Inherited Rickets

in

Corriedale Sheep

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ABSTRACT

Inherited rickets of Corriedale sheep is a newly discovered skeletal disease of sheep with simple autosomal recessive inheritance. The clinical signs resemble rickets in other species and include decreased growth rate, thoracic lordosis and angular limb deformities. Radiographic features include physeal thickening, blurred metaphyseal trabeculae and thickened porous cortices. Computed tomography scanning of long bones reveals increased bone mineral content and cortical area, but decreased bone mineral density. Gross lesions include segmental thickening of physes, growth arrest lines, collapse of subchondral bone of the humeral head, thickened cortices and enthesophytes around distal limb joints. Microscopically there is persistence of hypertrophic chondrocytes at sites of endochondral ossification, inappropriate and excessive osteoclastic resorption, microfractures and wide, unmineralised osteoid seams lining trabeculae and filling secondary osteons.

Affected sheep are persistently hypophosphataemic and hypocalcaemic. Normal serum 25-hydroxyvitamin D_3 concentration accompanied by a two-fold elevation in 1,25-dihydroxyvitamin D_3 ($1,25(OH)_2D_3$) suggested a defect in end-organ responsiveness to vitamin D as a likely mechanism, but this was not supported by *in vitro* studies using cultured skin fibroblasts. These studies revealed normal vitamin D receptor function and the presence of 24-hydroxylase mRNA in cells from affected sheep, even without induction by $1,25(OH)_2D_3$. Inappropriate overexpression of 25-hydroxyvitamin D_3-24-hydroxylase, the enzyme that breaks down active vitamin D, is therefore considered the probable cause of inherited rickets in Corriedale sheep. Such a

mechanism has not previously been described as a cause of inherited rickets in humans or other animal species. Treatment of affected sheep with high oral doses of vitamin D_3 weekly for 3 months showed a trend towards increased bone mineral density, thus supporting an intact vitamin D receptor. Preliminary studies on immune function revealed reduced numbers of CD4+ and CD8+ lymphocytes and reduced interferon-γ production by lymphocytes stimulated with parasite antigen.

This new form of inherited rickets may be widespread in the New Zealand Corriedale sheep population and has considerable potential as a model for studying aspects of vitamin D metabolism.

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Table of Contents

List of Figures

List of Tables

GLOSSARY

$1,25(OH)_2D$	1,25-dihydroxyvitamin D, active vitamin D, calcitriol
25(OH)D	25-hydroxyvitamin D, calcidiol
24-hydroxylase	25-hydroxyvitamin D-24-hydroxylase, CYP24, CYP24A1
$[^3H]$ $1,25(OH)_2D_3$	tritium labelled 1,25-dihydroxyvitamin D_3
ADHR	autosomal dominant hypophosphataemic rickets
AF2	ligand dependent activation function 2
alopecia	lack of hair or wool
ALP	alkaline phosphatase
ankylosis	fusion of joint
annexin-V	cell surface marker of apoptosis
ANOVA	analysis of variance
apoptosis	programmed cell death
ARHR	autosomal recessive hypophosphataemic rickets
basophilic	increase in blue staining
BMC	bone mineral content
BMD	bone mineral density
cAMP	cyclic adenosine monophosphate
CaSR	calcium sensing receptor
cholecalciferol	vitamin D_3, (3β,5*Z*,7*E*)-9,10-secocholesta-5,7,10(19)-trien-3-ol
ConA	*Concanavalia ensiformis* type IV-S
chondrocyte	cartilage cell
craniotabes	reduction in mineralisation of the skull, with abnormal softness of bone
CYP24	25-hydroxyvitamin D-24-hydroxylase, CYP24A1
CYP27B1	25-hydroxyvitamin D-1α-hydroxylase, 1α-hydroxylase
DEXA	dual energy x-ray absorptiometry
diaphysis	main or midsection (shaft) of a long bone
DMP1	dentin matrix protein 1
endosteum	tissue lining the medullary cavity of a bone
enthesophyte	calcification of a muscle or ligament attachment to bone
eosinophilic	increase in pink/red staining

epiphysis	the end of a long bone, separated from the shaft by the physis
ergocalciferol	vitamin D_2, (3β,5Z,7E,22E)-9,10-secoergosta-5,7,10(19),22-tetraen-3-ol
FGF	fibroblast growth factor
fibrous osteodystrophy	lesion where fibrous tissue replaces resorbed bone
GAPDH	glyceraldehyde-3-phosphate dehydrogenase
heterozygous	different alleles at one locus
HHRH	hereditary hypophoshataemic rickets with hypercalciuria
HVDRR	hereditary vitamin D-resistant rickets
hypercalcaemia	higher than normal serum calcium
hypercalciuria	excess of calcium in the urine
hyperphosphataemia	higher than normal serum phosphate
hyperplasia	increased formation of new cells
hypocalcaemia	lower than normal serum calcium
hypophosphataemia	lower than normal serum phosphate
hypoplasia	incomplete growth of a tissue
hypotonia	abnormally decreased strength
IFN-γ	interferon-gamma
lordosis	downward curvature of the spine
LPS	lipopolysaccharide
lymphopenia	lower than normal number of lymphocytes in the blood
MEPE	matrix extracellular phosphoglycoprotein
metaphysis	area of trabecular bone and thin cortex in between the epiphysis and diaphysis, consisting of primary and secondary spongiosa
monocytopenia	lower than normal number of monocytes in the blood
mRNA	messenger ribonucleic acid
myelofibrosis	replacement of bone marrow by fibrous tissue
nephrocalcinosis	deposition of calcium phosphate in the renal tubules
olsen P	test for soil phosphorus levels
OPG	osteoprotegerin
osteoblast	mesenchymal bone cell that makes osteoid
osteocalcin	non-collagenous bone protein
osteoclast	multinucleated bone cell that resorbs bone
osteogenesis imperfecta	inherited defect in type I collagen formation, leading to fragile bones

osteoid	matrix of collagen and non-collagenous bone proteins produced by osteoblasts
osteomalacia	impaired mineralisation of bone, leading to softening and accumulation of excess osteoid
osteopetrosis	hereditary disease that results in abnormally dense bone and retention of the primary spongiosa
osteoporosis	pathological loss of bone but the remaining bone is structurally normal
periosteum	connective tissue covering the outside of bones
phagocytosis	process of engulfing and destroying of bacteria and other foreign material
PHEX	phosphate-regulating gene with homologies to endopeptidases on the X-chromosome
phosphaturia	excess phosphate in the urine
physis	cartilaginous growth plate, plural=physes
pQCT	peripheral quantitative computed tomography
primary spongiosa	newly-formed bone beneath the physis with calcified cartilage core
PTH	parathyroid hormone
rachitic rosary	enlarged costochondral junctions
RANK	receptor activator of NF-κB
RANKL	receptor activator of the NF-κB ligand
recessive gene	gene that expresses itself in the homozygous state, but not in the presence of a dominant allele
rickets	bone disease due to a failure of endochondral ossification and lack of mineralisation of newly formed osteoid
RT-PCR	reverse transcriptase polymerase chain reaction
Runx2	runt related transcription factor 2
RXR	retinoid X receptor
sclerosis	hardening
secondary hyperparathyroidism	abnormally increased secretion of PTH as a result of either nutrtional deficiency of calcium or renal failure
secondary spongiosa	formed when the primary spongiosa undergoes remodelling
sFRP4	secreted frizzled-related protein 4
SMAD	mothers against decapentaplegic homolog
SSI	stress-strain index
SUG1	suppressor for gal, a regulatory component of the 26S proteosome

TGF-β	transforming growth factor-β
TRPV 5,6	transient receptor potential vanilloid 5, 6, calcium channels
valgus	deformity where the angulation is away from the midline of the body
varus	deformity where the angulation is towards the midline of the body
VDDR I	vitamin D-dependent rickets type I, also called pseudovitamin D-deficiency rickets, due to a defect in 1α-hydroxylase enzyme
VDDR II	vitamin D-dependent rickets type II, also called hereditary vitamin D-resistant rickets, due to a defect in vitamin D receptor
VDR	vitamin D receptor
VDRE	vitamin D-response element
VEGF	vascular endothelial growth factor
voxel	volume element, region in a tissue slice that corresponds to a pixel in an image
XLH	X-linked hypophosphataemic rickets
Wnt	wingless-ints