Role of vitamin D₃ in Treatment of Lumbar Disc Herniation—Pain and Sensory Aspects: Study Protocol for a Randomized Controlled Trial

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Abstract

**Background:** Vitamin D receptors have been identified in the spinal cord, nerve roots, dorsal root ganglia and glial cells, and its genetic polymorphism association with the development of lumbar disc degeneration and herniation has been documented. Metabolic effects of active vitamin D metabolites in the nucleus pulposus and annulus fibrosus cells have been studied. Lumbar disc herniation is a process that involves immune and inflammatory cells and processes that are targets for immune regulatory actions of vitamin D as a neurosteroid hormone. In addition to vitamin D’s immune modulatory properties, its receptors have been identified in skeletal muscles. It also affects sensory neurons to modulate pain. In this study, we aim to study the role of vitamin D₃ in discogenic pain and related sensory deficits. Additionally, we will address how post-treatment 25-hydroxy vitamin D₃ level influences pain and sensory deficits severity. The cut-off value for serum 25-hydroxy vitamin D₃ that would be efficacious in improving pain and sensory deficits in lumbar disc herniation will also be studied.

**Methods/Design:** We will conduct a randomized, placebo-controlled, double-blind clinical trial. Our study population will include 380 cases with one-level and unilateral lumbar disc herniation with duration of discogenic pain less than 8 weeks. Individuals who do not have any contraindications, will be divided into three groups based on serum 25-hydroxy vitamin D₃ level, and each group will be randomized to receive either a single-dose 300,000-IU intramuscular injection of vitamin D₃ or placebo. All patients will be under conservative treatment. Pre-treatment and post-treatment assessments will be performed with the McGill Pain Questionnaire and a visual analogue scale. For the 15-day duration of this study, questionnaires will be filled out during telephone interviews every 3 days (a total of five times). The initial and final interviews will be scheduled at our clinic. After 15 days, serum 25-hydroxy vitamin D₃ levels will be measured for those who have received vitamin D₃ (190 individuals).

**Trial registration:** Iranian Registry for Clinical Trials ID: IRCT2014050317534N1 (trial registration: 5 June 2014)

**Keywords:** Inflammation, Lumbar disc herniation, Pain, Sensory, Vitamin D₃

Background

Medical treatment is the first step in therapy for lumbar disc herniation (LDH), except for patients who require immediate surgical decompression. Drugs that are utilized in treatment of LDH pain and sensory deficits include muscle relaxants [1-3], analgesics [1,2,4-9], corticosteroids [1,2,10], antidepressants [4,8,11,12] and antiepileptics [4,8,11-17].

Vitamin D is a secosteroid hormone that has many skeletal and nonskeletal functions [18-94]. In addition to its classic action on bone metabolism and osteoporosis [18,19], its links and roles in relation to other diseases have been addressed in the literature (diabetes mellitus [18,20-23], hypertension [24,25], cardiovascular diseases [18,26-29], multiple sclerosis [30-35], neurodegenerative diseases [36-39], neuropsychiatric diseases [39-44], inflammatory bowel disease [33,45-49], dermatologic diseases [50-58], rheumatoid arthritis [47,53,59-61], systemic lupus erythematosus [60,62-67], transplant rejection [68-70], cancer [18,52,68,71-73], postherpetic neuralgia [74], corneal neuralgia [75], respiratory diseases [76-79], pregnancy...
complications [80-82], human reproductive issues [83-85], migraine headache [86], chronic low back pain [87,88], chronic painful conditions and fibromyalgia [89,90], and diabetic neuropathy [91-93]). Studies that have shed light on areas that have given us the scientific underpinning for our present proposal are described below.

1. Vitamin D has been called a neurosteroid hormone [39,74,94-109], given its protective role against neurotoxicity and detoxification pathways [74,94,96-108] and also its receptors in different parts of the central nervous system [36,94-96,106-114].

2. Vitamin D receptors are present in the spinal cord, nerve roots, dorsal root ganglia and glial cells [94,96,97,113,115-118].

3. Vitamin D receptor gene polymorphism has a role in the development of lumbar disc degeneration and herniation [119-123].

4. Discs are composed largely of avascular tissue with a great sensitivity to its nutritional supply and excretion of waste products, and the balance between these two processes is an important factor that could lead to disc degeneration [124-127]. The effects of active vitamin D metabolites in nucleus pulposus and annulus fibrosus cells have been studied [128]. Vitamin D inhibits and decreases production of monocyte chemoattractant protein 1, thrombopoietin, vascular endothelial growth factor and angiogenin by human annulus cells in vitro [129]. As mentioned above, vitamin D affects detoxification pathways which are of importance in disc cell nutritional balance.

5. Vitamin D possesses immune regulatory properties which can downregulate proinflammatory cytokines and upregulate anti-inflammatory cytokines [22,32,36,46-48,58,67,70,74,78,90,94,96,130-146].

6. Vitamin D has properties that defend against cell injury caused via free radicals, reactive oxygen species, glutathione and glutamate [74,94,96-108,136,147-149].

7. Vitamin D has a role in pain by downregulating inflammatory cytokines that produce pain (a) directly, (b) by stimulating release of pain mediators, (c) by upregulating anti-inflammatory cytokines to help the body combat inflammation, (d) by its role in eliminating toxic metabolites or (e) by increasing the antioxidant pool. It also affects sensory neurons to modulate pain [114], influences neuron excitability [96] and acts at the level of substantia gelatinosa and spinal ganglion in the process of sensory perception [118]. In addition, its status affects pain sensitivity and opiate activity [150].

8. The role of the vitamin D receptor in skeletal muscles [151-155] and its effects on muscle strength and function have been identified [156-159].

In addition to the information described above, many studies about changes that occur in LDH have been done, as outlined below.

1. The contribution of inflammatory cytokines in the pathogenesis of LDH has been widely addressed in the literature. The herniated nucleus pulposus, either with immunogenic properties itself or by inducing an immunologic response in the nerve roots, dorsal root ganglia and surrounding muscles, is the starting point for the cascade of inflammation initiated through immune cell activation and infiltration and cytokine release [160-184].

2. Neuropathic pain involves the activation of neurons, glial cells and the immune system [185,186]. Dorsal root ganglia and dorsal roots play important roles in LDH, not only by the effect of released inflammatory cytokines but also by actively amplifying inflammation by producing proinflammatory cytokines and pain mediators that affect pain perception and nociception. Among these substances is brain-derived neurotrophic factor. Its receptor has been identified in intervertebral discs, with its expression being increased during inflammatory conditions such as LDH and its neuroimmunomodulatory role in the dorsal root of the spinal cord [185,187-204]. The other factor is glial cell-derived neurotrophic factor (GDNF). It has been shown that GDNF reduces neuropathic pain states [188,190,205-208]. Interestingly, vitamin D affects neuropathic pain by directly suppressing inducible nitric oxide that is expressed in glial cells [96,136] or by affecting other substances, such as reactive oxygen species or glutamate. Given the immunomodulatory action of vitamin D, it is possible that it could downregulate inflammatory chemokines released by glial cells [96,185-189,209-215]. It has been suggested that vitamin D attenuates ischemia-induced brain injury that is thought to be mediated through upregulation of GDNF, in addition to its role in nitric oxide (NO) suppression [216]. The results of other studies support the hypothesis that GDNF is upregulated by vitamin D [90,94,96,190,217]. Interleukin 6 (IL-6) and tumor necrosis factor α produced by glial cells were shown to be downregulated by vitamin D [94,96,136], as were glial cell release of NO [188,218,219], prostaglandin [188], IL-1 and IL-6 [218], which, as described below, could be suppressed by vitamin D administration. Glial cells
have glutamate receptors that are important in the process of nociception [220-224]. Therefore, vitamin D, through its immunoregulatory properties, affects another important cell population that is inflamed in disc herniation, either through suppressing neurotoxic agents or by its action on neurotrophins.

Some specific inflammatory cytokines and pain mediators that are involved in LDH and vitamin D immunomodulatory effects with regard to these specific substances are described in Table 1.

3. Detailed study of inflammatory cytokines and subsequent pain mediators released in LDH has shown that there is a shift toward type 1 T-helper cell activity [164,177,181,182,228].

4. Vitamin D decreases the number and function of type 1 T-helper cells [47,48,67,90,253].

5. Muscle changes associated with low back pain have been studied [254-258]. Studies have shown how muscles are affected by LDH [259-266]. Atrophy of type II muscle fibers [259-261,263] or atrophy of both types I and II muscle fibers [260] and adipocyte enlargement are examples of how muscles are targeted by LDH [264]. Vitamin D deficiency–associated histochemical changes in muscles somehow resemble those seen in LDH-affected muscles with atrophy of type II muscle fibers [267-271] and enlarged interfibrillar spaces and fat infiltration and glycogen granules [271-274]. Another interesting aspect of vitamin D deficiency is how it promotes skeletal muscle hypersensitivity and sensory hyperinnervation [275]. Vitamin D supplementation was shown to increase the diameter of type II muscle fibers [181,276]. It also influences transdifferentiation of muscle cells to adipose cells [277]. With regard to the presence of vitamin D receptor in skeletal muscles [151-155], its effect on muscle growth and proliferation [278-282] and the changes seen in muscles after LDH, we propose that vitamin D supplementation also influences muscle changes in this condition.

### Methods/Design

**Design of the study**

We will conduct a randomized, placebo-controlled, double-blind clinical trial.

**Statement of ethical approval**

This study was approved by the local research ethics committee of Shiraz University of Medical Sciences, Shiraz, Iran (CT-P-92-6632).

**Informed consent**

Informed consent will be obtained from all participants.

**Setting**

We will recruit patients who have appointments at the neurosurgery outpatient departments of the university-affiliated hospitals of Shiraz, Iran.

**Participants**

We will recruit 380 patients with LDH proven by physical examination and confirmed by magnetic resonance imaging.

**Intervention**

Patients in the intervention arm will receive single-dose intramuscular injections of 300,000 IU of vitamin D₃ (1 ml). Individuals will be informed about the nature of this study.

**Inclusion criteria**

The following are the inclusion criteria:

1. Single-level LDH
2. No coexistent or preexisting spine pathology (for example, spondylolysis, spondylolisthesis, infection, tumors, fracture)
3. Discogenic pain duration less than 8 weeks from onset to physician’s evaluation

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**Table 1 Vitamin D effects on substances involved in lumbar disc herniation**

| Vitamin D actions [references] | LDH [references] |
|-------------------------------|-----------------|
| IFN-γ: D [46,65,72,88,94,144] | E [160,171,179,180] |
| IL-1: D [46,65,72] | E [173,225-227] |
| IL-2: D [46,65,72,88,94,139] | E [179] |
| IL-4: D [46] | E [179] |
| IL-5: D [67] | E [179] |
| IL-6: D [32,46,72,94,136,141] | E [165,176,181,228-230] |
| IL-8 | E [164,225,231] |
| IL-10: U [32,46,72,94,136,141] | E [165,176,181,228-230] |
| IL-12: D [22,32,67,139,140] | E [181,182] |
| IL-17: D [47,90] | E [181] |
| MCP: I [129] | E [164,175] |
| MMP: I [232-240] | E [176,190,228,241-243] |
| ROS: I [98,101,102,106,238] | E [244] |
| NO: I [245] | E [126,148,176,190,228,246-249] |
| Glutamate: I [101,147] | E [220,221] |
| Glutathione: I [96,106,148] | E [176,190,228,243,251,252] |

D, Downregulation; E, Expression; I, Inhibition; IFN-γ, Interferon γ; IL, Interleukin; LDH, Lumbar disc herniation; MCP, Monocyte chemoattractant protein; MMP, Matrix metalloproteinase; NO, Nitric oxide; PG, Prostaglandin; ROS, Reactive oxygen species; U, Upregulation.
4. Compliance with the study protocol
5. Normal laboratory studies that do not contraindicate vitamin D₃ injection

**Exclusion criteria**
The following are the exclusion criteria:

1. Daily supplementation of more than 800 IU of vitamin D₃
2. Serum calcium level above 10.5 mg/dl
3. Hypercalciuria (spot urine calcium/creatinine ratio above 0.4)
4. Lymphoma, sarcoidosis, tuberculosis (TB), hyperparathyroidism, celiac disease or malabsorption syndromes
5. History of kidney stones
6. History of inflammatory back pain
7. Impaired renal function tests (glomerular filtration rate less than 30 ml/min/1.73 m²)
8. Impaired hepatic function tests
9. Abnormal serum phosphorus, alkaline phosphatase and parathyroid hormone values
10. Fasting blood sugar above 126 mg/dl
11. Previous spine surgery
12. History of trauma
13. Taking anticonvulsant, anti-TB medications or vitamin D₃ analogues
14. Cauda equine syndrome that requires emergency surgical decompression

**Laboratory Assessments**
The following laboratory workups will be performed for all included participants: serum 25-hydroxy vitamin D₃ level, serum calcium, serum phosphorus, alkaline phosphatase, parathyroid hormone, liver function tests (bilirubin (direct and total), alanine transaminase, aspartate transaminase, total protein, total albumin), blood urea nitrogen, creatinine, spot urine for calcium and fasting blood sugar. Clinic-based pre-intervention interviews and physical examinations will include the following:

1. McGill Pain Questionnaire: The McGill Pain Questionnaire is used to evaluate different pain qualities and intensities. This questionnaire consists of four major descriptors: sensory, affective, evaluative and miscellaneous. Each descriptor has its own rank value. The sum of these rank values is the pain rating index. Present pain intensity is measured on scale from 0 to 5 [281].
2. Visual analogue scale (VAS) to evaluate low back pain and radicular pain: A VAS is a pain measurement scale that incorporates numbers and faces to depict the severity of pain. It is usually a 100-mm line. Its ends show the pain extremes [229,282].
3. A physical examination to detect any sensory deficits.

**Randomization**
Patients will be categorized on the basis of their serum 25-hydroxy vitamin D₃ levels into three groups:

- **Group 1**: Optimum 25-hydroxy vitamin D₃ level (32 to 50 ng/ml)
- **Group 2**: Deficient 25-hydroxy vitamin D₃ level (less than 10 ng/ml)
- **Group 3**: Insufficient 25-hydroxy vitamin D₃ level (less than 32 ng/ml)

Each of the groups will be randomized, based on randomly computer-generated numbers, into two groups to receive intramuscular injection of either 300,000 IU of vitamin D₃ (1 ml) or distilled water (1 ml). All patients will be prescribed daily 15 mg Meloxicam capsules. Our study population will be warned verbally and in writing about the potential for severe adverse side effects of vitamin D₃ (nausea, vomiting, abdominal pain, metallic taste, breathing difficulties). They will have access to emergency department care should side effects occur.

The study will last 15 days. After vitamin D₃ injection, patients will be contacted by telephone every 3 days to assess the sensory and pain effects of vitamin D₃ with the McGill Pain Questionnaire and the VAS (a total of five times). Participants will be provided with the VAS so that they can look at the scale and report their pain severity during the telephone interviews.

The following are the final post-treatment evaluations that will be carried out at the clinic:

1. McGill Pain Questionnaire
2. VAS (for low back pain and radicular pain)
3. Physical examination to detect any sensory deficits

Post-treatment 25-hydroxy vitamin D₃ levels (after 15 days) will be measured for those participants who have received vitamin D₃ (N = 190).

**Statistical analysis**
Data will be assessed by analysis of variance and paired tests.

**Discussion**
On the basis of the inflammatory nature of disc herniation and the immunomodulatory effects of vitamin D, as well as the existence of vitamin D receptors in various parts of areas that are affected in the process of disc herniation, we propose a novel role for vitamin D in the treatment of discogenic pain and sensory deficits related
to this pathology. We hypothesized that vitamin D₃ plays a role in reducing the severity of discogenic pain and that vitamin D₃ can improve discogenic-related sensory deficits.

The following are our general objectives in this trial:

1. Effect of vitamin D₃ on discogenic pain
2. Effect of posttreatment 25-hydroxy vitamin D₃ level on pain and sensory deficit severity
3. Determining a cut-off level of 25-hydroxy vitamin D₃ that is efficient in improving pain and sensory deficits

The following are our applicative objectives:

1. Proposing vitamin D₃ as part of medical treatment for LDH
2. Improving LDH patients’ quality of life
3. Decreasing the economic and health burden of LDH

Our ultimate goal in this study is to introduce a new treatment strategy for the treatment of discogenic pain.

**Trial status**
The study protocol has been approved by the Vice-Chancellor for Research of Shiraz University for Medical Sciences. Recruitment has not been initiated.

**Abbreviations**
ALT: Alanine transaminase; AST: Aspartate transaminase; D: Downregulation; E: Expression; I: Inhibition; IFN-γ: Interferon γ; LDH: Lumbar disc herniation; MCP: Monocyte chemotactant protein; MMP: Matrix metalloproteinase; NO: Nitric oxide; PG: Prostaglandin; ROS: Reactive oxygen species; U: Upregulation.

**Competing interests**
The authors declare that they have no competing interests.

**Authors’ contributions**
MS contributed to the acquisition and study of background data, proposed the novel role for vitamin D in the treatment of lumbar disc herniation, suggested the design of the study and how it will be carried out, and helped develop the inclusion and exclusion criteria and laboratory studies. AH participated in the design of the study and how it will be carried out and developing the inclusion and exclusion criteria. Both authors read and approved the final manuscript.

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**References**
1. Smeal WL, Tyburski M, Allewa J: Discogenic/radicular pain. Dir Mon 2004, 50:636–669.
2. Valat J-P, Genievry S, Marty M, Rozenberg S, Koes B: Sciatica. Best Pract Res Clin Rheumatol 2010, 24:241–252.
3. Legrand E, Boudard B, Audran M, Fournier D, Valat JP: Sciatica from disk herniation: Medical treatment or surgery? Joint Bone Spine 2007, 74:530–535.
4. Stafford MA, Peng P, Hill DA: Sciatica: a review of history, epidemiology, pathogenesis, and the role of epidural steroid injection in management. Br J Anaesth 2007, 99:461–473.
5. Van Boxem K, Cheng J, Patijn J, Van Kleef M, Lataster A, Melikshah N, Van Zundert J: Lumbosacral radicular pain, Pain Pract 2010, 10:339–358.
6. Koes BW, Van Tulder MW, Peul WC: Diagnosis and treatment of sciatica. BMJ 2007, 334:1313–1317.
7. Tarulli AW, Raynor EM: Lumbosacral radiculopathy. Neurol Clin 2007, 25:387–405.
8. Pinto RZ, Maher CG, Ferreira MS, Ferreira PH, Hancock M, Oliveira VC, Mclachlan AJ, Koes B: Drugs for relief of pain in patients with sciatica: systematic review and meta-analysis. BMJ 2012, 344.
9. Ito T, Takano Y, Yuasa N: Types of lumbar herniated disc and clinical course. Spine 2001, 26:648–651.
10. Green LN: Dexamethasone in the management of symptoms due to herniated lumbar disc. J Neurol Neurosurg Psychiatry 1975, 38:1211–1217.
11. Chou R: Treating sciatica in the face of poor evidence. BMJ-British Med J 2012, 344:12.
12. Levin KH: Nonsurgical interventions for spine pain. Neurol Clin 2007, 25:495–505.
13. Kasimcan O, Kaptan H: Efficacy of gabapentin for radiculopathy caused by lumbar spinal stenosis and lumbar disk hernia. Neurol Med Chir 2010, 50:1070–1073.
14. Eisenberg E, Damunni G, Hoffer E, Baum Y, Kivoy N: Lamotrigine for intractable sciatica: correlation between dose, plasma concentration and analgesia. Eur J Pain 2003, 7:485–491.
15. Zaremba PD, Bielak M, Blaszczak B, Cioczek P, Czuczwar SA: Non-epilepsy uses of antiepileptic drugs. Pharmacol Rep 2006, 58:1–12.
16. Saldana MT, Navarro A, Pérez C, Masamón X, Rejas J: Patient-reported-outcomes in subjects with painful lumbar or cervical radiculopathy treated with pregabalin: evidence from medical practice in primary care settings. Rheumatol Int 2010, 30:1005–1015.
17. Leo RJ: Treatment considerations in neuropathic pain. Curr Treat Options Neurol 2006, 8:389–400.
18. Holick MF: Vitamin D: importance in the prevention of cancers, type 1 diabetes, heart disease, and osteoporosis. Am J Clin Nutr 2004, 79:362–371.
19. Holick MF: The vitamin D epidemic and its health consequences. J Nutr 2005, 135:2739S–2748S.
20. Pittas AG, Lau J, Hu FB, Dawson-Hughes B: The role of vitamin D and calcium in type 2 diabetes. A systematic review and meta-analysis. J Endocrinol Metab 2007, 92:2017–2029.
21. Mathieu C, Badenhoop K: Vitamin D and type 1 diabetes mellitus: state of the art. Trends Endocrinol Metab 2005, 16:261–266.
22. Aronson Y, Antal M, Shoenfeld Y: Vitamin D and autoimmunity: new aetiological and therapeutic considerations. Ann Rheum Dis 2007, 66:1137–1142.
23. Kamen DL, Tangpricha V: Vitamin D and molecular actions on the immune system: modulation of innate and autoimmunity. J Mol Med (Berl) 2010, 88:441–450.
24. Forman JP, Giovannucci E, Holmes MD, Bischoff-Ferrari HA, Twarogger SS, Willett WC, Cuthran GC: Plasma 25-hydroxyvitamin D levels and risk of incident hypertension. Hypertension 2007, 49:1063–1069.
25. Li YC, Qiao G, Uskokovic M, Xiang W, Zheng W, Kong J: Vitamin D: a negative endocrine regulator of the renin–angiotensin system and blood pressure. J Steroid Biochem Mol Biol 2004, 89:387–392.
26. Dey Y, Green SA, Hoshen M, Feldmane I, Baldwin RD, Feldman BS: Vitamin D levels for preventing acute coronary syndrome and mortality: evidence of a nonlinear association. J Clin Endocrinol Metab 2013, 98:2160–2167.
27. Nemerovski CW, Dorsch MP, Simpson RU, Bone HG, Aaronson KD, Blieske BE: Vitamin D and cardiovascular disease. Pharmacother: J Human Pharmacol Drug Ther 2009, 29:691–708.
81. Shand AW, Nassar N, Von Dadelzen P, Innis SM, Green TJ: Maternal vitamin D status in pregnancy and adverse pregnancy outcomes in a group at high risk for pre-eclampsia. BJOG 2010, 117:1593–1598.

82. Robinson CJ, Alanis MC, Wagner CL, Hollis BW, Johnson DD: Vitamin D3 regulates the synthesis of gamma-glutamyl transpeptidase and of vitamin D(3). Ann N Y Acad Sci 2007, 1094:25–32.

83. Malcok U, Sengul G, Kadioglu H, Aydin I: Association of the polymorphisms of vitamin D receptor and aggrecan genes with degenerative disc disease. Spine 2001, 26:2543–2547.

84. Eyles DW, Feron F, Cui X, Kesby JP, Harms LH, Ko P, McGrath JJ, Burne TH: The nuclear vitamin D receptor: biological and molecular regulatory properties revealed. J Bone Miner Res 1998, 13:325–349.

85. Shand AW, Nassar N, Von Dadelzen P, Innis SM, Green TJ: Metabolic effects of vitamin D active metabolites on the intervertebral disc. J Bone Miner Res 2001, 16:254–260.

86. Shand AW, Nassar N, Von Dadelzen P, Innis SM, Green TJ: Metabolic effects of vitamin D active metabolites on the intervertebral disc. J Bone Miner Res 2001, 16:254–260.

87. Shand AW, Nassar N, Von Dadelzen P, Innis SM, Green TJ: Metabolic effects of vitamin D active metabolites on the intervertebral disc. J Bone Miner Res 2001, 16:254–260.

88. Shand AW, Nassar N, Von Dadelzen P, Innis SM, Green TJ: Metabolic effects of vitamin D active metabolites on the intervertebral disc. J Bone Miner Res 2001, 16:254–260.

89. Shand AW, Nassar N, Von Dadelzen P, Innis SM, Green TJ: Metabolic effects of vitamin D active metabolites on the intervertebral disc. J Bone Miner Res 2001, 16:254–260.
195. Ha SO, Kim JK, Hong HS, Kim DS, Cho HJ: Expression of brain-derived neurotrophic factor in rat dorsal root ganglia, spinal cord and gracile nuclei in experimental models of neuropathic pain. Neuroscience 2001, 107:381–309.

196. Ohtori S, Takahashi K, Motiya H: Existence of brain-derived neurotrophic factor and vanilloid receptor subtype 1 immunoreactive sensory DRG neurons innervating L5/S1 intervertebral discs in rats. J Orthop Sci 2003, 8:44–57.

197. Cho HJ, Kim JK, Zhou XF, Rush RA: Increased brain-derived neurotrophic factor immunoreactivity in rat dorsal root ganglia and spinal cord following peripheral inflammation. Brain Res 1997, 764:269–272.

198. Obata K, Tsujino H, Yamana K, YD D, Fukuda T, Hashimoto N, Yonenobu K, Yoshikawa H, Noguchi K: Expression of neurotrophic factors in the dorsal root ganglion in a rat model of lumbar disc herniation. Pain 2002, 99:121–132.

199. Costigan M, Woolf CJ: Pain: Molecular mechanisms. J Pain 2000, 1:35–44.

200. Marcol W, Kotsukia K, Larzez-Bryz M, Kowalik JI. BDNF contributes to animal model neuropathic pain after peripheral nerve transaction. Neurosurg Rev 2007, 30:235–243, discussion 243.

201. Fukuda T, Kondo E, Dai Y, Hashimoto N, Noguchi K: Brain-derived neurotrophic factor increases in the uninjured dorsal root ganglion neurons in selective spinal nerve ligation model. J Neurosci 2001, 21:4891–4900.

202. Gruber HE, Ingram JA, Hoelscher G, Zinchenko N, Norton HJ, Hanley EN Jr: Brain-derived neurotrophic factor and its receptor in the human and the sand rat intervertebral disc. Arthritis Res Ther 2008, 10:936.

203. Zhou XF, Chie ET, Deng YS, Zhong JH, Xue Q, Rush RA, Xian CJ: Injured primary sensory neurons switch phenotype for brain-derived neurotrophic factor in the rat. Neuroscience 1999, 92:841–853.

204. Onda A, Murata Y, Rydevik B, Larsson K, Ikuki S, Olmarker K: Immunoreactivity of brain-derived neurotrophic factor in rat dorsal root ganglion in a rat model of lumbar disc herniation. Pain 2002, 99:121–132.

205. Nagano M, Sakai A, Takahashi N, Umino M, Yoshikawa K, Suzuki H: Decreased expression of glial cell line-derived neurotrophic factor signaling in rat models of neuropathic pain. Br J Pharmacol 2003, 140:1252–1260.

206. Boucher TJ, Okuse K, Bennett DL, Munson JB, Wood JN, McMahon SB: Potent analgesic effects of GDNF in neuropathic pain states. Science 2000, 290:124–127.

207. Wang R, Guo W, Ossipov MH, Vanderah TW, Poree F, Lai J: Glial cell line-derived neurotrophic factor normalizes neurochemical changes in injured dorsal root ganglion neurons and prevents the expression of experimental neuropathic pain. Neuroscience 2003, 121:815–824.

208. Gardell LR, Wang R, Ehrenfels C, Ossipov MH, Rossonando AJ, Miller S, Buckley C, Cai AK, Tse A, Foley SF, Gong B, Walus L, Carmillo P, Worley D, Huang C, Engbert T, Pepinsky B, Nate C, Vanderah TW, Lai J, Sah DW, Cones F: Multiple actions of systemic artemin in experimental neuropathic pain. Nat Med 2003, 9:1383–1389.

209. Scholz J, Woolf CJ: The neuropathic pain triad: neurons, immune cells and glia. Nat Neurosci 2007, 10:1361–1368.

210. Costigan M, Scholz J, Woolf CJ: Neuropathic pain: a maladaptive response of the nervous system to damage. Annu Rev Neurosci 2009, 32:1–32.

211. Sommer C, Kress M: Recent findings on how proinflammatory cytokines cause pain: peripheral mechanisms in inflammatory and neuropathic hyperalgesia. Neurosci Lett 2004, 361:184–187.

212. Czechick JC, Hagenacker T, Scharen M, Busselberg D: TNF-alpha differentially modulates ion channels of nociceptive neurons. Neurosci Lett 2008, 434:293–296.

213. Saccabone P, Franchi S, Trovato AE, Valsecchi AE, Panerai AE, Colomei M: Transient early expression of TNF-alpha in sciatic nerve and dorsal root ganglia in a mouse model of painful peripheral neuropathy. Neurobiol Lett 2008, 436:210–213.

214. Tsduda M, Inoue K, Salter MW: Neuropathic pain and spinal microglia: a big problem from molecules in “small” glia. Trends Neurosci 2005, 28:101–107.

215. Hanisch UK: Microglia as a source and target of cytokines. Glia 2002, 40:140–155.

216. Wang Y, Chiang YH, Su TP, Hayashi T, Morales M, Hoffer BJ, Lin SZ: Vitamin D3 attenuates cortical injury induced by middle cerebral arterial ligation in rats. Neuropharmacology 2000, 39:573–880.

217. Navelhan P, Neveu I, Wion D, Brachet P: 1,25-Dihydroxyvitamin D3, an inducer of glial cell line-derived neurotrophic factor. Neuroreport 1996, 7:2171–2175.
218. Kawakami M, Matsumoto T, Kuribayashi K, Tamaki T: mRNA expression of interleukins, phospholipase A2, and nitric oxide synthase in the nerve root and dorsal root ganglion induced by autologous nuclear pulplosis in the rat. J Orthop Res 1999, 17:941–946.

219. Levy D, Zachodnie DW: NO pain: potential roles of nitric oxide in neuropathic pain. Pain Pract 2004, 4:11–18.

220. Harrington JF, Messier AA, Bereste D, Barnes B, Epstein MH: Herniated lumbar disc material as a source of free glutamate available to affect pain signals through the dorsal root ganglion. Spine 2000, 25:929–936.

221. Harrington JF, Messier AA, Hoffman L, Yu E, Dykhuizen M, Barker K: Physiological and behavioral evidence for focal nociception induced by epidermal glutamate infusion in rats. Spine 2005, 30:600–612.

222. Persson JK, Lindh B, Eldér R, Robertson B, Rivero-Mellan C, Eriksson NP, Hofkén T, Aldskogius H: The expression of different cytochemical markers in normal and herniated dorsal root ganglia cells projecting to the nucleus gracilis in the adult rat. Exp Brain Res 1995, 105:331–344.

223. Wildling TJ, Huettner JE: Differential antagonism of alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid-prefering and kainate-prefering receptors by 2,3-benzodiazepine analogs. J Pharmacol Exp Ther 1995, 273:823–830.

224. Wong LA, Mayer ML: Differential modulation by cyclohexidase and concanavalin A of desensitization at native alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid- and kainate-prefering glutamate receptors. Mol Pharmacol 1993, 44:504–510.

225. Ahn SH, Cho YW, Ahn MW, Jang SH, Sohn YK, Kim HS: mRNA expression of cytokines and chemokines in herniated lumbar intervertebral discs. Spine 2002, 27:991–997.

226. Xuistra E, Kusumakar S, Boswell S, Peek E, Urry Z, Richards DF, Adikibi T, Xystrakis E: Reversing the defective induction of IL-10-secreting regulatory T cells in glucocorticoid-resistant asthma patients. J Clin Invest 2006, 116:146–155.

227. Almerighi C, Sinistro A, Cavazza A, Ciaprini C, Rocchi G, Bergamini A: Reactive oxygen species (ROS) play an important role in a rat model of neuropathic pain. Pain 2004, 111:116–124.

228. Funusawa N, Baba H, Miyoshi N, Maezawa Y, Uchida K, Kubo K, Fukuda M: Differential antagonism of alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid and kainate-prefering glutamate receptors in an animal model of neuropathic pain. J Neurosci Res 1999, 60:207–209.

229. Suzuki A, Tokuda K, Koyano Y, Ito Y, Oiso Y, Kozaka O: Effect of vitamin D3 on prostaglandin E2 synthesis in osteoblast-like cells. Prostaglandins Leukot Essent Fatty Acids 1994, 47:27–31.

230. Takahashi M: A mechanism for sciatic pain caused by lumbar disc herniation—involvement of inflammatory cytokines with sciatic pain. Nihon Seikeigeka Gakkai Zasshi 1995, 95:197–201.

231. O’Donnell JL, O’Donnell AL: Prostaglandin E2 content in herniated lumbar disc disease. Spine 1996, 21:1653–1655. discussion 1655-1656.

232. Munamoto T, Atsuta Y, Iwahara T, Sato M, Takematsu Y: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

233. Kanemoto M, Hukuda S, Komiya Y, Katsuura A, Nishioka J: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

234. Lavender P, Lee TH, Corrigan C, Hawrylowicz CM: Differential antagonism of alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid and kainate-prefering glutamate receptors. Mol Pharmacol 1993, 44:504–510.

235. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

236. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

237. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

238. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

239. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

240. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

241. Kanemoto M, Hukuda S, Komiya Y, Katsuura A, Nishioka J: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

242. Kanemoto M, Hukuda S, Komiya Y, Katsuura A, Nishioka J: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

243. Benoist M: The natural history of lumbar disc herniation and radiculopathy. Joint Bone Spine 2002, 69:155–160.

244. Kim HK, Park SK, Zhu J, Tagliatela G, Chung K, Coggeshall RE, Chung JM: Physiological evidence for focal nociception induced by epidermal glutamate infusion in rats. Spine 2005, 30:600–612.

245. Holmén S, Eide K, Snekvik KA, Wasmund NE, Sjovold H: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

246. Holmén S, Eide K, Snekvik KA, Wasmund NE, Sjovold H: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

247. Holmén S, Eide K, Snekvik KA, Wasmund NE, Sjovold H: The action of prostaglandin E2 and triaminoclonine acetamide on the firing activity of lumbar nerve roots. Int Orthop 1997, 21:172–175.

248. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

249. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

250. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

251. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

252. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.

253. Levy D, Hoke A, Zochodnie DW: Local expression of inducible nitric oxide synthase in an animal model of neuropathic pain. Neurosci Lett 1999, 260:207–209.
comparative study between diseased and normal sides. Spine 2000, 25:2191–2199.

262. Franke J, Hesse T, Tournier C, Schubert W, Maxwin C, LeHuex JC, Grasshoff H. Morphological changes of the multifidus muscle in patients with symptomatic lumbar disc herniation. J Neurosurg Spine 2009, 11:70–714.

263. Mattila M, Hurme M, Alaranta H, Paljarvi L, Kalimo H, Falck B, Lehto M, Einola S, Jarvinen M. The multifidus muscle in patients with lumbar disc herniation. A histochemical and morphometric analysis of intraoperative biopsies. Spine 1986, 11:732–738.

264. Hodges P, Holm AK, Hansson T, Holm S. Rapid atrophy of the lumbar multifidus follows experimental disc or nerve root injury. Spine 2006, 31:2926–2933.

265. Hyun JK, Lee JY, Lee SJ, Jeon JY. Asymmetric atrophy of multifidus muscle in patients with unilateral lumbosacral radiculopathy. Spine 2007, 32:E598–E602.

266. Kader DF, Wardlaw D, Smith FW. Correlation between the MRI changes in the lumbar multifidus muscles and leg pain. Clin Radiol 2000, 55:145–149.

267. Boland R. Role of vitamin D in skeletal muscle function. Endocr Rev 1986, 7:434–448.

268. Floyd M, Ayyar DR, Barwick DD, Hudson P, Weightman D. Myopathy in chronic renal failure. Q J Med 1974, 43:509–524.

269. Lazaro RP, Kirshner HS. Proximal muscle weakness in uremia. Case reports and review of the literature. Arch Neurol 1980, 37:555–558.

270. Snijder MB, van Schoor NM, van Hall GM, van der Meer NJM, Lips P. Vitamin D status in relation to one-year risk of recurrent falling in older men and women. J Clin Endocrinol Metab 2006, 91:2980–2985.

271. Ceglia L. Vitamin D and skeletal muscle tissue and function. Mol Aspects Med 2008, 29:407–414.

272. Yoshikawa S, Nakamura T, Tanabe H, Imamura T. Osteomalacic myopathy. Endocrinol Jpn 1979, 26:65–72.

273. Oh JH, Kim SH, Kim JH, Shin YH, Yoon JP, Oh CH. The level of vitamin D in the serum correlates with fatty degeneration of the muscles of the rotator cuff. J Bone Joint Surg British Volume 2009, 91:1587–1593.

274. Tagliafico AS, Ameri P, Bovio M, Puntoni M, Capaccio E, Murialdo G, Martinoni C. Relationship between fatty degeneration of thigh muscles and vitamin D status in the elderly: a preliminary MRI study. AJR Am J Roentgenol 2010, 194:728–734.

275. Tague SE, Clarke GL, Winter MK, McCarrison KE, Wright DE, Smith PG. Vitamin D deficiency promotes skeletal muscle hypersensitivity and sensory hyperinnervation. J Neurosci 2011, 31:13728–13738.

276. Sorensen OH, Lund B, Saltin B, Lund B, Andersen RB, Hjorth L, Melsen F, Mosekilde L. Myopathy in bone loss of ageing: improvement by treatment with 1 alpha-hydroxycholecalciferol and calcium. Clin Sci (Lond) 1979, 56:157–161.

277. Ryan KJ, Daniel ZC, Craggs LJ, Parr T, Brameld JM. Dose-dependent effects of vitamin D on transdifferentiation of skeletal muscle cells to adipose cells. J Endocrinol 2013, 217:45–58.

278. Wu Z, Woodring PJ, Bhakta KS, Tamura K, Wu F, Feramisco JR, Karin M, Wang YJ, Puri PL, p38 and extracellular signal-regulated kinases regulate the myogenic program at multiple steps. Mol Cell Biol 2000, 20:3951–3964.

279. Widmann C, Gibson S, Jarpe MB, Johnson GL. Mitogen-activated protein kinase: conservation of a three-kinase module from yeast to human. Physiol Rev 1999, 79:143–180.

280. Buitrago C, Boland R, de Boland AR. The tyrosine kinase c-Src is required for 1,25(OH)2-vitamin D3 signalling to the nucleus in muscle cells. Biochim Biophys Acta 2001, 1541:79–87.

281. Buitrago CG, Pardo VG, de Boland AR, Boland R. Activation of RAF-1 through Ras and protein kinase Calpha mediates 1alpha,25(OH)2-vitamin D3 regulation of the mitogen-activated protein kinase pathway in muscle cells. J Biol Chem 2003, 278:2199–2205.

282. Buitrago C, Gonzalez Pardo V, de Boland AR. Nongenomic action of 1 alpha,25(OH)2-vitamin D3. Activation of muscle cell PLC gamma through the tyrosine kinase c-Src and PtdIns 3-kinase. Eur J Biochem 2002, 269:2506–2515.

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