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IDENTIFICATION OF MICROBES FROM SURGICALLY EXCISED LUMBAR DISC HERNIATIONS: A STUDY AMONG SRI LANKAN SUBJECTS

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Introduction

Disc herniation is a common disorder not only among elder people but also among young teenage population. It is hypothesized that traumatic work and genetic predisposition play an important role in disc herniation. Even though the main causative factor is not yet identified, the number of admission is currently increasing. Studies have reported severe back pain and other related symptoms occur mainly due to disc herniation and disc degeneration interfering with the daily lives of many subjects by not only making them bed ridden but also decreasing the quality of life. It is said that approximately 80% of the population may have experienced lower back pain during their lifetime which is considered to be one of the major medical issues at work place absence. According to the prevailing data the prevalence of disc prolapse in European countries is about 1-3%.

Treatments and diagnostic procedures (such as neuro surgical procedures and Magnetic Resonance Imaging - MRI) associated with disc herniation are very costly and does not provide a permanent remedy. Recurrent surgical invasion had been evidenced in several lumbar discectomy patients. In addition to the medical cost, indirect costs, such as decreased productivity in the population are of major issue. It is estimated that £ 12 billion per annum in UK and 1.7% of the gross national product in the Netherlands is expensed for this health issue.

Studies carried out in other countries have stated that there could be a role played by the microorganisms in association with disc herniation other than traumatic work and genetic predisposition (vitamin D receptor gene, aggrecan gene). Although there is increasing number of hospital admissions related to disc herniation and degeneration in Sri Lankan population, so far there had not been any studies carried out to identify the role of microbes leading to the above condition. Hence the present study was conducted to identify the presence of bacteria in disc herniated patients and to provide a better patient compliance if identified.

Methodology

After confirming lumbar herniation with MRI by a consultant neurosurgeon and consultant radiologist thirty patients admitted to the neurosurgical unit of a selected hospital in Colombo for lumbar discectomy were included in this study. Ethical approval was obtained from Ethics Review Committee of University of Sri Jayawardenapura and informed written consent was obtained from all individual participants prior to the study. Standardized, 45-item, interviewer administered questionnaires was given to each patient enquiring their personnel information, daily activities and behaviours, work status, general health status and current health condition. Major causative factor for disc herniation as stated by the patients was noted from the clinical history.

Standard protocol was used for disinfecting the skin pre operatively-2 times with 70% (v/v) isopropyl alcohol and 3 times with povidone iodine solution before the skin incision was performed. After skin disinfection, skin scrapings, and muscle biopsies were obtained. Each sample was transferred into individual Robertson's cooked meat enrichment broth (RCM) using sterile forceps. Surgically removed disc was also transferred into RCM for anaerobic studies and another portion of the excised disc material was taken into sterile container for aerobic studies. All surgical instruments and transferring instruments were sterilized before invasion. Direct Gram stain was performed for disc samples and were cultured on blood agar, chocolate agar, MacConkey agar and incubated at 37 °C for 48 hours for isolation of aerobic bacteria. In addition, samples were enriched on Brain heart infusion (BHI) agar for 24 hours. After 24 hours of incubation, enriched samples were sub cultured on blood agar, chocolate agar and MacConkey agar and further incubated for 24 hours at 37 °C. Blood agar and MacConkey agar plates were incubated in room air, while chocolate agar plates were kept in candle jar in 5-10% CO_2.

For anaerobic isolation, removed disc samples in RCM enrichment broth were incubated at 37 °C for 48 hours. It was sub cultured on blood agar, bacteriods bile esculin agar and brucella blood agar and incubated at 37 °C anaerobically with anaerobic packs. Anaerobic plates were read at day 2, 7 and 21. Bacteriological cultures were done for skin scrapings and muscle biopsy samples obtained from the surgical site. This is to ensure bacteria isolated from the lumbar disc are not a contaminant. Gram stain was performed for isolations. Anaerobic culture identification was done using RAPID ANA ID kit. Gram stain, catalase and coagulase tests were used for identification of *Staphylococcus* spp.

Results and Discussion

Among the study subjects, two were positive for anaerobic cultures, and four were positive for aerobic cultures, when intervertebral disc tissue was analyzed following dissectomy. All disc samples that were positive for aerobic cultures were identified as, coagulase negative *Staphylococci spp.* Among the disc samples that were positive for anaerobic culture, one-disc sample was identified to have *Gemella morbillorum* (According to kit analysis probability of isolation of *Gemella morbilorum* is > 99.9% in the positive culture). In addition, this patient had recurrent dissectomy and also presented with family history of lumbar disc prolapse. The other subject who was identified with anaerobic positive disc had suffered with lower back pain for more than 20 years. The anaerobic species are yet to be identified. Among the aerobic positive subjects who were confirmed as coagulase negative *Staphylococci* spp one presented with recurrent dissectomy.

Table 1. Summary of main categories stated by the patients as the causative factor for disc herniation in the study subjects

Category	No. of individuals = 30 *Age (18 – 73 y), Male-15, Female-15*
Family history of disc prolapse	02 (6.6 %)
Lifting weight, fallen	08 (26.6 %)
Traumatic occupation	02 (6.6 %)
Recurrent dissectomy	04 (13.3 %)
Patients without any special clinical history	14 (46.6 %)

Previous studies carried out in other counties have also stated the evidence of 24/64 (38%) *Propionibacterium acnes* (*P. acnes*) and coagulase negative *Staphylococci* spp (5/64 - 8%) in intervertebral disc tissue cultures following lumbar dissectomy (Rollason *et al.*, 2013), where as in another study it was found that 16/36 (84%) *P. acnes*, 2/36 (11%) coagulase negative *Staphylococci* spp and 1/36 (5%) *Corynebacteruim propinquum* were present in the intervertebral disc tissue cultures (Stirling *et al.*, 2001). This present study also had a similar percentage of (13% of total) coagulase negative *Staphylococci* spp.

Even though 2/30 was positive for anaerobes in the disc materials, none of these patients had either anaerobe positive muscle or skin scrapings. Further, the aerobic positive patients in this population did not show positive aerobic cultures on muscle scraping. Even though few subjects had skin positive for aerobes, this concludes further that microbe positive disc could not be due to contamination.

In addition, according to the results (Table 1), only 10/30 subjects have traumatic occupation, lifting heavy weights and fallen history, while another two patients had family history whereas more than half (50%) of the study population (14/30) did not engage in such traumatic work. This provides evidences to anticipate the traditional hypothesis that traumatic work and genetic predisposition plays a major role in disc herniation.

Conclusions

Present study indicates in patients who had lumbar discectomy a good percentage (20%) had either aerobic or anaerobic positive microbial cultures in the disc samples. Hence this study provides further evidence to support a possible association of microbes in low grade infection in disc herniation causing symptoms of low back pain and inflammation.

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