J Korean Med Sci 2005; 20: 702-3
ISSN 1011-8934

■ BRIEF COMMUNICATION ■

Indigo Carmine for the Selective Endoscopic Intervertebral Nuclectomy

This study was undertaken to prove that the selectively infiltrated parts of nucleus pulposus with indigo carmine was degenerated parts of nucleus pulposus. This study was done, between August and October 2002, in 5 patients, who received endoscopic discectomy, due to intervertebral disc herniation. Discogram was done with mixture of indigo carmine and radioactive dye. Blue discolored part was removed through endoscope, and small undiscolored part was removed together for the control. The two parts were stained with hematoxylin and eosin and compared under the microscope. Undiscolored part was normal nucleus pulposus, composed of chondrocytes with a matrix of type II collagen and proteoglycan, mainly aggrecan. However, in discolored part, slits with destruction of collagen fiber array and ingrowth of vessel and nerve were observed. Using indigo carmine in endoscopic discectomy gives us selective removal of degenerated disc.

Key Words : *Indigo Carmine; Indigotindisulfonate Sodium; Intervertebral Disk; Diskectomy, Percutaneous*

Inn-Se Kim, Kyung-Hoon Kim, Sang-Wook Shin, Tae-Kyun Kim, Jeung-Il Kim*

Departments of Anesthesia and Pain Medicine, and Orthopedic Surgery*, College of Medicine, Pusan National University, Busan, Korea

Received : 17 January 2005
Accepted : 28 March 2005

Address for correspondence
Kyung Hoon Kim, M.D.
Department of Anesthesia and Pain Medicine, College of Medicine, Pusan National University, 1-10 Ami-dong, Seo-gu, Busan 602-739, Korea
Tel : +82.51-240-7394, Fax : +82.51-242-7466
E-mail : pain@pusan.ac.kr

*Supported by a Grant-in Aid from Pusan National University Hospital in 2003.

According to the development of spinal endoscope instrument and concept of minimally invasive spine surgery (MISS), selective chromoendoscopic nuclectomy for the degenerative nucleus pulposus (NP) has been designed by using a vital or contrast stain (1).

Agents used for chromoendoscopy are categorized according to their working principle. A vital stain such as methylene blue is absorbed into the cells. However, a contrast stain such as indigo carmine is not absorbed but accumulates in pits and valleys between cells highlighting mucosal architecture (2). The low values of pH seen in some degenerate discs are thus likely to be involved in breakdown of the disc matrix (3). Indigo carmine has a molecular weight of 466.35 with its pH 11.6-14.0. This study was undertaken to prove that the selectively infiltrated parts of nucleus pulposus with indigo carmine was degenerated parts of nucleus pulposus.

This study was done, between August and October 2002, in 5 patients, who received endoscopic discectomy, due to intervertebral disc herniation.

Discogram was done with mixture composed of 1 mL of 0.8% indigo carmine (Carmine®, United Korea Pharma., Yeongi, Korea) and 4 mL of radioactive dye (Telebrix®, Gluerbet, Aulnay-sous-Bois, France). Blue discolored part was removed through the endoscope, and small undiscolored part was removed together for the control. The two parts were stained with hematoxylin and eosin (HE) and compared under the microscope.

Undiscolored part was normal nucleus pulposus, composed of chondrocytes with a matrix of type II collagen and proteoglycan, mainly aggrecan (Fig. 1). However, in discolored part, slits with destruction of collagen fiber array and vessel and nerve ingrowth were observed (Fig. 2).

There are typical features of discal degeneration in the three components of the intervertebral disc (IVD), the end-plate (EP), nucleus pulposus, and annulus fibrosus (AF). The normal structure of the NP consists of chondrocytes within a matrix of type II collagen and proteoglycan, mainly aggrecan. The type II collagen fibers do not give the same level of order to the structure or the same degree of mechanical stability to the matrix as in articular cartilage. The proteoglycans are hydrophilic, causing the NP to swell. On HE stained sections, the NP appears homogenous and pale lilac-blue, consistent with its complement of proteoglycans. With the exception of the outer third of the AF, the normal adult IVD is avascular and aneural; there is a relatively clear demarcation between the AF and the NP; and the end-plate forms an intact layer of cartilage overlying cortical bone.

The NP is disrupted in degeneration, with changes in the proportion and types of proteoglycans and collagens (making the NP more eosinophilic), a reduction in the total number of lacunae containing viable chondrocytes, but the formation of chondrocyte clusters in many lacunae, just as in osteoarthritis of articular cartilage, which may even increase the total number of cells present in the diseased tissue. In addition, there is breakdown of the matrix with the formation of permeative 'slit-like' spaces. There is often also disruption of the

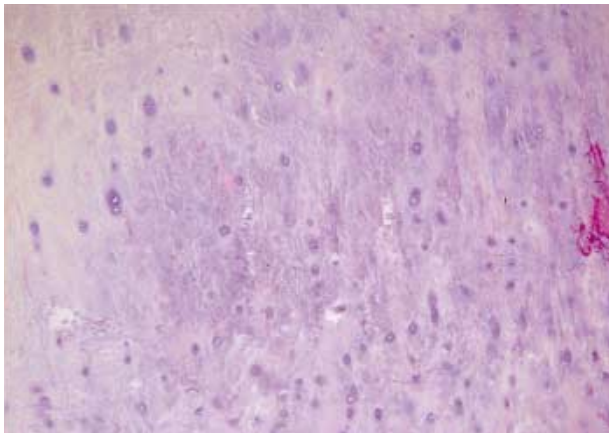

Fig. 1. Normal nucleus pulposus appears homogenous and pale lilac-blue and shows regular chondrocytes array with a matrix of type II collagen and proteoglycan, mainly aggrecan in undiscolored part on H&E stain. With the exception of the outer third of the AF, the normal adult IVD is avascular and aneural (×10).

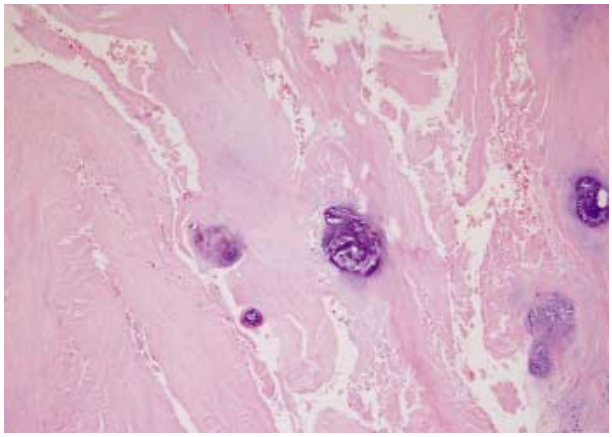

Fig. 2. Degenerated nucleus pulposus shows some slits with destruction of collagen fiber array, vessel and nerve ingrowth, and irregular chondrocytes aggregation (H&E, ×200).

collagen fiber arrays in the AF, traumatic damage to the end plate, and vessel and nerve ingrowth into the inner AF and NP (4). In this study, normal nucleus pulposus, undiscolored part, was composed of chondrocytes with a matrix of type II collagen and proteoglycan, mainly aggrecan. However, discolored part had proper characteristics of degenerated nucleus pulposus, with some slits, destruction of collagen fiber array, and vessel and nerve ingrowth.

Normal discal chondrocytes are characterized by expression of type II collagen and proteoglycans and regulated by the master chondro-regulatory gene, SOX-9. However, in discal degeneration, chondrocyte synthesis of matrix molecules changes differentially with the degree of degeneration, leading to an increase in the synthesis of collagens I and III and decreased production of aggrecan. The regulation of matrix turnover is deranged, affecting both synthesis and degradation. There is a net increase in matrix-degrading enzyme activity over natural inhibitors of such activity, which leads to loss of discal matrix.

Indigo carmine, Indigotindisulfonate, consists essentially of a mixture of disodium 3,3′-dioxo-2,2′-bi-indolylidene-5,5′-disulfonate and disodium 3,3′-dioxo-2,2′-bi-indolyldene-5,7′-disulfonate ($C_{16}H_8N_2Na_2O_8S_2$) and subsidiary coloring matters together with sodium chloride and/or sodium sulfate as the principal uncolored components. Molecular weight is 466.35 and pH is 11.6-14.0. Indigo carmine, like methylene blue, may inhibit endothelium-dependent relaxation by a mechanism that involves two levels. The major action of indigo carmine appears to be at the level of nitric oxide generation and/or release from the endothelial cell. In addition, indigo carmine appears to inhibit vascular smooth muscle guanylyl cyclase. Thus, indigo carmine may elevate blood pressure by interfering with these nitric oxide-mediated vasodilatory mechanisms (5). In this study, intradiscal injection of indigo carmine had little risk to be exposed to blood vessels owing to scant blood supply in the NP.

Selective chromoendoscopy in the spine surgery is based on the specific indicator of indigo carmine that is highly reactive with acidic extracellular matrix in degenerated NP. This study showed a strong evidence for usefulness of application of indigo carmine for selective endoscopic intervertebral nuclectomy in degenerated nucleus. However, there is no strict difference between normal aging and degeneration in IVD. Acidic condition of IVD does not always mean pathologic condition. Removal of all blue-colored nucleus pulposus means removal of all degenerated nucleus pulposus. So as to remove the neural compression of herniated nucleus, decompression has to become the only and main purpose.

REFERENCES

1. Yeung AT. *The evolution of percutaneous spinal endoscopy and discectomy: state of the art. Mt Sinai J Med 2000; 67: 327-32.*
2. Kiesslich R, Neurath MF, Galle PR. *Chromoendoscopy and magnifying endoscopy in patients with gastroesophageal reflux disease. Useful or negligible? Dig Dis 2004; 22: 142-7.*
3. Razaq S, Wilkins RJ, Urban JP. *The effect of extracellular pH on matrix turnover by cells of the bovine nucleus pulposus. Eur Spine J 2003; 12: 341-9.*
4. Freemont AJ, Watkins A, Le Maitre C, Jeziorska M, Hoyland JA. *Current understanding of cellular and molecular events in intervertebral disc degeneration: implications for therapy. J Pathol 2002; 196: 374-9.*
5. Chang KS, Zhong MZ, Davis RF. *Indigo carmine inhibits endothelium-dependent and -independent vasodilation. Hypertension 1996; 27: 228-34.*