Introduction. The Intervertebral Disc (IVD) remains the most implicated structure with regards to low back pain. Its health is critically linked to the concentration of proteoglycans within the nucleus pulposus that confer a large swelling potential upon the disc, causing it to imbibe water and swell. In vivo, this occurs on a daily basis as high loads experienced during the day lead to a loss in disc height that is replaced overnight when the load is removed during recumbent sleep when the disc swells ([Adams et al., 1981]). The movement of water through the disc not only accounts for the disc's capacity for load bearing and recovery from mechanical stress, but also provides nutrition of the disc. For these reasons, numerous studies have sought to quantify the swelling pressure in relation to disc nutrition (e.g. Urban and Maroudas, 1981) and examine the effects of loading characteristics on the hydration of the disc. The majority of these studies have only quantified hydration in terms of the change in mass after the wet tissue is dried, it is unclear how swelling affects the IVD morphologically both from an external and internal perspective. Indeed, external morphological changes may be important with regards the way in which the IVD interacts with its surrounding structures. Also, internal structural changes as the disc swells may lead to the progression of pathological abnormalities within the AF. Therefore, the objectives of this study were to observe and quantify the external and internal morphological changes of the disc by using the novel approaches of time-lapse photography and acoustic microscopy respectively. Several hypotheses were tested: (1) that the disc expands equally in all directions, (2) the internal morphology changes with swelling and, (3) the lamellar thickness decreases as the disc swells.

Materials and Methods. Specimens. Three Porcine tails were obtained fresh from an abattoir, two were frozen until required. All soft tissue was removed from the specimens to leave only the vertebral bodies and intervening IVDs. IVDs approximately 1cm diameter were selected and excised from the tail by transecting the adjacent vertebra in the centre. The IVD was then mounted on a specially constructed specimen holder. Approximately 5 or 6 small marks of permanent histological dye were placed on the anterior of both cartilage end-plates (CEPs) in approximate vertical alignment with one another using a hypodermic needle. Two marks were then placed on the left and right sides of the disc itself. Once marked the specimen was mounted in the acoustic microscope and oriented so that the marks faced the digital camera and were clearly visible.

Time-Lapse Photography. A colour video camera was mounted on a tripod facing the anterior of the disc. A frame grabber card was used to acquire still images with a resolution of 768 by 576 pixels with a user-set period between each acquisition. Illumination of the disc was provided by a fluorescent lamp to avoid heating the specimen.

Acoustic Microscope. The acoustic microscope consists of a 50 MHz transducer, three motorised scanning stages driven by stepper motors, and a bath for coupling media. A series of electronics are used to generate and detect transmitted and reflected signals, all of which are digitized by an oscilloscope and transferred to PC where an image is constructed. The transducer operates in pulse-echo mode and all images are displayed as a brightness-modulated timebase (B-scan) in which the amplitude of echoes reflected from tissue boundaries determines the brightness of the display. The time difference between the propagation of the initial pulse and the echo’s return gives a measure of the depth of the reflecting surface.

Image Analysis. Time-lapse images were individually processed using Scion Image Analysis (Scion Corporation.). Markers on the CEPs were identified and a distance between corresponding markers calculated to give the IVDs vertical dimensions, similarly, markers on the disc itself were identified and the horizontal distance between them calculated to give the radial dimensions. Acoustic B-scans were inspected visually. Lamellae consistently appeared in each image and the distance across these were measured. Changes in vertical, radial and internal lamellar dimensions are all expressed as a percentage change from the original size at time zero.

Experimental Procedure. The PC was configured to acquire an image of the specimen every 10 minutes for a duration of 6 hours. After each image acquisition a B-scan was performed transversely across the centre of the disc. Before each scan the transducers distance from the disc surface was checked and standardised to ensure the focus of the beam remained a constant distance within the disc and no erroneous structures appeared due to the disc swelling.

Results. Time-lapse photography revealed changes in both the vertical and radial dimensions. Vertical dimensions increased slowly for the first 50 minutes but then increased almost linearly at the rate of approximately 0.007min⁻¹ until 250 minutes when a plateau was reached at 16% (Figure 1). Changes in radial dimensions were much smaller than those in vertical height (Figure 1). Initially, the disc swells rapidly in the radial direction before returning to its original dimensions and then shrinking.

Discussion. The results of this study clearly show that the disc does not swell equally in both the vertical and radial directions. Initially swelling is greater in the radial direction, which then decreases due to the increasing tension within the annular fibres as a result of the rapid gain in vertical height. We have demonstrated that the internal structure of the disc changes with swelling. Lamellar thickness changes in a similar manner to the external radial changes and, whilst small, these changes may play a role in excentuating an annular tear. The small porcine caudal discs examined have a circular cross-section, swelling characteristic may be affected by the re-entrant posterior profile exhibited by human lumbar discs.
