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Iliac crest histomorphometry and skeletal heterogeneity in men

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ABSTRACT

Purpose: The cortical characteristics of the iliac crest in male have rarely been investigated with quantitative histomorphometry. Also it is still unknown how cortical microarchitecture may vary between the iliac crest and fractures related sites at the proximal femur. We studied the microarchitecture of both external and internal cortices within the iliac crest, and compared the results with femoral neck and subtrochanteric femoral shaft sites.

Methods: Undecalcified histological sections of the iliac crest were obtained bicortically from cadavers ($n = 20$, aged 18–82 years, males). They were cut (7 μm) and stained using modified Masson-Goldner stain. Histomorphometric parameters of cortical bone were analysed with low (×50) and high (×100) magnification, after identifying cortical bone boundaries using our previously validated method. Within cortical bone area, only complete osteons with typical concentric lamellae and cement line were selected and measured.

Results: At the iliac crest, the mean cortical width of external cortex was higher than at the internal cortex ($p < 0.001$). Also, osteon structural parameters, e.g. mean osteonal perimeter, were higher in the external cortex ($p < 0.05$). In both external and internal cortices, pore number per cortical bone area was higher in young subjects (≤50 years) ($p < 0.05$) while mean pore perimeter was higher in the old subjects (>50 years) ($p < 0.05$). Several cortical parameters (e.g. osteon area per cortical bone area, pore number per cortical area) were the lowest in the femoral neck ($p < 0.05$). The maximal osteonal diameter and mean wall width were the highest in the external cortex of the iliac crest ($p < 0.05$), and the mean cortical width, osteon number per cortical area were the highest in the subtrochanteric femoral shaft ($p < 0.05$). Some osteonal structural parameters (e.g. min osteonal diameter) were significantly positively correlated ($0.29 \le R^2 \le 0.45$, $p < 0.05$) between the external iliac crest and the femoral neck.

Conclusions: This study reveals heterogeneity in cortical microarchitecture between the external and internal iliac crest cortices, as well as between the iliac crest, the femoral neck and the subtrochanteric femoral shaft. Standard iliac crest biopsy does not reflect accurately cortical microarchitecture of other skeletal sites.

1. Introduction

It has been well established that age-related bone loss occurs throughout the skeleton and affects both cortical and cancellous bone in the normal population (Vedi et al., 1982; Zebaze et al., 2010). In humans, histomorphometric analysis of the underlying changes in bone remodelling and microarchitecture that may predispose to bone loss has been mostly carried out in the iliac crest (Podenphant et al., 1986; Recker and Barger-Lux, 2006). Although iliac crest may not be representative of the clinically relevant sites such as the proximal femur (Dempster, 1989; Eventov et al., 1991), it has been selected internationally as the standardized site for biopsy in histological investigations of the metabolic bone disease (Dempster and Shane, 2001).

Traditionally, studies of iliac crest biopsy have focused almost exclusively on cancellous bone, since disorders in cancellous bone remodelling might be widely held responsible for common metabolic bone disorders in adults, such as postmenopausal osteoporosis (Arlot et al., 1990; Kimmel et al., 1990). In recent years, it has become apparent that excluding cortical bone from analysis may limit the ability to detect fundamental aspects of bone characteristics. Many studies focused on age-related changes in the iliac crest cortical structures (Schnitzler and Mesquita, 2006; Vedi et al., 2011), and the effects of growth as expressed in differences between the external and internal iliac crest cortices were demonstrated by studies on children (Schnitzler et al.,

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Table 1
Basic anthropometric data of the cadavers. Individual values and the mean ± SD are shown.

Age [years]	Group*	Height [cm]	Weight [kg]	BMI [kg/m^2]
17	1	178	74	23.4
22	1	186	106	30.6
29	1	184	105	31
32	1	171	69	23.6
34	1	187	102	29.2
36	1	177	74	23.6
39	1	185	84	24.5
43	1	171	98	33.5
44	1	179	96	30
46	1	185	85	24.8
48	1	178	85	26.8
50	1	185	108	31.6
52	2	180	136	42
53	2	176	73	23.6
58	2	175	73	23.8
58	2	169	96	33.6
62	2	170	68	23.5
74	2	166	64	23.2
77	2	177	72	23
82	2	165	53	19.5
47 ± 18.2		177 ± 6.9	84 ± 20.4	27.2 ± 5.3

* Group 1 (≤50 years, $n = 12$); Group 2 (>50 years, $n = 8$).

2009; Rauch et al., 2006). Chappard et al. found no significant differences for any cortical parameters between biopsies from the right and left iliac crests in the same individual (Chappard et al., 2008), while a difference in bone formation rate between the two cortices of the same iliac crest biopsy in women with osteoporosis was reported by Balena et al. (1992). Misof and co-workers indicated a difference in calcium content between the iliac cortices, and they also revealed that the bone mineralization density distribution (BMDD) in cortices of a transiliac biopsy generally correlates with the corresponding values in the trabecular compartment (Misof et al., 2014). However, few data is available for comparison of structural characteristics between the iliac crest cortices in healthy adults.

Studies analysing the skeletal microarchitecture enables better understanding of bone alterations due to aging and pathology (Amling et al., 1996). However, most of these studies are based on restricted sites of the skeleton, e.g. iliac crest, spine and proximal femur. Previous skeletal heterogeneity related studies have predominantly focused on the differences in structural and remodelling parameters of cancellous bone (Hildebrand et al., 1999; Lochmüller et al., 2008; Aaron et al., 2015), or the physical measurements of cortical composition (Boskey et al., 2016; Scerpella et al., 2016). Except for the cortical width (Dempster et al., 1993; Castillo et al., 2012), structural characteristics of the cortical bone have rarely been compared between different skeletal sites. Considering the fact that the cortical microarchitecture is complex and a considerable skeletal heterogeneity exists between the axial and appendicular subdivisions of the skeleton (Marcus et al., 2009), the analysis of cortical bone throughout the skeleton of the same individual is needed.

In this study, cortical properties were compared between both cortices of the iliac crest, as well as to those reported earlier in the femoral neck and subtrochanteric femoral shaft of the same subject (Tong et al., 2015a; Tong et al., 2016). Both non-fracture (iliac crest) and fracture (proximal femur) skeletal sites were studied.

2. Materials and methods

2.1. Subjects

Iliac crests were obtained from 20 male cadavers (mean age, 47 ± 18.2 years, range 17–82 years) at Kuopio University Hospital, Kuopio, Finland. The subjects were divided into two sub-groups based on their age: young (≤50 years, $n = 12$), and old (>50 years, $n = 8$) (Table 1). There was no previous history of medical conditions or use of drugs known to affect bone metabolism. Ethical approval for collection of samples was granted by the National Authority for Medicolegal Affairs (permission number: 5783/04/044/07).

2.2. Sample preparation

Iliac crest biopsies were taken bicortically from a standardized site located 2 cm below and posterior to the anterosuperior iliac spine (Tamminen et al., 2011) (Fig. 1). Samples were dehydrated in ethanol before being embedded in polymethylmethacrylate (PMMA) according to standard protocols (Raum, 2008). After embedding, 7-μm-thick sections were cut using a microtome (Reichert-Jung; Cambridge Instruments, Heidelberg, Germany) and stained with modified Masson Goldner trichrome stain. The entire section of the iliac crest was scanned using an auto-image scanner (Particle Analyzer; Carl Zeiss, Jena, Germany) to acquire a complete histological image (×50) for histomorphometric analysis (Fig. 1). An image program (GNU Image

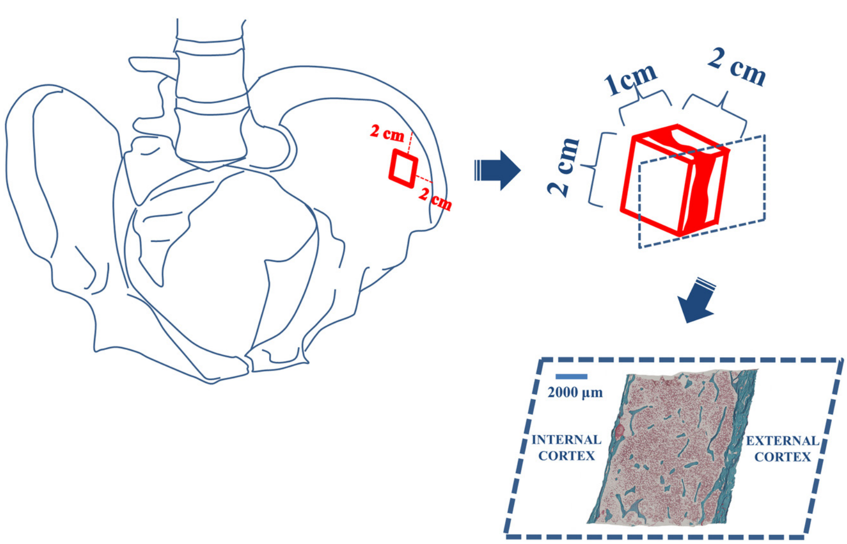

Fig. 1. Iliac crest biopsies taken bicortically from a standardized site (2 cm below and posterior to the anterosuperior iliac spine) were indicated with red square. The anterior section was scanned to acquire a complete histological image (×50) (include both cortices) for histomorphometric analysis.