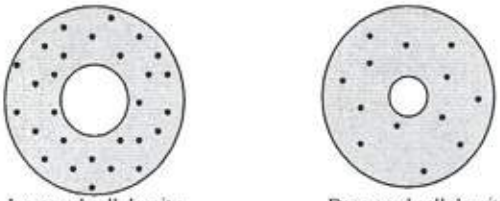
Abstract 79: Orthopaedic Research Society; Vol. 34. 2009.

Do Osteocyte Densities Control Osteon Wall Thickness? The Hypothesis is Supported in Non-human Appendicular Bones But not in Male Human Ribs

*Clark, G C; +*Skedros, J G; Taylor, K W; Sorenson, S M; **Qiu, S
+*University of Utah Department of Orthopaedic Surgery, Salt Lake City, Utah
Senior author jskedros@utahboneandjoint.com

INTRODUCTION: In appendicular skeletons of many species, secondary osteons (Haversian systems) are important in maintaining mechanical competence and mineral homeostasis. Marotti and co-workers [1] hypothesized that during the formation of secondary osteons, the newly formed osteocytes send inhibitory signals to surrounding osteoblasts. This signal causes the osteoblasts to cease bone formation and differentiate into osteocytes as they become embedded in the newly formed matrix. Martin expanded on this concept with his 'unifying theory of bone remodeling' [2], which suggested that osteoblasts remaining at the termination of osteon infilling differentiate into a distinct cell type — bone lining cells (BLC) — that maintain communication with the embedded osteocytes via gap junctions. At the termination of osteon infilling, instead of regulating osteocyte formation, the interconnected osteocytes send inhibitory signals to the BLC that suppresses the activation of osteoclast-mediated resorption that would lead to a new remodeling cycle. Martin further hypothesized that the strength of the inhibitory signal is proportional to the number of osteocytes in communication with the BLC, with a relatively greater repression exerted by progressively greater numbers of osteocytes (i.e. increased numbers of osteocytes correlate with thinner osteon walls) as shown in the figure below.



Increased cell density increases inhibitory signal of osteon in-filling

Decreased cell density decreases inhibitory signal of osteon in-filling

Although this hypothesis of an inverse relationship between cell number and tissue volume has clear implications for the maintenance of skeletal mass in diseases where this relationship is impaired (e.g. osteoporosis), it has been adequately tested in only a few bones. Experimental data reported by Metz, Martin, and coworkers [3] in ulnae of four-to-five year old, skeletally mature sheep support the idea that osteocytes form a repressive network. Although Qiu et al. [4] argued in favor of this relationship, their analysis of adult human rib osteons dealt with Ot.Lc.N/B.Ar vs. HC.Ar and HC.Ar/On.Ar , but not specifically with osteon wall thickness (HC = Haversian canal, Ot.Lc = osteocyte lacunae, On.Ar = osteon area, B.Ar = bone area). These comparisons in human rib osteons, however, showed no or little correlation (0.011 and 0.104, respectively, and only the latter is statistically significant). Additionally, to our knowledge, the only other study that we could locate that has addressed this hypothesis examined osteons in cortices of human proximal femora that were possibly confounded by the affects of aging and/or disease (osteoporosis) [5,6]. The purpose of the present investigation was to test this hypothesis in formed osteons from human ribs and various non-human limb bones including those of horses, deer, elk, and sheep.

METHODS: 50X backscattered electron images were obtained from the mid-third diaphyses of 10 mature (ages 2-10) equine radii and third metacarpals (MC3s), and from sheep, deer, elk, and equine calcanei. Human rib data were from Qiu et al., [4]. Over 10,000 fully formed osteons were examined and a regression analysis was performed on osteons in all species to show relationships between osteon wall thickness and lacunae number (Ot.Lc.N) and osteon wall thickness and osteocyte lacunae number per bone area (Ot.Lc.N/B.Ar).

RESULTS: A summary of results of the correlation analyses for each species is shown in the table below. Each non-human appendicular bone showed a statistically significant negative correlation between Ot.Lc.N/B.Ar and osteon wall thickness ($p < 0.001$), supporting the hypothesis of Martin [2] and Metz et al. [3]. Analysis of human rib data, however, showed a significant ($p = 0.04$) but comparatively weaker negative correlation ($r = -0.10$). By contrast, in all species Ot.Lc.N showed a very strong positive correlation ($r \geq 0.72$) with osteon wall thickness in: **1)** all non-human bones, and **2)** human ribs, which showed the strongest positive correlations ($r = 0.89$; $p \leq 0.001$).

Ot.Lc.N/B.Ar and Ot.Lc.N vs. Osteon Wall Thickness

Species	Cortex	Ot.Lc.N/B.Ar r	Ot.Lc.N/B.Ar p	Ot.Lc.N r	Ot.Lc.N p
Sheep, Calcaneus	Dorsal	-0.35	<0.0438	0.83	<0.001
	Plantar	-0.50	<0.001	0.77	<0.001
	Medial+Lateral	-0.46	<0.001	0.84	<0.001
	All locations (all osteons)	-0.47	<0.001	0.80	<0.001
Deer Calcaneus	Dorsal	-0.28	<0.001	0.76	<0.001
	Plantar	-0.42	<0.001	0.75	<0.001
	Medial+Lateral	-	-	-	-
	All locations (all osteons)	-0.34	<0.001	0.74	<0.001
Elk Calcaneus	Dorsal	-0.43	<0.001	0.80	<0.001
	Plantar	-0.33	<0.006	0.87	<0.001
	Medial+Lateral	-0.37	<0.002	0.82	<0.001
	All locations (all osteons)	-0.36	<0.001	0.83	<0.001
Equine Calcaneus	Dorsal	-0.40	<0.001	0.78	<0.001
	Plantar	-0.43	<0.001	0.74	<0.001
	Medial+Lateral	-0.56	<0.001	0.84	<0.001
	All locations (all osteons)	-0.47	<0.001	0.79	<0.001
Equine Radius	Cranial	-0.39	<0.001	0.74	<0.001
	Caudal	-0.33	<0.001	0.73	<0.001
	Medial+Lateral	-0.47	<0.001	0.74	<0.001
	All locations (all osteons)	-0.39	<0.001	0.73	<0.001
Equine MC3	Dorsal	-0.42	<0.001	0.73	<0.001
	Plantar	-0.44	<0.001	0.73	<0.001
	Medial+Lateral	-0.40	<0.001	0.76	<0.001
	All locations (all osteons)	-0.42	<0.001	0.74	<0.001
Human Rib	All locations (all osteons)	-0.10	<0.04	0.89	<0.001

DISCUSSION: The inverse relationship between osteocyte density and osteon wall thickness reported by Martin [2] and Metz et al. [3] is supported by our data in all non-human bones. This finding is important in understating the influence osteocytes have on osteon in-filling and its implications for the pathogenesis of osteoporosis. The weak correlations of the human rib data, however, might reflect fundamental differences between ribs and appendicular bones, supporting the argument that osteon remodeling dynamics in ribs should not be broadly applied to all bones. For example, ribs: **1)** are metabolically more active (e.g. have higher basal remodeling rates) and are more sensitive to hormonal changes (e.g. during pregnancy, lactation, and antler growth) [7,8,9], **2)** receive frequent low-strain loading, **3)** are derived from sclerotomes of the somites, and **4)** are phylogenetically primitive, appearing in the fossil record well before limb bones, which could allow for the evolution of morphogenetic fields that differ in the degree that cellular/hormonal or other biomechanical mechanisms have in controlling the requisition of calcium from non-weight-bearing bones (ribs) when compared to weight-bearing limb bones. Therefore, in these perspectives, it is plausible that rib osteons are inherently biased toward physiologic interactions that affect molecular exchange between surface area (i.e., Haversian canal perimeter) and bone volume (i.e., osteon wall area) in ways that are not generally applicable, and may be deleterious by increasing porosity and reducing tissue density, if employed by osteons in the appendicular skeleton. In addition to data showing that the basal remodeling rate in ribs is not only greater than in limb bones [7,8], the suggested possibility that osteon dynamics fundamentally differ between ribs and limb bones might also be reflected in the generally greater sensitivity that the axial skeleton (e.g. ribs and skull) has to hormonal and/or other types of non-strain stimuli [10].

REFERENCES: 1) Marotti 1996 Ital J Anat Embryol 101:25-79; 2) Martin 2000 Bone 26:1-6; 3) Metz et al. 2003 Bone 33 753-9; 4) Qiu et al. 2003 Anat Rec 272A:520-25; 5) Power et al. 2001 Calcif Tissue Int 69:13-19; 6) Power et al. 2002 Bone 30:859-65; 7) Tommerup et al., 1993 J Bone and Min Res 8:1053-57; 8) Raab et al., 1991 J Bone and Min Res 6:741-49; 9) Vajda et al., 1999 Biol Reprod 61:1439-44; 10) Rawlinson et al. 1995 J Bone and Min Res 10:1225-32

** Bone Res. Lab, Henry Ford Hosp., Detroit, MI; NIH grant DK43858