

## SUPPLEMENTARY MATERIAL

TABLE SIV

**Characteristics of the animal and veterinary science studies that reported the use of BTMs in different types of research.**

Authors	Population	Type of Study	Markers	Time	Conclusion
Chavassieux et al. (1991)	Fourteen ewes	Determine the effects of ossein-hydroxyapatite compound on bone remodeling	Serum ALP, OC, Ca and P	90 days	Possibly the OHC is able to reduce the seasonal effect on bone turnover
Wan Zahari et al. (1994)	Ten sheep	Effects of nutrition on bone growth	Serum 1, 25 Vit. D, ALP and TRAP	6 weeks	Diets rich in phosphate do not have effect on skeletal mineralization
Turner et al. (1995)	Thirty ewes	Represent changes in bone mass in ovariectomized ewes	Serum BALP	6 months	This model may be useful for estrogen deficiency studies inducing bone loss
Chavassieux et al. (1997)	Thirty-two ewes	Use of glucocorticoid for decrease of bone formation	Serum OC and BALP	7 months	Use of glucocorticoid in ewe may represent valid model for bone loss
Scott et al. (1997)	Twenty- four sheep	Effects of nutrition on bone growth	Serum TRAP, OC and BALP; Urinary PYD and DPD	90 days	Suggest that markers may be useful for diagnosis and treatment of bone disease and early detection of nutrition deficiencies
Nicodemo et al. (1999)	Twenty-four sheep	Influence of diet in bone growth	Serum BALP, OC, Ca and P; Urinary PYD and DPD	11 weeks	Markers are unsuitable for assessment of bone growth induced by different diets
Chanetsa et al. (2000)	Forty sheep	Effects of an estrogen agonist on growth and bone mineral accretion	Serum BALP, TRAP, 1, 25 Vit. D, Ca, Mg and P	163 days	This study has clinical relevance for treating children with delays in growth and bone mineral accretion
Chavassieux et al. (2001)	Forty ewes	The effects of OVX in ewe associated or not with lentaron and effects of a new selective estrogen receptor modulator	Serum OC and BALP; Urinary CTx	6 months	OVX induced an increase in bone turnover and MDL may be useful for prevention of postmenopausal bone loss
Lill et al. (2002a)	Eight sheep	Which method is more effective to induce osteoporosis		6 months	The most effective method to induce osteoporosis is a combination of diet, ovariectomy, and glucocorticoid
Lill et al. (2002b)	Thirty-two sheep	Induce severe osteoporosis in an ovine model		7 months	The model may be useful for studies on osteoporosis
Windhagen et al. (2002)	Fourteen sheep	What is the response of turnover markers during distraction osteogenesis	Urinary DPD and PYD; Serum OC	74 days	Showed a pattern of osteoblast cellular activation during distraction osteogenesis
Liesegang et al. (2003)	Twelve goats and sheep	Determine the diurnal variation in bone markers	Serum BALP, ICTP, CL and OC	2 weeks	During the day there was a variation in concentration of markers, goats presented a higher bone turnover than sheep
Klein et al. (2004)	Fourteen sheep	Use of BTMs to represent callus consolidation in bone healing	Serum PICP, BALP and PIIINP	9 weeks	The markers used were not useful for represent bone healing
MacLeay et al. (2004a)	Fifty-two ewes	Develop an animal model for human postmenopausal osteoporosis		90 days	Model is sensitive for bone loss due to a dietary metabolic acidosis
MacLeay et al. (2004b)	Twenty-four sheep	Influence of diet and OVX on bone turnover	Urinary DPD; Serum BALP, Ca and P	180 days	Model is sensitive for bone loss due to a dietary metabolic acidosis

**TABLE SIV (continuation)**

<b>Authors</b>	<b>Population</b>	<b>Type of Study</b>	<b>Markers</b>	<b>Time</b>	<b>Conclusion</b>
Newton et al. (2004)	Twelve ewes	Effects of ovariectomy on the trabeculae of ovine iliac bone	Serum OC; Urinary PYD	12 months	The ovine model is adequate for changes in trabecular bone architecture studies
Liesegang and Risteli (2005)	Six sheep and six goat	Influence of diet	Serum ICTP, BALP, OC, Ca, CL and 1, 25 Vit. D	8 weeks	Due to short duration, it is difficult to associate the diet with bone turnover
Seebeck et al. (2005)	Sixteen sheep	Use bone markers to represent callus formation during fracture healing	Serum PICP, ALP, BALP, PIIINP, Ca and P	9 weeks	The markers used were not useful in representing callus formation
Liesegang et al. (2006)	Twelve goat and sheep	Determine the effects of pregnancy and lactation on markers	Milk and serum Ca; Serum OC, BALP, CTx, ICTP and 1, 25 Vit. D	11 months	Markers showed that bone turnover occurred during gestation and lactation
Arens et al. (2007)	Eight sheep	Measure the seasonal variations in quantity and quality of bone turnover	Serum BALP, PYD; Urinary DPD	18 months	Seasonal variation must be considered when using an ovine model in osteoporosis studies
Liesegang et al. (2007)	Twelve goats and sheep	Determine the effects of a second pregnancy and lactation on markers in comparison with a first	Milk and serum Ca; Serum OC, BALP, CTx, ICTP and 1, 25 Vit. D	11 months	The bone loss in the second pregnancy and lactation is lower than in the first, possibly due to an adaptation of the organism
Sigrist et al. (2007)	Fourteen sheep	The effect of ovariectomy on bone metabolism in sheep	Serum BALP, PYD; Urinary DPD	18 months	The ovine model is not an appropriate model for human postmenopausal osteoporosis
Dias et al. (2008)	Eighteen ewes	Measurement of bone markers in ewes under controlled environmental factors, and the study of their correlation with serum minerals	Serum ALP, BALP, OC, Ca, P, Mg and Ca <sup>2+</sup>	6 weeks	References for the serum values of bone turnover parameters in sheep could be of great value, possibly for obtaining an early prognosis of fracture healing
Goebel et al. (2009)	Eighty-five sheep	Verification of FGF23 as a possible marker of bone healing and regeneration	Serum ALP, Ca and P; Urinary P	42 days	FGF23 is a promising marker for indicating bone healing
Ding et al. (2010)	Eighteen sheep	Use of glucocorticoid for inducing osteopenia in cancellous bone		10 months	This method is useful for induced osteoporosis in sheep
Vernon et al. (2010)	Twenty sheep	Influence of exercise on the degradation of the articular cartilage	Serum LOX and C2C	5 months	Markers were unable to demonstrate the effects of forced exercise
Liesegang et al. (2013)	Twenty-four sheep	Influence of diet	Serum ICTP, BALP, Ca, P and 1, 25 Vit. D	4 months	Changes in bone turnover associated with diet
Tralman et al. (2013)	Seven sheep	Compare two methods of osteosynthesis in sheep	Serum ALP and OC	10 weeks	Use of RTP fixator is more effective than plate fixation in osteotomies of long bones in sheep
Kreipke et al. (2014)	Thirteen sheep	The effect of ovariectomy on vertebral bodies and femoral condyles in sheep after 1 and 2 years		24 months	The vertebral bodies are preferable for trabecular microarchitecture studies
Sousa et al. (2014a)	Ninety sheep	Measure the values of bone markers and evaluate the correlation between those and serum minerals in sheep of various ages and different physiologic stages	Serum ALP, BALP, Ca, Mg and P	1 day	The measure of lifespan in sheep is useful in preclinical orthopedic research and provide information complementary for other analyses with imaging

## BONE TURNOVER MARKERS IN SMALL RUMINANTS

TABLE SIV (continuation)

Authors	Population	Type of Study	Markers	Time	Conclusion
Sousa et al. (2014b)	Eighteen sheep	Assessment of the short-term variation in the BTMs serum levels	Serum ALP, BALP, OC, PIIINP, DPD, TRAP, Ca and P	12 weeks	The variability in short-term does not seem to be a limitation for studies with bone markers
Andreasen et al. (2015)	Twenty ewes	Assessment of cellular events during the remodeling process induced by glucocorticoid in ovariectomised sheep	Serum CTx and OC	7 months	It is a useful animal model due to significant bone loss compared to osteoporosis in postmenopausal women
Kielbowicz et al. (2015, 2016)	Forty-nine ewes	Assessment of different factors after osteoporosis induction with glucocorticoid	Serum BALP, CTx, estradiol, cortisol, progesterone and parathormone		Glucocorticoid treatment in ovariectomised sheep was considered by the authors as a very appropriate method for osteoporosis induction