Retinol binding protein 4 is expressed in chondrocytes of developing mouse long bones: implications for a local role in formation of the secondary ossification center

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Online Resource 3 Rbp4 is present in cells giving rise to the nucleus pulposus

a) Alcian blue/nuclear fast red staining of vertebrae showing the developing nucleus pulposus of the intervertebral disc (arrow). The developing centre of ossification (o) of the vertebrae surrounded by chondrocytes undergoing hypertrophy (h) is also shown. Cartilage, blue; nuclei, red. b) Magnified view of boxed section in (a). c) Corresponding serial section showing Rbp4 in these cells (Rbp4, green; nuclei, blue). Scale bar a) 420 µm, b and c) 70 µm.

Examination of P1 vertebrae showed that Rbp4 was present in developing intervertebral disks (a, boxed and c), but not within the developing vertebrae (data not shown). Close examination of the developing disk shows that Rbp4 was restricted to cells within the nucleus pulposus (c). Ossification center-associated hypertrophic chondrocytes (h) did not express Rbp4 above background levels (data not shown).