobility) (Fig 1, Step 2).[3] However, a description of ADH development within the Z joints (Fig 1, Step 2) has not been reported in the peer-reviewed literature, even though ADH have been demonstrated in hypomobile knee,[11,12] shoulder,[13] and temporomandibular joints (TMJ).[14] In fact, Hase[14]

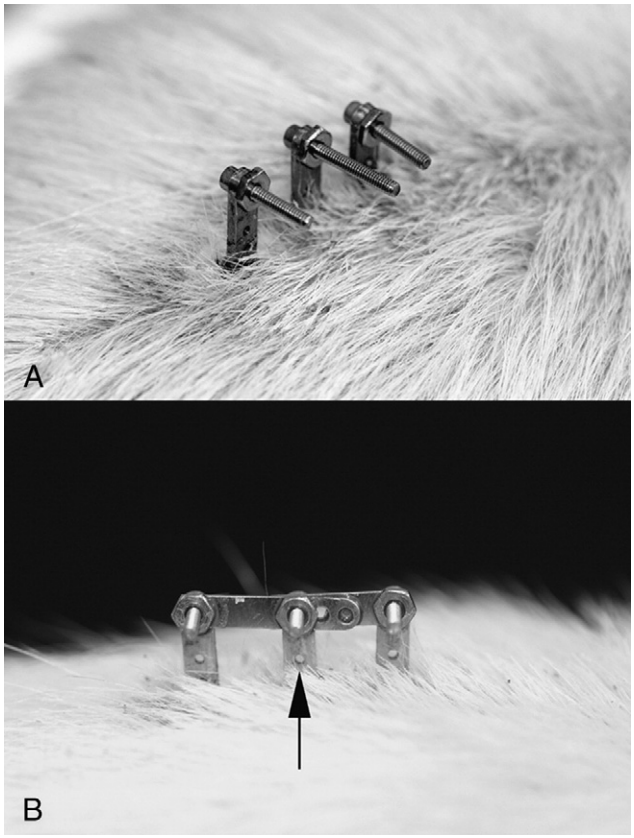

Fig 3. *The upper figure (A) is of one type of control animal (C_{LINK}, see text) and shows the vertical stems of three SAUs passing through the skin of the back. The lower figure (B) is an experimental group animal and shows the SAUs linked to create hypomobility. Linking was done for 4, 8, 12, or 16 weeks, depending upon the experimental group. The arrow indicates a hole in the neck of the SAU used for biomechanical testing in other studies (not used in the study reported here). (Adapted with permission from Henderson et al[16] Journal of Manipulative and Physiological Therapeutics 2007;30:239-45.)*

concluded that "A progressive maturation of ADH [thicker, more abundant] was observed which was directly related to the length of time of clinical symptoms of internal (TMJ) derangement." The TMJ is a modified fibrous joint that differs somewhat from the purely synovial, planar Z joints, and TMJ derangement is different than pure hypomobility (although hypomobility is frequently associated with TMJ derangement); however, Hase's findings and the findings of the other groups suggest that ADH develop in a time dependent manner in hypomobile joints. In addition, Laroche[15] et al used isotonic saline under pressure to separate the humeral head from the glenoid fossa of the shoulder joint (inducing joint gapping) to treat adhesive capsulitis, indicating that gapping of joints is a biologically plausible treatment for breaking up intra-articular ADH (Fig 1, Step 4).

In this study, a previously developed small animal model, the External Link Model[16-18] was used to evaluate

Table 1. *Animal and Z joint counts*

Groups	Link period or equivalent control period—No. of animals (no. of Z joints)[a]				
	4 wk	8 wk	12 wk	16 wk	Total
Controls	5 (16)[b]	7 (28)	6 (22)[b]	6 (24)	24 (90)
C_{LINK}	2 (8)	2 (8)	2 (7)[b]	2 (8)	8 (31)
C_{SAU}	2 (5)[b]	1 (4)	2 (8)	2 (8)	7 (25)
C_{SURG}	1 (3)[b]	4 (16)	2 (7)[b]	2 (8)	9 (34)
Experimental	5 (18)[b]	6 (23)[b]	3 (11)[b]	3 (12)	17 (64)

[a] Numbers to the left of parentheses are the number of animals examined for the time period. Numbers in parentheses are the number of Z joints analyzed in the group and time period.

[b] Indicates that some Z joints in the group and time period were not analyzed due to deformation during sectioning (optimally, 4 Z joints were analyzed per animal).

ADH formation in the Z joints of rats following induced intervertebral (segmental) hypomobility (fixation) at 4, 8, 12, or 16 weeks. These changes were then compared to the Z joints of control rats (3 control configurations).

Methods

The Institutional Animal Care and Use Committees of the National University of Health Sciences and the Palmer College of Chiropractic, Davenport, Iowa, approved this study.

External Link Model

Using the External Link Model,[16-18] 3 contiguous lumbar segments (L4, L5, L6—the rat has 6 lumbar vertebrae) were rendered hypomobile by means of specially designed and engineered spinal fixation devices (Figs 2 and 3). These devices, known as spinal attachment units (SAUs), were surgically implanted, like saddles, over the L4, L5, and L6 vertebral spinous processes. The "saddle" of each SAU was affixed by drawing its sides firmly together against the spinous process with a small screw. A small stem extended vertically from the saddle of each SAU and passed through the skin of the back to permit linking of SAUs at a later date (Fig 3). Following a 1-week surgical recovery period, the vertical stems of experimental group rats, but not control rats, were linked (yoked) together. Linking the SAUs completed the fixation induction procedure. A previous study demonstrated that this linking significantly decreases but does not completely eliminate intersegmental motion.[17] In these previous studies, the average intervertebral stiffness in control animals with no fixation was 14.52 ± 4.47 N/mm, and the average stiffness during hypomobility induction (with links in place) was 44.26 ± 11.06 N/mm.[17] The fixation device also significantly reduced extension of the spine (by 9.5°-18°) as measured by the angle of lines extended from the vertebral bodies of L4 and L6 from x-rays taken in full extension of control and fixation animals.[17] Flexion was not assessed in the study.

Control and Experimental Groups

Three groups of control animals were used. One control group (C_{LINK}, Fig 3A) had the SAUs implanted, but they were never linked in fixation. The second control group (C_{SAU}) underwent the surgical implant procedure, but SAUs were not implanted. The final control group (C_{SURG}) had no surgical procedure whatsoever. Z joints of 4, 8, 12, and 16 week linked experimental group rats were compared with the 3 groups of control rats that were age equivalent and survived for the same time periods.

Forty-one animals (24 control and 17 experimental) providing 154 Z joints were used in this study. Animal and Z joint counts are reported in Table 1. Some Z joints were lost to analysis due to deformation during sectioning.

At the end of the experimental or control period, the animals were harvested for study. The L4 through L6 vertebrae were removed en bloc, formalin fixed, and decalcified. These decalcified spine segments were then cut transversely through the L4, L5, and L6 vertebral bodies to isolate the L4-L5 and L5-L6 Z joint segments. All Z joint segments were embedded in paraffin and sectioned horizontally at 45 μm. Every 10th horizontal section through each Z joint pair (left and right) was stained in Ehrlich's hematoxylin, counter stained in light green, dehydrated, cleared, and mounted in Permount. The left and right L4-L5 and L5-L6 Z joints (ie, 4 joints per animal) were then systematically examined and photomicrographed under 10× and 20× brightfield microscopy.

Assessment of ADH

The Z joints were assessed for the presence of connective tissue ADH bridges within the joint space. An ADH was defined as connective tissue material located within the Z joint space and completely connecting two distinct Z joint structures (ie, superior articular process to inferior articular process, superior articular process to a synovial fold, or inferior articular process to a synovial fold: see Fig 4A and B). Adhesion sizes were graded by a trained observer as small (thin, threadlike), medium (intermediate thickness, not over 10% of a quarter region of the joint), and

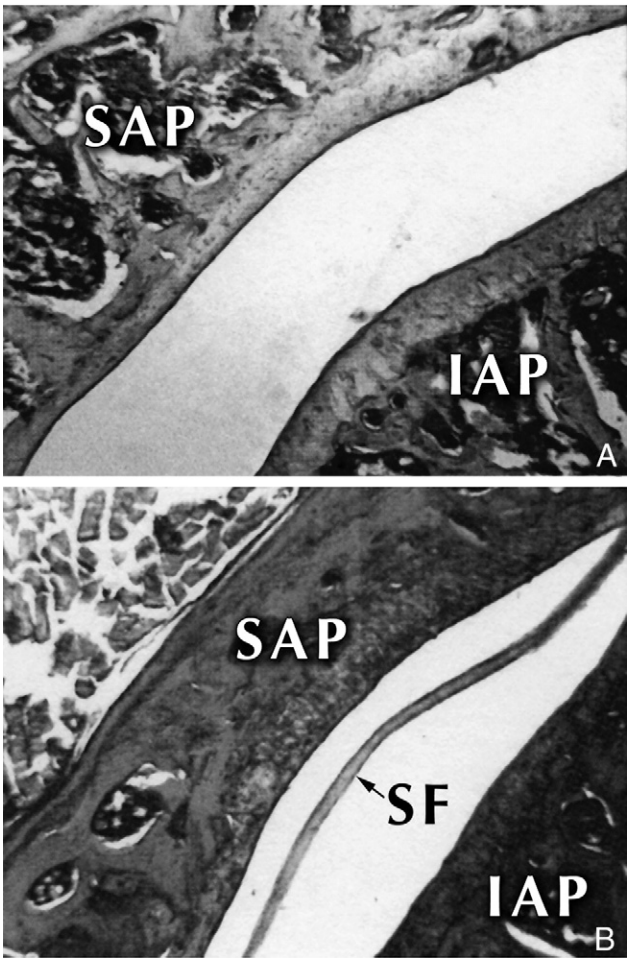

Fig 4. *Horizontal sections of Z joints without ADH. A, Superior (cephalad) articular process (SAP) and inferior (caudad) articular process (IAP) are shown. B, Notice the synovial fold (SF, arrow) within the center of the joint space.*

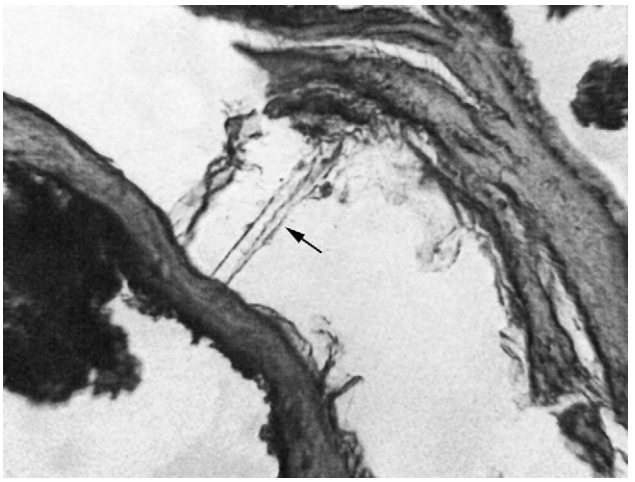

Fig 5. *Small ADH (original magnification ×20, arrow). Notice the thin, threadlike appearance.*

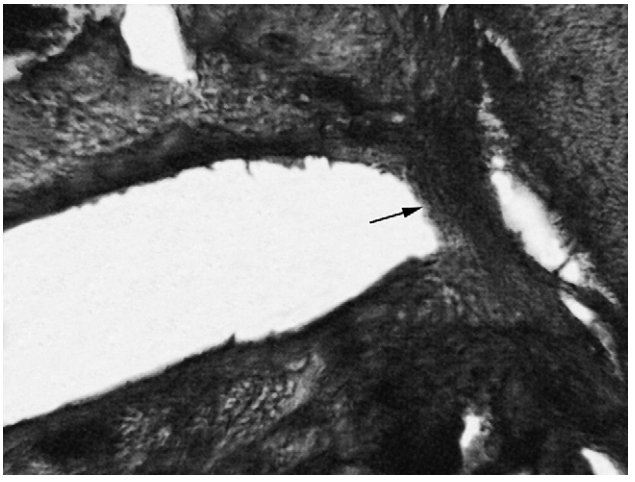

Fig 6. *Medium ADH (original magnification ×20, arrow). Intermediate thickness, not over 10% of a quarter (quadrant) of the joint.*

large (thick, >10% and up to 50% of a quarter region of the joint) (see Figs 5-7 for examples of each type of ADH). The small, medium, and large ADH were carefully examined under light microscopy and the distinguishing characteristics of each were recorded.

In addition, the location of the ADH within the joints from superior to inferior and medial to lateral were recorded by dividing each joint into two sets of quadrants, the two sets of quadrants being positioned perpendicular to each other. The number of ADH were calculated from cephalad to caudad using cephalad, superior middle, inferior middle, and caudad quadrants for each joint. These quadrants were defined using percentages of the total number of sections (ie, 25% increments) in the cephalad to caudad extent of each joint. Similarly, each joint space was also evaluated from medial to lateral, using medial, medial middle, lateral middle, and lateral quadrants, which were determined by visually dividing the joint space into quadrants (the entire medial-lateral extent of the joint space was visualized under low-power magnification).

Interobserver Reliability Study

A study was conducted to determine the reliability with which trained observers could identify Z joint ADH. Two observers, blinded to each other, were assigned 28 Z joints (observers were instructed to assess 4 joints per day for 7 days). The joints were chosen by an independent investigator and intentionally contained all 3 sizes of ADH in order to test the observers' ability to discriminate among the various categories of ADH. Several of the 28 test Z joints were chosen because they had no ADH; however,

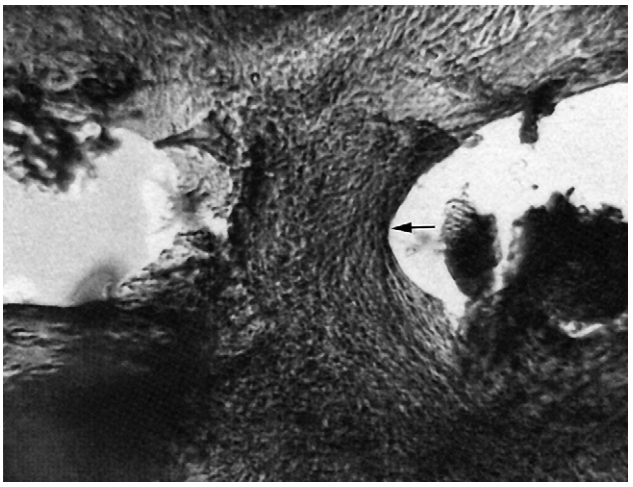

Fig 7. *Large ADH (original magnification ×25, arrow). Thick, up to 50% of a quadrant of the joint.*

most had several ADH of small and/or medium sizes, and 8 joints had large ADH. The observers were given no information regarding the number or type of ADH in the joints and were instructed to record the largest ADH identified within a joint, using a 0-3 scale (0 = no ADH, 1 = small ADH, 2 = medium ADH, and 3 = large ADH). Data were analyzed using the weighted kappa statistic to determine the level of agreement.

Assessment of Z Joints in the Primary Study

A total of 154 Z joints (left and right L4/L5, L5/L6) were randomized and evaluated microscopically by one of the observers (JL) who completed the reliability study. The Z joints were assessed for the presence of ADH bridges within the Z joint space. The same criteria used in the reliability study were used for identification of ADH and for ADH size in the primary study. However, for this part of the study, all ADH within a joint were identified and counted. In addition, the location of the ADH from superior to inferior and from medial to lateral were recorded (see "Assessment of ADH" section).

Statistical Analysis

Average number of ADH per joint (A_{AVG}) was the primary outcome. This value, with SD was calculated in each animal group (4-, 8-, 12-, and 16-week fixation and control groups) for small, medium, and large ADH. Because the data was not Gaussian in distribution, the Kruskal-Wallis (KW) test was used to determine differences among groups and followed by post hoc analysis with Dunn's pairwise comparison for differences between groups. Data were analyzed to determine the difference from 0 at the 5% level of significance for small, medium, and large ADH.

The location of ADH was also assessed as a secondary outcome. Mann-Whitney *U* tests were used to determine if differences existed between the left and right Z joints of control and fixation animals. In addition, descriptive comparisons of the mean small, medium, and large ADH were performed between quadrants to determine regions with the highest frequency from superior to inferior and medial to lateral. Statistical analysis was also performed, using the KW and Dunn's pairwise comparison tests described above, to determine statistically significant differences between fixation and control groups for each quadrant.

Results

Make-up of ADH

Small ADH. Small spinal ADH appeared to consist of a few connective tissue fibers spanning the region between individual bony trabeculae of the 2 sides of a Z joint (Fig 5). The fibers were mostly parallel with one another but were not pristinely aligned, as in a true regular connective tissue. Close to the attachment site of the ADH, a few shorter fibers were sometimes recognized and appeared more haphazardly arranged. No cells were discernable. There was either a sizeable amount of ground substance between fibers or else actual gaps in the tissue, indicating the ADH were probably made out of loose connective tissue.

Medium ADH. Medium-sized ADH had the most variation. All were composed almost entirely of connective tissue fibers, but in some ADH, the fibers had no regular arrangement; in others, there was some regular directional arrangement and, in others, a pristine regular arrangement (Fig 6). There was very scant to little amount of ground substance, and no cells were recognized. Consequently, medium ADH were best classified as dense irregular connective tissue (when the fibers were generally irregular in their arrangement) or dense regular connective tissue (when the fibers were regularly arranged).

Large ADH. The large Z joint ADH also consisted mostly of connective tissue fibers (Fig 7). These fibers were not regularly arranged but had some directionality to their arrangement. Little ground substance was present, and very few, if any, cells were visible. Therefore, large ADH appeared to be composed of dense irregular connective tissue.

Macroscopic View of ADH

The ADH identified in light micrographs of this study (Figs 5-7) were also seen macroscopically. The macroscopic view further demonstrates the nature of the ADH. The supplemental digital content (online at www.jmptonline.org) shows macroscopic views comparing and contrasting the Z joints of an 8-week control and an 8-week fixation animal.

Table 2. *Average number of ADH per joint in control and experimental fixation animals*

Wk of survival (control)/fixation	Small (SD)	Medium (SD)	Large (SD)
C4	4.9 (2.5)	0.3 (0.6)	0.0
F4	4.4 (3.8)	0.4 (0.6)	0.0
C8	5.5 (5.6)	0.6 (0.9)	0.0
F8	4.5 (4.6)	2.0 (2.4)	1.0 (1.8)‡
C12	6.2 (3.4)	0.7 (0.8)	0.0
F12	6.3 (3.6)	1.2 (1.2)	0.6 (0.7)*
C16	4.2 (5.0)	0.5 (1.1)	0.0
F16	9.9 (4.8)†	3.8 (2.9)†	1.8 (3.5)‡

Significant difference between control and fixation at *$P < .05$, †$P < .01$, and ‡$P < .001$.

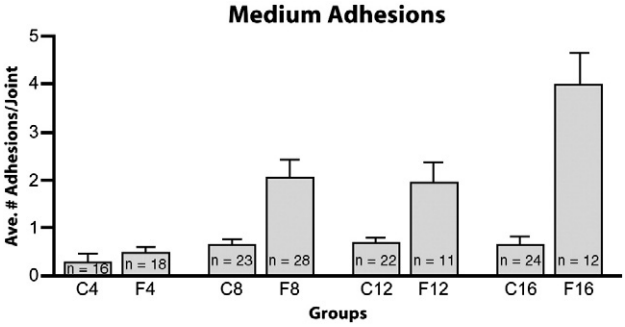

Fig 9. *Graph summarizing the average number of medium ADH per joint (Ave ADH).*

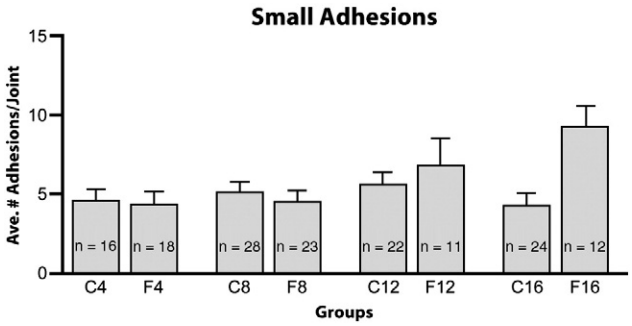

Fig 8. *Graph summarizing the average number of small ADH per joint (Ave ADH). C, control; F, experimental fixation; 4, 8, 12, 16, weeks linked or equivalent control period.*

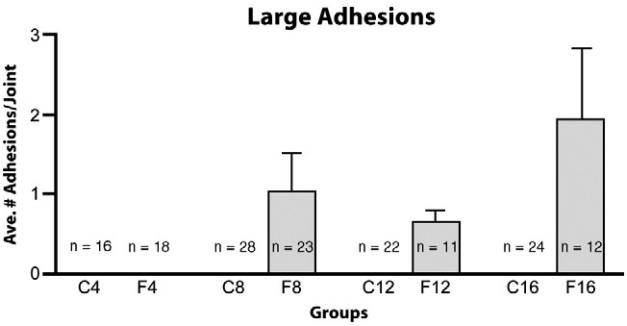

Fig 10. *Graph summarizing the average number of large ADH per joint (Ave ADH).*

Reliability Study

The weighted kappa score for the reliability study of two observers assessing 28 Z joints was 0.86 (SE = 0.18), indicating "almost perfect" agreement[19] between the 2 observers. This high level of agreement indicated that only one observer was needed to successfully complete the primary study.

Average Number of ADH per Z Joint

Control Groups. No significant differences were found in A_{AVG} among the C_{LINK}, C_{SAU}, and C_{SURG} groups for any of the link-equivalent time periods, that is, the 4-week (KW = 1.5, $P = .47$), 8-week (KW = 4.49, $P = .11$), 12-week (KW = 0.51, $P = .77$), or 16-week (KW = 0.1, $P = .95$) controls. Consequently, the values for C_{LINK}, C_{SAU}, and C_{SURG} were pooled into 4- (C4), 8- (C8), 12- (C12), and 16-week (C16) controls for comparisons with each link-equivalent time period of experimental fixation (F) animals (ie, F4, F8, F12, and F16).

Group Comparisons. Table 2 and Figures 8-10 summarize the Z joint ADH assessments for both experimental and control animals.

Small and medium ADH were found in all groups. However, large ADH were found only in the F8 (A_{AVG} = 1.1, SD = 1.9), F12 (A_{AVG} = 0.6, SD = 0.8), and F16 (A_{AVG} = 1.8, SD = 3.5) groups.

There were significant differences among groups for small (KW = 19.921.7, $P < .0103$), medium (KW = 37.032.7) ($P < .0001$), and large (KW = 56.7, $P < .0001$) ADH. The average number of ADH per joint generally increased with the length of hypomobility (Table 2 and Figs 8-10).

Post hoc analysis revealed differences between groups in the following ADH categories: small (C16 vs F16, $P < .01$; and F8 vs F16, $P < .05$), medium (C16 vs F16, $P < .01$; and F4 vs F16, $P < .01$), and large (C8 vs F8, $P < .001$; C12 vs F12, $P < .05$; and C16 vs F16, $P < .001$). (Because there were no large ADH found in the F4 group, this group was significantly different from the F8, F12, and F16 groups for large ADH.) Notice that significant differences were found between 16-week control and 16 week fixation animals for all three sizes of ADH.

Location of ADH within the Z Joints

Adhesions were found in both left and right Z joints, but there were no significant differences between the number of ADH found in left and right joints for

Table 3. *Small ADH: average small ADH/Z joint (SD)*

Quadrant	Lateral	Lateral middle	Medial middle	Medial
All joints[a]	1.84 (2.74)	0.91 (1.71)	0.97 (1.69)	1.71 (2.32)
Control joints[b]	1.38 (2.1)	1.07 (1.93)	1.11 (1.83)	1.6 (2.33)
Fixation joints[c]	2.48 (3.36)*	0.69 (1.32)	0.77 (1.46)	1.86 (2.31)
Quadrant	Cephalad	Superior middle	Inferior middle	Caudad
All joints	1.45 (1.75)	1.44 (2.3)	1.54 (2.17)	1.01 (2.22)
Control joints	1.3 (1.55)	1.61 (2.26)	1.54 (2.12)	0.74 (1.77)
Fixation joints	1.67 (1.99)	1.2 (2.35)	1.53 (2.25)	1.39 (2.7)

[a] Pooled data for all Z joints for both control and fixation animals of the 4-, 8-, 12-, and 16-week survival animals for small, medium, and large ADH.
[b] Pooled data for *control* animals only.
[c] Pooled data for *fixation* animals only.
* Significant at <.05 level for fixation vs control.

Table 4. *Medium ADH: average medium ADH/Z joint (SD)*

Quadrant	Lateral	Lateral middle	Medial middle	Medial
All joints[a]	0.36 (0.85)	0.16 (0.45)	0.16 (0.53)	0.36 (0.89)
Control joints[b]	0.13 (0.43)	0.13 (0.43)	0.1 (0.37)	0.18 (0.55)
Fixation joints[c]	0.69 (1.15)***,†	0.2 (0.48)	0.23 (0.68)	0.61 (1.18)**
Quadrant	Cephalad	Superior Middle	Inferior Middle	Caudad
All joints	0.34 (0.82)	0.23 (0.59)	0.3 (0.71)	0.18 (0.59)
Control joints	0.2 (0.55)	0.12 (0.39)	0.18 (0.51)	0.04 (0.21)
Fixation joints	0.55 (1.07)*	0.38 (0.77)	0.47 (0.89)	0.38 (0.85)**

[a-c] See footnotes of Table 3.
* Significant at <.05 level for fixation vs. control.
** Significant at <.01 level for fixation vs. control.
*** Significant at <.001 level for fixation vs control.
† Significant at <.05 level for fixation vs all.

Table 5. *Large ADH: average large ADH/Z joint (SD)*

Quadrant	Lateral	Lateral middle	Medial middle	Medial
All joints[a]	0.2 (0.88)	0.05 (0.29)	0.01 (0.11)	0.08 (0.36)
Control joints[b]	0 (0)	0 (0)	0 (0)	0 (0)
Fixation joints[c]	0.47 (1.32)***,†	0.11 (0.44)*	0.03 (0.18)	0.2 (0.54)***
Quadrant	Cephalad	Superior middle	Inferior middle	Caudad
All joints	0.07 (0.3)	0.08 (0.46)	0.08 (0.51)	0.1 (0.53)
Control joints	0 (0)	0 (0)	0 (0)	0 (0)
Fixation joints	0.16 (0.44)**	0.2 (0.69)**	0.2 (0.78)**	0.25 (0.8)**

[a-c] See footnotes of Table 3.
* Significant at <.05 level for fixation vs. control.
** Significant at <.01 level for fixation vs. control.
*** Significant at <.001 level for fixation vs control.
† Significant at <.05 level for fixation vs all.

small (P = .25), medium (P = .36), or large (P = .14) ADH.

Tables 3-5 show the distribution of ADH within the Z joints. Figures 11-13 graphically represent the data of the first row of Tables 3 to 5 (pooled data of all Z joints to best identify the location of ADH). Small ADH were more abundant than medium and large ADH (ranges: small ADH = 0.91-1.84, medium = 0.16-0.36, large = 0.01-0.20 ADH/joint). Small ADH were generally evenly distributed within the joint from cephalad to caudad; however, they

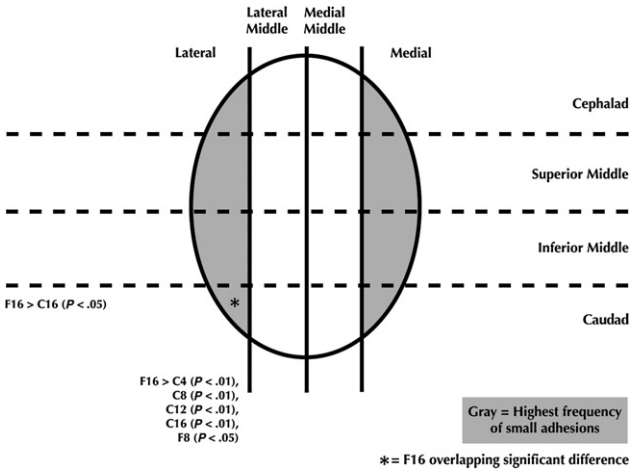

Fig 11. *Small ADH. Figures 11-13 show the two sets of quadrants, used to identify the location of ADH, superimposed on a superior articular facet. The gray shading represents quadrants that have >70% more ADH/joint than another quadrant. The figures also show the results of comparisons among and between specific groups of animals (e.g., F16 vs C16 or F16 vs F8, etc). Symbols are used to identify regions of "overlapping significance." These regions are created when significant differences in ADH exist between control and fixation animals for a particular survival period (4-, 8-, 12-, or 16-week survival animals) in medial-lateral and cephalad-caudad quadrants that overlap one another. The highest frequency of small ADH occurred along the periphery in the medial and lateral quadrants. The lateral and caudad aspect of the Z joints was an area of high concentration of small ADH in the long (16-week) survival period. *Overlapping significance between F16 vs C16 groups. (See Results section of text for further details.)*

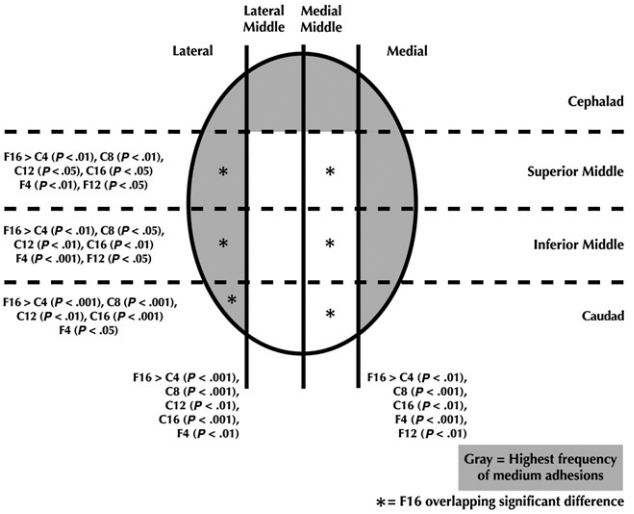

Fig 12. *Medium ADH. Overlapping significant regions were found for the F16 group in the lateral and in the medial middle portions of the joint. *F16 vs C16 groups. (See Results section of text for further details.)*

were more commonly located along the medial and lateral surfaces of the joint (medial and lateral quadrants, Table 3 and Fig 11). Similarly, medium and large ADH were found most frequently in the medial and lateral quadrants (Tables 4 and 5; Figs 12 and 13). Medium ADH were also commonly located in the cephalad quadrant, whereas large ADH were more abundant in the caudal quadrant. Notice that ADH were significantly greater in the lateral and caudal quadrants of the F16 group for small (F16 vs C16: lateral, $P < .05$; caudal, $P < .05$), medium (F16 vs C16: lateral, $P < .001$; caudal, $P < .001$), and large (F16 vs C16: lateral, $P < .001$; caudal, $P < .001$) ADH. In addition, the F16 group had a high concentration of medium ADH in the superior middle (F16 vs C16 $P < .05$), inferior middle (F16 vs C16 $P < .01$), and medial middle (F16 vs C16 $P < .01$) quadrants, indicating that medium ADH were also found toward the center of the Z joints in these long-term-survival animals. There were many fewer large ADH than small or medium ADH. Large ADH were significantly greater in fixation animals in the medial (F8 vs C8, $P < .01$; F12 vs C12, $P < .01$) and lateral middle quadrants (F16 vs C16, $P < .01$).

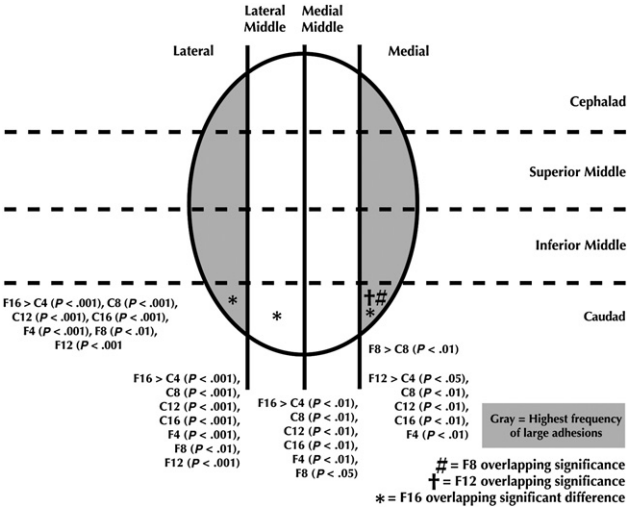

Fig 13. *Large ADH. The medial middle quadrant had so few ADH (0.013 ADH per joint) that the quadrant with the second lowest number of ADH (lateral middle, 0.045 ADH per joint) was used for calculations of 70% more ADH than the quadrant with the lowest number of ADH. Overlapping significant regions were found in the inferior aspect of the joint, specifically in the caudal, lateral middle, and lateral quadrants. #F8 vs C8; †F12 vs C12; *F16 vs C16. (See Results section of text for further details.)*

Discussion

Summary of Findings

We found that the assessment of ADH was accomplished reliably and the light microscopic structure of the Z joint ADH could be clearly visualized

and described (see subsection Nature and Source of ADH).

The ADH were approximately equal in numbers in the left and right Z joints and were most commonly found in the periphery of the Z joints, both along the medial and lateral (ie, the medial and lateral quadrants) and to a lesser extent the superior and inferior aspects of the joints (cephalad quadrant, medium ADH; caudad quadrant, large ADH). The number of ADH was generally related to the length of time a joint underwent induced hypomobility (see subsections Location of ADH and Time-Dependent Nature of ADH Development).

Several specific issues of this study can benefit from further discussion. These issues include: the effect of the surgery on the development of ADH, the nature and source of the ADH, the time-dependent nature of ADH development, the study's limitations, and the potential clinical relevance and directions of future research. These issues are addressed in the following sections.

Effect of Surgery and the SAUs on the Development of ADH

The effect of the surgeries and/or the SAUs used to induce hypomobility in this study was of high importance in assessing the relevance of the research. If the surgeries or implanted SAUs contributed to the creation of intra-articular ADH, then the effects of hypomobility alone on ADH development would be difficult, or impossible, to evaluate. For this reason more control animals were used in this study than fixation animals (24 controls vs 17 fixation animals). The controls animals either had: no surgery at all (C_{SURG} group); surgery with the spinous processes prepared for implantation of the SAUs, but the SAUs were not implanted (C_{SAU} group); or they had the surgery and the SAUs implanted, but the SAUs were never linked together to create hypomobility (C_{LINK} group). The C_{LINK} animals were the most closely related to the experimental fixation animals, the only difference being that the linking device was never put in place. Th C_{LINK} animals showed no difference from the other two types of controls, regardless of duration of survival (4, 8, 12, or 16 weeks) or type of ADH (small, medium, or large), yet the control animals were significantly different from the fixation animals in many categories. The same held true for the (C_{SAU}) animals. Recall that these animals had surgery but the SAUs were not implanted. This group also showed no difference from the other control animals, including the C_{SURG} Group that had no surgery at all. These findings indicate that neither the surgical procedures themselves nor the implantation of SAUs had an effect on ADH development. Consequently, we are confident that the differences found between the experimental fixation animals and the control animals represent the effects of hypomobility and not the effects of the surgical procedures or the implantation devices (SAUs).

Nature and Source of ADH

Small ADH were composed primarily of loose connective tissue; medium ADH were most variable and composed either of dense irregular connective tissue (if the connective tissue fibers were irregularly arranged) or dense regular connective tissue (if the fibers were regularly arranged) and large ADH were composed primarily of dense irregular connective tissue.

Hase[14] addressed the issue of the source of intra-articular ADH in his study of the TMJ. He was convinced that the ADH were the result of a "deposition of fibrinoid material" secondary to "degeneration of type-A cells of the synovial membrane."[14] Although the TMJs are modified fibrous joints and the Z joints are planar synovial joints, the same mechanism is reasonable for the Z joints, because the Z joints have an ample synovium found not only along the inner joint capsule but also surrounding the Z joint synovial folds.[20,21] Further research is required to verify the source of Z joint intra-articular ADH. Additional work characterizing the ultrastructural and biochemical composition of the ADH is also warranted.

Location of ADH

The ADH were most abundant along the periphery of the Z joints (primarily the medial and lateral quadrants, but also to a lesser extent the cephalad and caudad quadrants). Even though large and medium ADH were significantly more abundant along the periphery, the medium ADH in the F16 group also extended into the more central quadrants of the Z joints. The most likely explanation of this is that the Z joint synovial folds are located along the periphery of the joints, and the broad capsular attachment sites of these folds lie along the medial and lateral aspects of the joints. Recall that Hase[14] believed the ADH he found in the TMJ were caused by the breakdown of synoviocytes, which would leave small joint inclusions around which collagen-based ADH could develop. The synoviocyte-rich Z joint synovial folds provide an abundant source of synoviocytes whose natural (and relatively rapid) turnover may present a plentiful source of small joint inclusions around which Z joint ADH could develop. Again, further ultrastructural and biochemical analysis focusing on the formation of the ADH is needed to better understand why the ADH are more prominent along the periphery of hypomobile Z joints.

Time-Dependent Nature of ADH Development

The number of ADH was generally directly related to the length of time a joint underwent induced hypomobility. Small ADH were common in the Z joints of control and fixation rats; however, medium and large ADH seemed closely related to the duration of hypomobility (spinal fixation) with significant differences found between 16 week control and fixation animals for all

sizes of ADH. Although slightly more medium and large ADH were found in 8-week fixation animals compared to 12-week fixation animals, the differences were not significant.

The results indicate that although small ADH are relatively common, medium and large ADH develop with increasing durations of hypomobility, suggesting that small ADH may develop into medium or eventually large ADH with continued hypomobility. One possible interpretation of these findings is that small ADH act as "seed structures" that are in dynamic equilibrium, that is, building up and breaking down, depending on the absence or presence of joint motion. Consequently, ADH may build up and, possibly, become irreversible with chronic hypomobility of a joint. This might explain the finding that in 16-week fixation animals (the maximum survival length in this study), medium ADH were found not only in the periphery of the joint, but also more centrally as well, an indication that small ADH had enlarged into medium ADH. One might anticipate that with longer periods of hypomobility, the medium ADH would further develop into large ADH. A study similar to that reported here, but extending the fixation period to 20 and 24 weeks would help to confirm this hypothesis. One might also speculate that the rapid Z joint motion (gapping) induced by spinal manipulation may act to modulate such a dynamic ADH development system. This is conjecture and more research is needed to clarify the role spinal manipulation may play in modulating ADH development.

Limitations

A relatively small number of animals were available for some of the control subgroups and only three animals were available in the 12- and 16-week fixation groups. Consequently, some caution should be used when interpreting the data. However, the total number of joints was relatively high and the changes were significant in several areas (eg, 16-week control vs 16-week fixation animals for small, medium, and large ADH), indicating that the findings reported here are due to real differences.

A large number of statistical tests were performed to analyze the secondary outcome of ADH location within the joint. Although we are confident the results provide useful information regarding the location of ADH within this study, further research with larger numbers of animals is needed to reach the statistical power necessary to assess ADH location as a primary outcome.

In addition, this study did not assess the ultrastructure or biochemical make up of the ADH. Such analyses in future investigations could provide useful information regarding the origins and the nature of the ADH, and the possible mechanisms by which ADH development in the Z joints could be slowed and/or their breakdown enhanced by therapeutic interventions.

Clinical Relevance and Future Research

These findings are consistent with the hypothesis that joint hypomobility leads to increased ADH development (Fig 1). The results reported here are also consistent with previously reported findings that osteophyte formation and degenerative changes of the articular facets increase with induced hypomobility.[3] Additional research is needed to determine the clinical significance of both ADH size and the effects of spinal manipulation on Z joint ADH. Experiments assessing the effects of standardized high-velocity, low-amplitude thrusts and low-velocity, variable-amplitude mobilizations on degenerative changes of the Z joints in this animal model are currently underway.

Conclusions

Experimentally induced segmental hypomobility (fixation) of the lumbar Z joints resulted in time dependent intra-articular ADH formation. The ADH were found in approximately equal numbers in the left and right Z joints and were most prevalent in the peripheral regions of the joint from medial to lateral and cephalad to caudal. These findings are consistent with the hypothesis that hypomobility results in time-dependent degenerative changes and ADH development of the Z joints.

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.jmpt.2010.08.002.

Practical Applications

- Adhesions were found in approximately equal numbers in left and right zygapophyseal joints and were more common in the peripheral regions of the joints (ie, medial and lateral aspects and to a lesser extent superior/cephalad and inferior/caudad).
- Small sized ADH were commonly found in the zygapophyseal joints of rats (even without induced intervertebral hypomobility, ie, spinal fixation) and were composed of loose irregular connective tissue.
- Medium and large sized ADH seemed closely related to the duration of induced intervertebral hypomobility and were composed primarily of dense irregular connective tissue.
- Significant differences were found between 16-week control and induced hypomobility animals (longest duration of induced intervertebral hypomobility) for all sizes of ADH.
- These findings may have implications in the mechanism of action of spinal manipulation (ie, spinal manipulation theoretically may affect the time profile of ADH development with intervertebral hypomobility).

Acknowledgment

We thank Barclay Bakkum, DC, PhD for assistance in assessing the morphological make-up of the ADH.

Funding Sources and Potential Conflicts of Interest

Funding for this study was provided by NCMIC, administered by the Foundation for Chiropractic Education and Research (FCER, grant number 97-10-18). No conflicts of interest were reported for this study.

References

1. Janse J. In: Hildebrandt RW, editor. Principles and practice of chiropractic: an anthology. Wheaton: Kjellberg & Sons; 1976. p. 326.
2. Triano JJ. Interaction of spinal biomechanics and physiology. In: Haldeman S, editor. Principles and practice of chiropractic. 2nd ed. East Norwalk, Conn: Appleton & Lange; 1992. p. 225-57.
3. Cramer GD, Fournier JT, Henderson CN, Wolcott CC. Degenerative changes following spinal fixation in a small animal model. J Manipulative Physiol Ther 2004;27:141-54.
4. Cramer GD, Tuck NR, Knudsen JT, Fonda SD, Schliesser JS, Fournier JT, et al. Effects of side-posture positioning and side-posture adjusting on the lumbar zygapophysial joints as evaluated by magnetic resonance imaging: a before and after study with randomization. J Manipulative Physiol Ther 2000; 23:380-94.
5. Cramer GD, Gregerson DM, Knudsen JT, Hubbard BB, Ustas LM, Cantu JA. The effects of side-posture positioning and spinal adjusting on the lumbar Z joints: a randomized controlled trial with sixty-four subjects. Spine 2002;27: 2459-66.
6. Cramer GD, Fournier JT, Henderson C. Zygapophysial joint changes following spinal fixation. International Conference on Spinal Manipulation; 2000 September 21-23; Minneapolis. Brookline: Foundation for Chiropractic Education and Research; 2000. p. 85-7.
7. Pickar JG, Wheeler JD. Response of muscle proprioceptors to spinal manipulative-like loads in the anesthetized cat. J Manipulative and Physiol Ther 2001;24:2-11.
8. Ianuzzi A, Khalsa PS. Comparison of human lumbar facet joint capsule strains during simulated high-velocity, low-amplitude spinal manipulation versus physiological motions. Spine J 2005;5:277-90.
9. Bakkum BW, Henderson CN, Hong SP, Cramer GD. Preliminary morphological evidence that vertebral hypomobility induces synaptic plasticity in the spinal cord. J Manipulative Physiol Ther 2007;30:336-42.
10. Cramer GD, Cantu JA, Pocius JD, Balester F, Simpson DRJ, Horner TB, et al. Reliability of zygapophysial (Z) joint measurements taken from MRIs of subjects with acute low back pain. FASEB J 2009;23:649.
11. Enneking WF, Horowitz M. The intra-articular effects of immobilization on the human knee. J Bone Joint Surg Am 1972;54:973-85.
12. Trudel G, Seki M, Uhthoff HK. Synovial adhesions are more important than pannus proliferation in the pathogenesis of knee joint contracture after immobilization: an experimental investigation in the rat. J Rheumatol 2000;27:351-7.
13. Hannafin JA, Chiaia TA. Adhesive capsulitis. A treatment approach. Clin Orthop Relat Res 2000:95-109.
14. Hase M. Adhesions in the temporomandibular joint: formation and significance. Aust Dent J 2002;47:163-9.
15. Laroche M, Ighilahriz O, Moulinier L, Constantin A, Cantagrel A, Mazieres B. Adhesive capsulitis of the shoulder: an open study of 40 cases treated by joint distention during arthrography followed by an intraarticular corticosteroid injection and immediate physical therapy. Rev Rhum Engl Ed 1998;65:313-9.
16. Henderson CN, Cramer GD, Zhang Q, DeVocht JW, Fournier JT. Introducing the external link model for studying spine fixation and misalignment: part 1—need, rationale, and applications. J Manipulative Physiol Ther 2007;30:239-45.
17. Henderson CN, Cramer GD, Zhang Q, DeVocht JW, Fournier JT. Introducing the external link model for studying spine fixation and misalignment: part 2, biomechanical features. J Manipulative Physiol Ther 2007;30:279-94.
18. Henderson CN, Cramer GD, Zhang Q, DeVocht JW, Sozio RS, Fournier JT. Introducing the external link model for studying spine fixation and misalignment: current procedures, costs, and failure rates. J Manipulative Physiol Ther 2009;32: 294-302.
19. Landis J, Koch G. The measurement of observer agreement for categorical data. Biometrics 1977;33:174.
20. Giles LG. Human zygapophyseal joint inferior recess synovial folds: A light microscopic examination. Anat Rec 1988; 220:117-24.
21. Giles LG. The pathophysiology of the zygapophysial joints. In: Haldeman S, editor. Principles and practice of chiropractic. 2nd ed. East Norwalk, Conn: Appleton & Lange; 1992. p. 197-210.