INVESTIGATION OF THE STRUCTURE OF THE INTERVERTEBRAL DISC USING HIGH FREQUENCY ULTRASOUND AND ULTRASOUND CT

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Introduction. The lumbar Intervertebral Disc (IVD) is a common source of low back pain. It consists of a tough outer fibrocartilagenous Annulus Fibrosus (AF), a soft inner Nucleus Pulposus (NP), both of which are bounded above and below by a thin layer of hyaline cartilage, the cartilage end-plates. Its structure is key to understanding not only its mechanical functioning but also the pathological processes that affect the IVD and the way in which the IVD subsequently fails and causes pain. Thus far, study of the detailed structure of the IVD have been restricted to laborious dissection techniques (Marchand and Ahmed, 1991) and histology, both of which lead to the destruction of the IVD. The objective of this study was to assess the potential of observing the detailed internal structure of the intervertebral disc non-destructively using two methods of ultrasonic imaging: acoustic microscopy (AM) and continuous wave ultrasound computer tomography (CW Ultrasonic CT). Specifically the hypotheses that: (1) that the acoustic images correspond to the structure observed using conventional histological techniques and, (2) ultrasound is sensitive to pathological abnormalities within the disc, were tested.

Materials and Methods. Specimens. Two bovine tails and four porcine tails and lumbar spines were obtained fresh from an abattoir and frozen en-bloc until required. Specimens were defrosted and dissected to remove all soft tissue to leave only the vertebral bodies and intervening IVDs. IVDs were removed from the spine/tails via a transverse cut through the mid-body of the adjacent vertebral bodies. IVDs were then mounted into either imaging unit using specially constructed specimen holders.

Acoustic Microscope. The acoustic microscope consists of a 50MHz transducer, a series of three motorised stages driven by stepper motors, a bath for coupling media all of which is supported by an aluminium frame. The stages give three axis of motion: linear (X-axis), vertical (Y-axis) and rotation (R-axis). The transducer operates in pulse-echo mode. All images are displayed as brightness-modulated timebase (B-scan) scans in which the amplitude of the reflected echoes from tissue boundaries determines the brightness of the display of that boundary. The time between the propagation of the initial pulse and the return of the echo gives a measure of the depth of the reflecting boundary. A complete transverse section through the tissue was achieved using spatial compounding, a process in which a series of B-scans are performed at different tissue orientations and superimposed. On a selection of IVDs marks were placed on the specimen using dye to identify the scanned area, these IVDs then underwent histological processing.

CW Ultrasonic CT. The acoustic scanner consists of two 10 MHz transducers, one for transmitting and the other for receiving continuous wave ultrasound. The specimen is rotated at 300 rpm. As the object rotates with respect to the stationary ultrasound transducers, the scattering centres within the object return echoes that are Doppler shifted in frequency by an amount depending on the velocity of the individual scatterers. An instantaneous plot of amplitude as a function of frequency can be interpreted as a tomographic projection. The back-projection method is used to reconstruct the image. This method of imaging is discussed in more detail elsewhere (Liang et al., 2000).

Histology. Marked IVDs were dehydrated and embedded in LR White resin. Porcine tail IVDs were sectioned on a conventional microtome whereas the larger bovine tail and porcine spine IVDs were sectioned on a sledge-micromote; section thickness 8-15μm. Sections were either stained with Goldner’s Trichrome or left unstained and viewed under polarised light.

Experimental Procedure. Transverse sections through the porcine lumbar discs and bovine tail discs were acquired using CW Ultrasound CT. Detailed B-scan images, in both transverse and longitudinal planes, were conducted in all discs using the acoustic microscope. Spatial compounding was performed on porcine tail IVDs with a diameter of approximately 1 cm.

Results. CW ultrasound CT revealed a repetitive banding structure within the periphery of the image characteristic of the lamellar organisation within the AF: the path of the lamellae could be followed around sections of the AF. Images also show clearly that the thickness of the lamellae increase towards the centre of the disc. Banding and reflections were less apparent in the centre of the disc that corresponds to the NP. AM images of the disc provided more detailed images. As with CW ultrasound CT, AM images revealed a repetitive banding pattern representing the lamellae within the AF, spatial compounding of porcine tail discs enabled individual bands to be observed around the circumference of the AF (Figure 1A) in which discontinuities were noted. Lamellar thickness varied: 48-65μm, 120-162μm and 45-129μm for porcine and bovine tail discs and porcine lumbar discs respectively. Lamellar thickness was less varied in the tail discs due to the circular profile, however, in lumbar discs there was a clear distinction between anterior and posterolateral regions. Spatial compounding also revealed a pathological circumferential tear in the substance of the AF in one of the porcine tail specimens; this was also shown in histological preparations of the same disc. Other structures were also clearly identifiable including the epiphyseal growth plate (Figure 1B) and also the posterior longitudinal ligament.