

REPRODUCTIVE TRADE-OFFS IN SKELETAL HEALTH AND PHYSICAL
ACTIVITY AMONG THE INDIGENOUS SHUAR OF ECUADORIAN
AMAZONIA: A LIFE HISTORY APPROACH

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DISSERTATION ABSTRACT

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Reproductive effort is a central element of human biology and ecology. Particularly for females, reproduction is energetically demanding, with elevated metabolic costs during pregnancy and lactation, followed by high child care costs. To satisfy energetic needs, women can adopt various physiological and behavioral strategies. On a physiological level, the energetic requirements of offspring may be met by adjusting metabolic allocation and/or drawing on maternal bodily reserves. On a behavioral level, women may reduce energy expenditure and/or increase energy intake.

This study examined reproductive trade-offs in activity and skeletal health among the indigenous Shuar forager-horticulturalists of Ecuadorian Amazonia and had two main objectives. First, this research examined trade-offs in energy use during female reproductive states and behavioral adjustments made by females and males to meet high reproductive demands. Second, this study investigated skeletal health profiles among Shuar, as well non-Shuar *Colonos*, to identify the relationships between female reproductive factors and skeletal health.

Research was conducted among adults in four Morona-Santiago communities. Skeletal health was measured using calcaneal ultrasonometry, and physical activity was measured using accelerometry. Extensive information on sociodemographics and reproduction was assessed through structured interviews.

Age-related declines in bone mineral density (BMD) were observed for Shuar and *Colonos*, while Shuar BMD was significantly higher than that of other populations. These results suggest that normative data from developed countries may reflect suboptimal bone density levels. Regarding reproductive effects on skeletal health, results indicate that earlier menarcheal age and greater stature are associated with better bone health in postmenopausal life. These conclusions suggest the importance of the timing of early developmental stages in establishing bone status in adulthood.

Results demonstrate that physical activity levels were similar between pregnant/lactating (P/L) and other women. However, P/L women appear to compensate for elevated energetic demands by relying on a male partner who has increased his energy expenditure, suggesting greater participation in subsistence activities.

Overall, this study demonstrates the importance of biocultural strategies among women to meet high reproductive costs. Further, it emphasizes the utility of a life history framework for identifying trade-offs in physiology and behavior.

This dissertation contains previously published and unpublished co-authored material.

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Madimenos FC, Snodgrass JJ, Blackwell AB, Liebert MA, Sugiyama LS. 2011. Physical activity in an indigenous Ecuadorian forager-horticulturalist population as measured using accelerometry. *Am J Hum Biol* 23:488-497.

Madimenos FC, Snodgrass JJ, Blackwell AB, Liebert MA, Cepon TJ, Sugiyama

LS. 2011. Normative calcaneal quantitative ultrasound data for the indigenous Shuar and non-Shuar Colonos of the Ecuadorian Amazon. *Archives of Osteoporosis* 6:doi: 10.1007/s11657-011-0056-x.

Madimenos FC, Snodgrass JJ, Blackwell AD, Liebert MA, Sugiyama LS. 2011. Reproductive trade-offs and bone health among Shuar and Colonos females from Amazonian Ecuador. *Am J Hum Biol* 23(2):264.

Blackwell AD, Snodgrass, JJ, Madimenos FC, Sugiyama LS. 2010. Life history, immune function, and intestinal helminths: Trade-offs among immunoglobulin E, C-reactive protein, and growth in an Amazonian population. *Am J Hum Biol* 22(6):836-848.

Nelson GC, Madimenos FC. 2010. Obelionic cranial deformation in the Puebloan Southwest. *Am J Phys Anthropol* 143(3):465-472.

Madimenos FC, Snodgrass JJ, Blackwell AD, Liebert MA, Sugiyama LS. 2010. Reproductive history and bone health among female Shuar and Colonos from the Ecuadorian Amazon. *Proc AAAS* 29(1):76-77.

Madimenos FC, Cepon TJ, Snodgrass JJ, Sugiyama LS. 2010. Corroborating accelerometry-based physical activity data with self-report data. *Am J Hum Biol* 22(2):261.

Madimenos FC, Liebert MA, Snodgrass JJ, Blackwell AD, Sugiyama LS. 2010. A profile of bone health in an Ecuadorian forager-horticulturalist population. *Am J Phys Anthropol* 141(S50):160.

Madimenos FC. 2009. Dental evidence for subsistence strategy differences in pre-contact Ipiutak and Tigara of Point Hope, Alaska. *Am J Phys Anthropol* 38(S48):180.

Madimenos FC, Snodgrass JJ, Blackwell AD, Cepon TJ, Gandolfo TR, Sugiyama LS. 2009. Physical activity measured using accelerometry in an indigenous lowland Ecuadorian population. *Am J Hum Biol* 20(2):260.

Madimenos FC, Snodgrass JJ, Leonard WR. 2008. Recent trends in human biology: a content analysis of the *American Journal of Human Biology*. *Am J Hum Biol* 20:226.

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CHAPTER I

INTRODUCTION

Natural selection is the only evolutionary mechanism that consistently adapts populations to their environments. Although it is common to view natural selection as a process that retains variations that increase survival and reproduction, ultimately it is the effects of heritable differences on differential reproduction that resonates through generations. Even survival only has selective value to the extent that it leads to the proliferation of an organism's genes in future generations, which is ultimately achieved through reproduction (Ellison, 2001). While variation in reproductive biology is not the sole conduit through which variation in reproductive success can be achieved, it is an extremely important one, and one that is shaped by ecological conditions. This highlights the critical importance of investigations in reproductive ecology within the field of evolutionary biology.

Reproductive ecology examines the regulation of reproductive effort, and, specifically, the allocation of resources to reproduction at the expense of competing physiological demands. For human females, reproduction represents the most energetically demanding phase of life, with high metabolic costs of pregnancy and lactation, followed by the high costs associated with provisioning dependent offspring, typically several at once. These costs may be so high that a forager woman could not bear them alone (Hill and Hurtado 2009; Hrdy, 2005; Kramer, 2005; Reiches et al., 2009;

Snodgrass, 2011). In order to satisfy these high energetic costs, women can adopt a variety of physiological, behavioral, or cultural strategies. On a physiological level, the high energetic and nutrient requirements of the fetus or neonate may be met by drawing on maternal energy and nutrient reserves available from fat, lean muscle, and/or skeletal tissues. Reproduction for females is sensitive to energy availability, which affects menarche, fecundity, gestation length, lactation, and the resumption of menses post-parturition (Ellison, 2001; 2003). On a behavioral level, pregnant and lactating women may compensate for the heightened energetic demands of reproduction through strategies such as reducing energy expenditure for other activities, or increasing energy intake, potentially by recruiting additional investment from others (e.g., Hrdy, 2005; Marlowe, 2003; Sugiyama and Chacon, 2005). On a cultural level, bridewealth and the widespread distribution of game among foragers, for instance, can be seen as normative customs that provide females with resources necessary to satisfy reproductive costs. Both local ecological conditions (Gray, 1994; Panter Brick, 1993; Singh et al., 1989) and the availability of social support (Piperata, 2009; Sellen and Smay, 2001) are thus critical to the strategies adopted by different women in a variety of contexts. Regarding the latter, assistance through provisioning of food by mates, kin, or others can increase maternal energy intake, whereas assistance with chores and work tasks can relieve a woman from the energetic requirements of doing work herself. This allows her to conserve energy and resources that may then be re-allocated to meet the woman's elevated metabolic needs of pregnancy, lactation, or feeding of her offspring (Gibson and Mace, 2005; Gurven and Kaplan, 2008; Hrdy, 2005; Kramer, 2005). Because both physical and social environmental conditions are essential to shaping female reproductive ecology, the

physiological and social impacts of reproduction will vary among and within human populations.

The three studies presented in this dissertation contribute to research in human biology in two main ways. First, this dissertation examines trade-offs in energy use during critical female reproductive states, and the biological and social accommodations that are made in response to the high costs of these states. This goal is achieved by investigating the relationship between reproduction and skeletal health among the subsistence-based, natural fertility Indigenous Shuar forager-horticulturalists of Ecuadorian Amazonia and then, by documenting the behavioral strategies used by Shuar women to compensate for these elevated reproductive needs. Second, this research establishes age- and sex- related skeletal health profiles among the Shuar and neighboring non-Shuar *Colonos*. Minimal data on skeletal health and measured physical activity are available for subsistence-based populations, largely because of the methodological challenges of gathering these data in field settings. However, recent advancements in calcaneal ultrasonometry and accelerometry have facilitated the collection of data on bone health and activity levels outside of laboratory and clinical settings. This dissertation takes advantages of these methodological advances in order to measure bone density and physical activity from a remote population of forager-horticulturalists in Ecuador. For comparative purposes, bone density data are also presented for the non-Indigenous *Colono* population who are biologically unrelated to the Shuar but live in the same geographical region.

The studies described in this dissertation employ a biocultural approach to understand aspects of health. A biocultural perspective predicates a dialectical

relationship between human biology and cultural forces, and its application in this dissertation addresses the influences of socio-cultural elements on biology and simultaneously, the ways that biology influence socio-cultural factors. Further, this research describes the energetic strategies used to meet the demands of reproduction, which, in an energy-constrained environment, will be at the cost of competing physiological demands. This fundamental principle of life history theory—that under conditions with scarce energetic resources, humans make decisions about the allocation towards competing physiological domains—is the underlying theoretical framework throughout this dissertation and is examined in greater detail in the next section. Following this discussion, the second half of this chapter will provide an ethnographic background of the participant population.

LIFE HISTORY THEORY APPROACH

Like all organisms, humans face trade-offs regarding the allocation of energetic resources towards competing functional demands. This basic tenet of life history theory posits that in an environment where energy resources are limited, energetic investment towards one function will impact and limit energy allocation towards other functions (Charnov and Schaffer, 1973; Gadgil and Bossert, 1970; Hill and Hurtado, 1996; Hill and Kaplan, 1999; Lessels, 1991; Stearns, 1976; Trivers 1972, 1974). Energetic resources may include both the energy available in the environment and the availability of metabolic energy. Because optimal energy allocation changes as the individual grows and then reproduces, natural selection is expected to have shaped age and context dependent strategies of energy allocation to different functions across the lifespan; these allocations

are influenced by multiple physiological, neurobiological, and behavioral mechanisms. Competing functions are often subsumed under the demands of growth, somatic maintenance, and reproduction, although each is comprised of a vast array of different functional mechanisms, with trade-offs expected between them.

Maternal Life-History Strategies: Physiological and Behavioral

In human females of reproductive age, energy is divided to support two main types of function, survival and reproduction. Energy allocated to survival is used to keep the female alive and support basal metabolic needs, cellular maintenance and repair, immune function, many cognitive functions, and non-reproductive behavior such as food acquisition. Reproduction, on the other hand, is the most energetically demanding stage of life for females and entails additional energy costs, which must be met either by increasing energy intake, or reducing energy allocated to other functions (Snodgrass, 2011). Energy allocated to reproduction is used, for example, to maintain ovarian and uterine function, support pregnancy and lactation, and allow the behaviors necessary for child care. The energetic costs of pregnancy and lactation are particularly high compared to other reproductive functions. For example, the menstrual cycle requires approximately 6% more energetic resources above that which is allocated to maintain basic biological functions, or basal metabolic rate (BMR) (Howe et al., 1993; Meijer et al., 1992; Volland , 1998). In contrast, pregnancy necessitates on average an additional 85-475 kilocalories (kcal) per day, depending on trimester, in order to ensure adequate fetal and placental growth, growth of maternal tissues such as breasts and uterus, and fat deposition (FAO/WHO/UNU, 2004). The energetic cost of lactation is even more physiologically

taxing, although these costs are shaped by the composition and volume of breast milk produced (Dewey, 1997). While energetic needs during lactation fluctuate with child's age and nursing intensity, it requires, on average, an additional 675 kcal/day when exclusively breastfeeding during the first six months of life, and 460 kcal/day in later months (FAO/UNU/WHO, 2004). In some populations, this cost can comprise up to half of the mother's total energy budget. It is important to note, therefore, that these estimates are determined from well-nourished women in industrialized nations and may overestimate the actual physiological requirements to females (Jasienska, 2001). Several ecologically and environmentally-shaped bio-behavioral options are available to meet these heightened demands, including increasing energy intake (EI), mobilizing body fat stores, decreasing energy expenditure (EE), or a combination of these strategies (Dufour et al., 2002; Goldberg et al., 1991; Piperata and Dufour, 2007; Valeggia and Ellison, 2001).

In affluent countries, pregnant and lactating women who are well nourished generally increase EI and reduce EE in order to maintain and conserve sufficient fat and nutrient reserves (Jasienska, 2004). However, among populations with insufficient food availability and high female workloads, increasing EI and decreasing EE are not always possible. Among Gambian women with poor nutritional status, pregnancy and lactation are marked by a down-regulation of BMR, which re-allocates energy from the female's own physiological demands to those of the growing fetus or infant (Jasienska, 2004). However, among nutritionally-compromised women with heavy workloads, the energetic savings afforded by reduction in BMR may not be sufficient to compensate for the intense physical work effort required by their daily life, which may inhibit a woman's

ability to store fat during pregnancy. In turn, insufficient fat stores can lead to poor lactational performance and milk quality. It can also reduce maternal fat reserves and lean tissue with successive births and lactation periods (Jelliffe and Maddocks, 1964; Ulijaszek, 1995). This phenomenon, called the maternal depletion syndrome, is notorious in developing countries and may result in premature labor and low neonatal birth weights. Tracer (1991) found that Au women of Papua New Guinea exhibited both a short-term decline in adiposity following childbirth and a long-term fertility-related decline. Similar trade-offs between maternal and fetal conditions have been documented in nutritionally-stressed populations in Colombia (Gil et al., 1981), Philippines (Popkin et al., 1993), Kenya (Little et al., 1992), and Guatemala (Kurz et al., 1993). Thus, while female reproductive biology allows for reproduction even when energy supply is limited, this may have long-term costs in the form of poor reproductive outcomes, maternal physiological deterioration, and diminished future reproductive potential (Jasienska, 2003).

Female physiology appears to be adapted to minimize the risk of maternal depletion and poor birth outcomes through facultative modulation of ovarian function in response to reductions in energy availability. This down-regulation of ovarian function reduces the chances of investing in a pregnancy that has a low probability of success. Further, it allows for the repair of maternal nutritional status between successive births (Jasienska, 2003). Reproductive down-regulation, which under intense energy stress may lead to reproductive suppression, has been observed across populations who are experiencing transient periods of energetic stress. For example, among !Kung hunter-gatherers of Botswana, low fertility rates have been associated with heavy EE linked to

foraging activities and a nomadic lifestyle (Bentley, 1985). In Lese women from the Congo, Ellison et al. (1989) found that during pre-harvest season when little food is available, ovarian suppression occurs in response to low energy availability, negative energy balance, and low energy flux, a state characterized by low EI and low EE. When harvest resumes and food resources are more secure, ovarian function resumes, resulting in a seasonal pattern of conceptions and births (Ellison, 2008). Similarly, among Tamang agriculturalists of Nepal, Panter-Brick and colleagues (1993) documented that down-regulation of female ovarian function occurs during seasonal periods of high workload, despite a fairly stable annual food supply. In both Lese and Tamang women, the effects of intense physical activity on reproductive function are confounded by the effects of negative energy balance (i.e., energy imbalance with EE greater than EI). When considering women in populations with seasonal periods of high energy flux (e.g., high EE and high EI), such as rural Polish farmworkers, ovarian suppression may occur as a result of increases in daily energy expenditure despite consistent availability of food (Jasienska and Ellison, 1998). These examples suggest that periods of energy imbalance associated with high energy expenditure from intense work output coupled with relatively low energy intake may signal that not enough energetic resources are available for reproduction, and that energy-saving mechanisms such as down-regulation of BMR may be insufficient to support pregnancy and lactation (Ellison, 2003; Ellison and Jasienska, 2007). Temporary down-regulation or full suppression of reproductive function appears, therefore, to be an adaptive response that optimizes birth spacing that serves to ensure longer term reproductive success.

The physiological strategies that females adopt to compensate for reproductive demands are further shaped by biosocial conditions within which they live. Both local ecological conditions (e.g., rainfall patterns, extent of seasonality, etc.) (Gray, 1994; Sellen and Smay, 2001; Singh et al., 1989) and adherence to cultural norms and practices such as food taboos and work restrictions immediately post-parturition (Piperata and Dufour, 2007; Piperata, 2008) can affect female energetic strategies. Recently, much focus has been paid to the effect of maternal social networks on reproduction, especially in subsistence-based economies. Kin support can relieve subsistence and domestic chores and provide food for the pregnant or lactating female, supplementing her dietary intake and activity patterns, and which allow her to modify the extent to which she must utilize her body stores (Piperata, 2009). For example, lactating Toba women from Argentina who had girl assistants were found to spend less time engaging in domestic work than women without support (Bove et al., 2002). Among the Turkana pastoralists of Kenya, women with fewer children and co-wives spent more time in herding activities than did women in larger, wealthier households (Gray, 1995). Among Amazonian *Ribeirinha* woman, Piperata (2009) noted that lactating women with social support had higher overall energy intake, and spent significantly less time in subsistence work than women with lower levels of social support. Furthermore, Marlowe (2003) observed that among the Hadza of Tanzania, men with pregnant or lactating partners had higher hunting returns, which served to compensate for the increased energy demands of their mates. Social networks are particularly important in subsistence-based populations where reduced work output often translates into a decrease in contribution to food production, with repercussions for the well-being of the entire family. Thus, an examination of the

physiological strategies used to conserve maternal energetic stores requires thoughtful consideration of the larger ecological and social complex that frame the maternal condition.

Reproduction and Skeletal Health

In addition to energy, nutrient stores in specific tissues are drawn upon during pregnancy and lactation. Bone is a composite tissue that varies in quantity and quality over the course of the lifespan in response to various endogenous and exogenous factors (Agarwal and Glencross, 2010). Some key factors implicated in bone integrity and maintenance include biological sex, heredity, nutrition, reproductive history (in females) and physical activity, the effects of which fluctuate across the lifespan (see details in Chapter II). Approaching skeletal health using a life course perspective acknowledges that every life stage or transition represents a point along a cumulative progression and that the dynamic interaction between developmental phases across the lifecycle is largely fueled by the availability of energy and energetic resources necessary to modulate these stages (Leidy, 1996). In the case of female reproduction, skeletal growth and maintenance may be compromised if reproductive effort is prioritized, and vice versa. However, while energetic studies over the past several decades have made great progress in applying a life history framework to understanding the regulation of fecundity, few studies have applied this perspective to understanding the regulation of reproductive trade-offs affecting female skeletal health.

Human females, like most mammals, exhibit determinate growth; all energy that exceeds what is needed for maintenance is allocated to longitudinal growth during the

juvenile period and then growth stops as the reproductive phase of life begins, coinciding with the onset of menarche (Charnov, 1993). Trade-offs between female reproduction and longitudinal skeletal growth have been documented in several studies, showing relationships between earlier age at menarche, first birth and shorter adult stature. With menarche, influxes of estrogen hormones that function to maintain bone mass also play a role in epiphyseal closure (see Chapter II). For example, Kramer and Greaves (2011) found that among the indigenous Pumé foragers of Venezuela, girls contribute significantly less to subsistence activity than juveniles in other cultures, despite having the skills and strength required to increase their subsistence effort. However, the authors suggest that the low levels of activity may be a strategy that conserves energy in order to support rapid juvenile skeletal growth during an early age at first reproduction.

Similar trade-offs have been documented for age at first pregnancy. In rural Gambia, researchers found that women who were younger at first parturition were significantly shorter than other women (Allal et al., 2004; Sear et al., 2004). Additional epidemiological studies have also shown that pregnancy during adolescence impairs the ability to gain in height (Gigante et al., 2006; Scholl and Hediger, 1993). While bone growth in length stops around the time of menarche, bone continues to gain mass and grow appositionally until peak bone mass is achieved between 25-35 years of age.

Pregnancy and lactation represent stages of elevated energetic costs and thus, life history trade-offs between energy use, additional key resources such as calcium, and other aspects of maternal physiology are expected. The mineralization of the fetal skeleton and continued infant skeletal growth requires adjustments to maternal calcium metabolism during pregnancy and lactation, respectively. Both fetal and neonatal calcium

and bone metabolism are uniquely adapted to satisfy the specific needs of these developmental periods. The fetus must actively transport sufficient calcium across the placenta to meet the large demands of the rapidly mineralizing skeleton, whereas the neonate must quickly adjust to the loss of calcium transport through the placenta, while continuing to undergo rapid skeletal growth (Kovacs and Kronenberg, 1997).

Following the same logic as the maternal depletion syndrome, which specifically focuses on fat reserves, if maternal calcium stores continue to be depleted with each successive pregnancy and lactation cycle, maternal well-being may be compromised, although it remains unclear the extent to which the fetus/infant may be prioritized at the expense of the mother. The transient and long-term trade-offs of reproduction are highly debated and there is currently no consensus on exactly how reproductive ecology shapes bone health (see Chapter II). This lack of consensus may be largely attributed to the following factors. First, reproductive biology and bone density are impacted by multiple, complex pathways. For example, pregnancy can both decrease maternal bone density through heightened calcium demand associated with fetal skeletal growth, and increase maternal bone density through increased body weight and elevated estrogen levels (Allali et al., 2007). Therefore, establishing causality between specific characteristics of pregnancy and lactation with bone density becomes challenging. Second, there may be threshold effects, such that after a certain number of births the effects of pregnancy and/or lactation shift (Cerroni et al., 2003). Finally, patterns of lactation are extremely variable across populations and are dependent on a variety of environmental and social conditions that may either permit or inhibit extended breastfeeding periods. Because of the heterogeneity of lactation behaviors, the effects of breastfeeding on bone density may

not be experienced uniformly across populations. Therefore, the relationship between reproductive factors and bone density appear to be inherently population-specific and as such, guidelines for preventing bone fragility and loss may not be applied similarly to different populations across ecological and geographical space.

Few studies to date have examined maternal bone status in non-Western, non-clinical populations and even fewer data are available for subsistence-based, natural fertility populations. This is a critical oversight given that reproductive patterns and hormonal cycles of women living in non-industrialized populations are dramatically different from women in industrialized nations. Women in non-industrialized populations typically experience relatively late menarche followed by early first birth, approximately 3-4 years of lactating, and early menopause (Sperling and Beyene, 1997; Weaver, 1998). The estimated average lifetime number of menstrual cycles in non-industrialized groups is approximately 50-100 cycles. In contrast, over the reproductive lifetime industrialized Western females typically experience early age at menarche, limited breast-feeding periods, and an estimated 420 menstrual cycles (Eaton et al. 1994; Strassman, 1997). The significantly greater number of cycles in the lifetime of a typical Western woman, accompanied by shifts in hormonal levels, undoubtedly has an effect on bone integrity that differs from typical non-Western women. Because contemporary subsistence populations have reproductive patterns more characteristic of most of our evolutionary past than those of Western groups, studies in these populations can improve our understanding of the conditions under which the life history trade-offs in skeletal metabolism and reproduction evolved. Furthermore, research in subsistence-based groups may allow insight into the complexities of bone loss without the confounding

effects of sedentary lifestyles and Westernized diets characteristic of industrialized groups. For these reasons, research was conducted among the Indigenous Shuar forager-horticulturalists of Amazonian Ecuador. Additional comparative data were collected on rural, agrarian non-Indigenous *Colonos* of the same region.

THE SHUAR OF ECUADOR

The Shuar¹ are an indigenous population who live in the southern Oriente (eastern) neo-tropical forest of Ecuador and the northeastern region of Peru (Figures 1.1 and 1.2). Traditionally, Shuar lived in scattered households across the Paute and Upano River Valley between the eastern Andean foothills and the Cordillera de Cutucu, a steep mountain range that forms a barrier to the Yaupi and Morona drainages to the east. In the 1890s, Shuar began expanding eastward into the territory of the neighboring Achuar so that Shuar now live on both sides of the Cutucu, and throughout the Upano River Valley from the Peruvian border past the Pastaza River and north almost to the Rio Napo (Harner, 1984; Rubenstein, 2001; Stirling, 1938). Recent population estimates for Ecuadorian Shuar vary from 50,000 to 110,000, depending on the source; however, census estimates for indigenous groups are notoriously inaccurate, often underestimating the actual size of the population. Currently, Ecuadorian Shuar are distributed over 668 communities and primarily reside in the Morona-Santiago and Zamora provinces of Ecuador (CODENPE, 2011).

¹ *Shuar* means “people”. The Shuar may be more precisely referred to as Untsuri Shuar, which specifies this particular group as opposed to indigenous people in general or non-indigenous *apaci*.

As part of the Jívaroan² ethno-linguistic group, Shuar are culturally similar to other Jívaroan speaking peoples including Aguaruna, Huambisa, and Mayna of Peru, and share many cultural features with the Achuar further to the East (Descola, 1994, 1996; Harner, 1984; Hendricks, 1993) and Shiwiar to the Northeast (Sugiyama and Chacon, 2000; Sugiyama, 2004). Other than some minor linguistic differences, extent of polygyny, and slight material differences associated with population density and local ecology (e.g., navigability of rivers, size of houses, etc.), little separate Shuar, Achuar, and Shiwiar in terms of cultural variation.

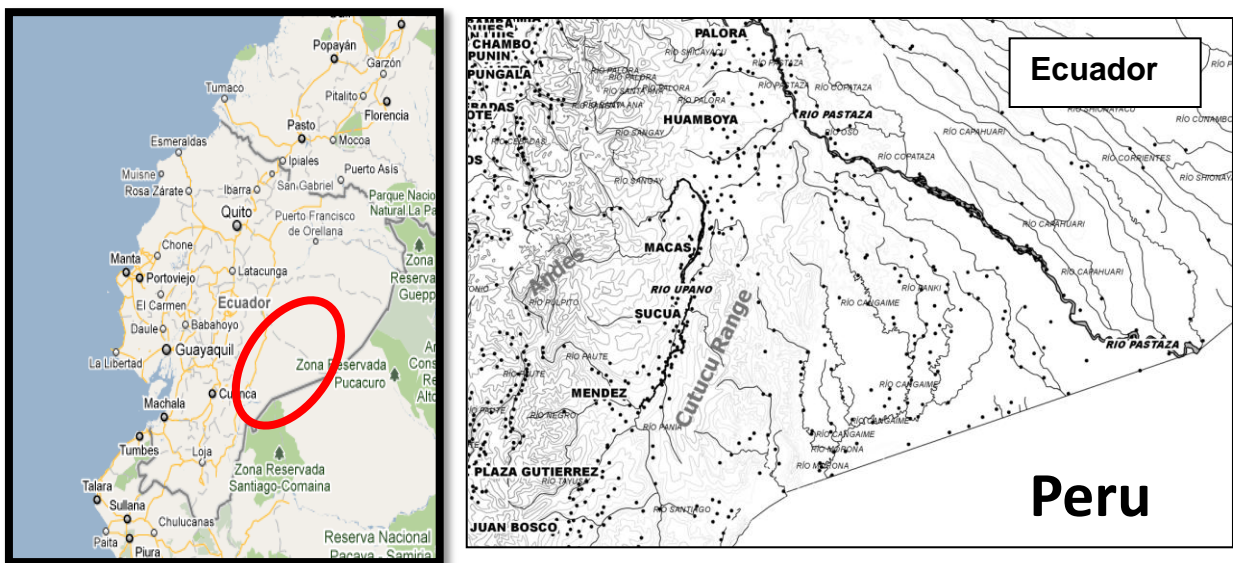


Figure 1.1. (left) Map of Ecuador with red oval indicating Ecuadorian Shuar territory and region of research. (Map Source: Google Maps)

Figure 1.2. (right) Map of Morona Santiago region. The map is based on data from the *Sistema Integrado de Indicadores Sociales del Ecuador* released by the Secretaria Técnica del Frente Social of Ecuador.

² *Jívaro*, in Ecuadorian Spanish, literally means “fierce”, “rebellious” or “savage” people. Until recently, Shuar were referred to by this name by Spaniards and foreign ethnographers but because of its derogatory implications, the term is not considered polite and is often replaced with other monikers.

Traditional Shuar Subsistence and Current Lifeways

Traditional³ Shuar subsistence was based on blowgun and spear hunting, fishing, and swidden (slash and burn) horticulture (Harner, 1984; Descola, 1994; Descola, 1996; Stirling 1938). In the interior Trans-Cutucu region (east of the Cutucu where road access ends), most Shuar continue to subsist primarily by hunting, fishing, and horticulture. Each Shuar household has either a single large garden (*finca* in Spanish; *aja* in Shuar), but usually two or three at different stages of production with intercropping and staggered planting seasons. These serve as the primary source of carbohydrates, which provide approximately 65% of dietary calories (Harner, 1984). Staple foods include tubers such as manioc (*yucca*; *mama*)⁴, sweet potato (*camote*; *inchi*), taro roots (*papa china*; *kenke*), lime zinger (*pelma*; *sanku*), and peanuts (*mani*; *nuse*). Shuar also grow non-tuberous food plants such as a variety of plantains (*plantanos*; *paantam*), a primary staple food, a variety of bananas (*bananas*; *páantam*), and maize (*mais*; *sháa*). Additional crops include oranges (*naranjillas*; *kukuch*), squash (*calabasa*; *yuwi*), onions (*sepui*; *cebollas*), papaya (*papaya*; *wapái*), sugar cane (*caña*; *paat*), achiote⁵ (*achiote*; *ipiáku*), chiles (*chiles*; *jimia*) and non-edible plants including *barbasco* roots and fish poisons (*timiu*), *hayahuasca* (a hallucinogen) and a variety of medicinal plants for healing. These crops continue to be grown in most Shuar villages, with manioc and plantains serving as primary dietary staples.

³ The term “traditional” is used here for convenience in referring to the basic pattern that can be seen consistently through sources beginning with Spanish chronicles in the 16th century and as recognized by Shuar.

⁴ Plant names are presented first in English followed by the Spanish name and when available, Shuar name.

⁵ Achiote is a fruit that is harvested for its subtle flavor and colorful red hue. It is often used as a food additive.

Fish and hunted game were traditionally the main Shuar protein sources, and continue to be staples in the Trans-Cutucu region. Fishing is done either with hooks and line or with fish poisons, baskets, spears, or by hand. In the rainy season, the bulk of fish are caught by hooks and line. During the transition to the dry season, emphasis on fishing gradually increases as the rivers become shallow and fishing with one of two cultivated poisons, *timiú* or *masu*, becomes increasingly efficient. In productive areas, these dry-season fish poisonings can be communal, village-wide events and can produce a large surplus (Figure 1.3). Fishing continues in the Upano Valley as well as in the Trans-Cutucu region. Ethnographic observations, interviews, and unpublished food frequency data suggest that fishing remains productive and an extremely important part of the subsistence regime in the Trans-Cutucu area. However, informants report declines in fishing returns within the Upano Valley to such an extent that fishing no longer provides a dietary staple, but rather a periodic supplement to the diet for some but not all households.

In many regions, hunted game also serves as an important source of protein. In a recent study focused on the sustainability of hunting in multiple Shuar communities in



Figure 1.3. Shuar women in the Upano Valley prepare barbasco root by pounding it with rocks to release the poison. The roots will be soaked in a shallow river bed and the fish will be sedated long enough to be caught by hand.

the Trans-Cutucu region, Zapata-Rios et al. (2009) recorded the following prey⁶: red brocket deer (*Mazama americana*; *penke hapa*), common woolly monkey (*Lagothrix lagotricha*; *Chuu*), tapir (*Tapirus terrestris*; *pama*), paca (*Agouti paca*; *kashai*) and nine-banded armadillo (*Dasypus novemcinctus*; *shushui*). Collared peccary (*Pecari tajacu*; *yankipik*) was the most important game species in terms of kilograms (kg) of meat taken. Other hunted game included howler monkey (*Alouatta sp.*; *yakump*), capuchin (*Cebus sp.*; *tsere*), coati (*Nasua nasua*; *kushi*), and Northern Amazonian red squirrel (*Sciurus igniventris*). These hunting data broadly correspond to data from Shiwiar living in more traditional communities (Sugiyama and Chacon 2000; Sugiyama 2004; Sugiyama personal communication, 2011), suggesting that hunting remains an important part of the subsistence regime. In addition to hunted game, several types of palm grubs and insects serve as important sources of protein. While at this time no direct observational data on hunting productivity are available for the Upano Valley region, Shuar informants report that good hunting can still be had by walking 6 hours or more into the Cordillera de Cutucu, but that hunting near the communities is unproductive. In the Upano Valley, chicken serves as the primary substitute for hunted game, but beef, pork and, to a lesser extent, guinea pig (*cuy*) are also consumed by some families (Figure 1.4). Meat is typically served either roasted or boiled. Fish and other meats may also be prepared in *ayampacos*, a traditional Shuar food stuffed with meat, palm hearts (*palmito*), chard (*acelga*), onions, and garlic in a banana leaf (*bijahua*) and roasted over a fire. *Ayampacos*

⁶ For ease of identification, primary prey species are listed here in the following way: common name (scientific name; Shuar name).

may also be prepared with animal intestines, bull testicles, or, in more acculturated areas, with a cheese filling.

Currently in the Upano Valley, many families also participate in small scale animal-husbandry, and cattle is typically raised and sold as money is needed (Figure 1.5). Timber and a variety of products from small scale agricultural production including plantains, papaya, and oranges are also sold.

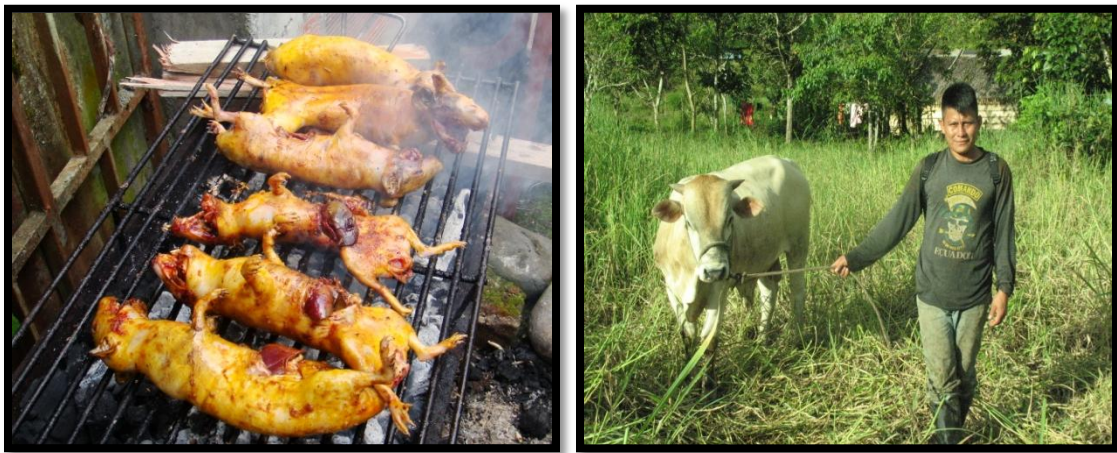


Figure 1.4. (*left*) Guinea pigs (*cuys*) roasting over a grill. Although not a traditional Shuar delicacy, many Upano Valley Shuar now raise guinea pigs for personal consumption and for sale. (Photo Credit: Liebert 2009)

Figure 1.5. (*right*) Shuar man from the Upano Valley showing off his cow. (Photo Credit: Sugiyama 2009)

Gender Relations and Sexual Division of Labor

Shuar gender relations have been subject to debate, with some authors describing a highly male dominated society (Seymour-Smith, 1988; Taylor, 1981), while others describe male and female gender roles and domains of influence that are complimentary

and complex (Descola, 1996; Karsten, 1935; Mader, 1999; Mader and Gippelhauser, 2000). What remains consistent throughout the literature is the complexity of gender roles and relations; this has been attributed to the fact that different aspects of gender relations are contextually-dependent, as well as complex and fluid (Mader, 1999).

The nuances of gender relations are beyond the scope of this review, but these patterns provide some insight into Shuar life, women's and men's work roles, what these activities typically entail, and the flexibility of these roles. For instance, the Shuar house is divided into male and female sections. The male domain includes the public space where visitors are greeted and seated, whereas the female domain constitutes the sleeping areas and cooking spaces. Male visitors typically do not have access to the woman's side and cooking areas unless they are close family or friends. In addition to gendered spaces, clear divisions of labor exist between men and women both on ideological and physical levels. A discussion of one set of ideal Shuar values helps put this in perspective. *Penker pujustin* or *shiir waras*, mean "good life" or "being," and includes gender roles and norms of behavior, such that to live a good life is to engage in practices expected of one's age and sex, and to engage in them with vigor. For men, this includes tasks such as hunting, fishing, clearing gardens, and hauling logs for firewood. It includes protecting and speaking strongly (*kakaram chicham*) in favor of the interests of oneself and one's family, being good to one's wife and children, and avenging transgressions against oneself or one's kin group. It also incorporates clear thinking, fearlessness and independence, and a refusal to submit to anyone. For women, *penker pujustin* involves being a hard worker and maintaining large, well-weeded, productive gardens, and harvesting the produce. Equally as important is making and serving good quantities of

manioc beer (*chicha*; *nihamanch*), caring well for one's family, preparing and serving food, and maintaining a clean house. Ideally, it also includes thinking clearly and speaking strongly in support of one's interests.

The Shuar concept of *kakaram* is a useful way to conceptualize how one should participate in daily activities. The term simultaneously refers to the personal and/or political power acquired through and associated with various spiritual qualities (Mader, 1999). *Kakaram* (literally: powerful ones) is used both to refer to men who are killers (i.e., mercenaries in warfare), as well as to any individual—man or woman—who speaks powerfully to advance or protect their interests, does not complain about hardship, and is recognized as a hard worker. As Sugiyama (2004) points out for Shiwiari (who hold similar values), the link between the values of *penker pujustin* and *kakaram* and everyday life has a logical connection to traditional Shuar subsistence economy and settlement pattern. Relatively low levels of traditional day-to-day foraging risk, along with large productive gardens meant that there was relatively high degree of household autonomy within a scattered, low-density pattern of settlement. Thus, it is unsurprising that household autonomy in production is closely linked with emphasis on hard work, individuality, and independence.

In actuality, men and women occasionally cross into other gender role activities as situations necessitate. With gardening, men are responsible for clearing forest trees (slashing) and some planting, as well as periodically helping with weeding or harvesting. They are also responsible for recruiting people for communal work parties (*mingas*). Women may help clear underbrush but bear primary responsibility for most planting, cultivation, weeding, and harvest activities. However, men with dependents but without a

current mate (e.g., widower) will engage in gardening tasks typically reserved for females. Conversely, a widow, for example, will organize a *minga*, although usually by recruiting a male relative to make the invitations.

As noted earlier, women are typically responsible for the cultivation of manioc as well as other tuber plants. Manioc may be consumed boiled or in the form of *chicha* (in Shuar: *nihamanchi*), a locally fermented beer that is made by boiling the tuber and mashing it to a soft consistency. A Shuar woman will simultaneously pound the boiled manioc into a mash, and chew handfuls of the softened manioc that she then spits back into the pot (Figure 1.6); an enzyme in the saliva ensures rapid fermentation of the manioc beer over the course of one to five days. While there is some fluidity in the gender roles of production, only Shuar women make *chicha* (Sugiyama personal communication, 2011). Drinking *chicha* is considered a central aspect of being Shuar and traditionally adult males can consume a daily average of 3-4 gallons while adult females may consume 1-2 gallons (Harner, 1984).



Figure 1.6. Upano Valley Shuar woman preparing *chicha*. (Photo Credit: Lim 2009)

When hunting is available, Shuar men embark on trips, sometimes for weeks at a time. Today, these hunting trips rarely involve women but in the past, men would often be accompanied by their youngest wives for companionship (Harner, 1984). Hunting dogs were often owned by women, so wives also hunted by pursuing small and medium-sized terrestrial game with dogs. Having sexual relations on the hunt is perceived as dangerous, since the Shuar believe that after intercourse the man will be more susceptible to a poisonous snake bite; however, in practice, this belief does little to curtail sexual activity during hunting trips (Harner, 1984).

In the past, a man's role in the system of production was not only to clear gardens and hunt, but also to present the casualties of war in a *tsantsa* ritual, the process of head shrinking. Although rarely practiced today, the *tsantsa* ceremony was the ultimate expression of male power (Rubinstein, 2002). During warfare, the enemy's head would be cut off and taken to be incorporated in the *tsantsa* ceremony. The process of the *tsantsa* ceremony involves removing the skin from the skull, and pouring warm sand in the skin to induce it to shrink. A *tsantsa* ceremony could last many hours as a result of the meticulous nature of this process in order to ensure that the avenging soul (*musiak*) of the dead person would not emerge and cause mischief. Further, the killer gained power from performance of the ritual, a power that ebbed with time and necessitated further *tsantsa* to reestablish. Although sometimes described as trophies of war that must be saved, *tsansta* were not displayed and were traditionally of no further use once the *tsantsa* ritual was performed (Harner, 1984; Rubenstein, 2002).

Socio-Political Organization

Egalitarian, polygynous, matrilocal, and without explicit lineage or clan structure, Shuar household clusters were traditionally centered around a man and his wives. Families typically lived in separated clusters of households that may also include daughters, unmarried sons, and sons-in-law (Rubinstein, 2002). A man's accomplishments, his courage as a warrior, hunting prowess, and respectability determined how many wives he could attain. Today, fewer marriages are polygynous, particularly among Upano River Valley populations, but older warriors, shamans (*uwishin*), or other highly productive men may still have more than one wife. The Shuar family unit is traditionally large, comprised of a male and one or more female mating pairs, and an average of 5-6 offspring. Because of this large family composition, allomothering is common practice and may involve older non-reproductive aged siblings and extended family members (e.g., grandmothers and maternal sisters).

The household is the primary economic unit, and traditionally, household clusters were separated by a distance of 2-3 kilometers. Similar to other Amazonian populations, Shuar power structure was traditionally decentralized with no formal political or social stratification. Authority was specific to achievements, and both shamans and powerful senior men (*juunt*) or warriors were respected for their knowledge and personal power. While inter-household cooperation remains important in several spheres (e.g., warfare, aid in health crises, healing, fish poisoning, forest clearing, and house construction), the daily household economy is largely independent (Harner, 1984). In general, even in the Upano Valley, Shuar remain exceedingly individualistic in outlook and so consensus is hard to achieve. This character, coupled with the formation of more concentrated

communities in the 1960s (see below: Shuar Federation), means that intra-village conflict is common and sometimes results in violence. Even with recent changes in overt political structure, including elected *Sindicos* (Spanish word for community union president), vice *Sindicos* (vice president), secretary and treasurer, these positions are largely ceremonial and have little actual authority or power.

CHANGING SHUAR LIFEWAYS

Market integration (MI) among the Shuar has, and continues to be, shaped by historical, political, economic, and social processes, as well as “traditional”⁷ cultural dynamics, including ideologies of power, shamanism, land rights, and politics (Mader 1999; Rubenstein 2001). Among the Shuar, MI is an ongoing process that is inherently intertwined with colonization. Recent clashes and high-profile conflicts between Shuar and the Ecuadorian government highlight an ongoing colonization that continues to threaten and encroach on Shuar land and alter cultural lifeways. In particular, government policies and deals with mining and oil companies since the mid 20th century have threatened traditional culture and well-being, although these relations have stimulated the establishment of several strong unified fronts that seek to preserve indigenous identity (e.g. Shuar Federation, CONAIE).

⁷ For convenience, and because Shuar employ similar terms to refer to this system, “traditional” is used to refer to Shuar life prior to significant MI and acculturation, yet recognize the complex and problematic issues that arise with the term in the larger anthropological literature.

Historical and Present-Day Colonization

In the 16th century, Spaniards defeated the Inca and were free to explore the Andean foothills in search of gold. Although trade networks between Shuar and Spaniards were established during this time, this relationship largely ended in 1599 with “the Jívaro Revolt,” when Shuar rose up to expel the Spanish in response to increasingly onerous Spanish taxation (Rubinstein, 2002). Not until the 1890s were peaceful trade relations reestablished with non-Shuar populations in the region (referred to locally as *Colonos*) prompting increased non-Shuar migration into Shuar territory. Over the next fifty years Upano Valley Shuar traded the machetes, axes, firearms, and other trade-goods they acquired with Shuar living further to the east.

In the 1950s, the collapse of the Panama hat industry in the Ecuadorian highlands altered the dynamics of these trade relations by introducing a new wave of non-Shuar migration into Shuar territory. A once lucrative export, the decline of the Ecuadorian Panama hat industry created a deficit in the economic well-being of the Ecuadorian workers who produced these hats. In an effort to incite economic development through increased access to lands and thus, agriculture, the Ecuadorian government promoted migration in and around Shuar territory by non-Shuar settlers. This marked one of the first government initiatives to appropriate Shuar land by encouraging movement into the region by non-Shuar Ecuadorians. The legacy of this move continues until today with conflict between Upano Valley Shuar and *Colonos* for access to land.

Within a few years, the discovery of oil in the Amazon in the early 1960s led to the creation of the government institution called the *Centro de Reconversión Económica del Azuay, Cañar y Morona-Santiago* (CREA) in order to establish the infrastructure for

massive colonization of Shuar land (Borrini-Feyerabend et al., 2005). The creation of CREA was integral in stimulating the formation of a unified Shuar body, which was established in 1964. In an effort to preserve the traditional lifeways and culture that were being threatened, many Shuar abandoned the Upano Valley and settled in the Trans-Cutucu region. Others aligned themselves with missionaries, converted to Catholicism, and adopted cattle raising in combination with the indigenous swidden horticulture practices (Borrini-Feyerabend et al., 2005). The Shuar Federation, or *Federación Interprovincial de Centros Shuar* (FISCH), was founded with missionary support, with the goal of defending indigenous land rights against agrarian reforms, ensuring the development of Shuar communities, and preserving indigenous cultural and ethnic identities (Borrini-Feyerabend et al., 2005). The federation also advocates for the registration of Shuar settlements as legally recognized cooperatives (*Centros*), the procurement of communal agricultural land titles, and the extension of bilingual education and modern health care. FISCH is directed by a board of officials who are elected every three years and who meet regularly to exchange information and coordinate strategies to defend their member's rights, which continue to be under the pressure of oil and mining companies (CODENPE, 2011).

Today, FISCH is comprised of approximately 490 *Centros* primarily from the Upano Valley region⁸. Communication between distant federation-affiliated communities has been increased since the early 1990s with the establishment of Shuar Radio, *Radio ARUTAM*. In 2004, the Shuar were one of four indigenous populations to present a

⁸ The *Federación Independiente del Pueblo Shuar* (FIPSE) is an organization formed by Trans-cutucu communities and works closely with FISCH.

lawsuit against the Texaco/Chevron oil company for reparations resulting from the destruction of Shuar ancestral land (Minority Rights Group International, 2008). The oil company is accused of intentionally dumping billions of gallons of toxic waste into the rainforest which has resulted in the decimation of indigenous groups by cancer and other oil-related health problems. Court decisions for this lawsuit regarding judgment payments are still pending. More recently, in September 2009, indigenous leaders including then FISCH president Pepe Acacho, organized protests against the Water Act, which would privatize water sources and allow transnational mining companies free reign over water supplies throughout Ecuador. The proposed act is especially threatening to indigenous communities who rely on local water sources for drinking and cooking water and fish. The Ecuadorian government under President Rafael Correa accused the protestors of terrorism and sabotage and, in early 2011, Acacho and others were arrested (Ordoñez, 2011). Among leaders of the Confederación de Nacionalidades Indígenas del Ecuador (CONAIE) and the indigenous Pachakutik political party, the arrests were seen as a political persecution by the Correa regime to silence the Shuar resistance and to further exploitation of mining and oil. Shuar responded through peaceful protests leading to the eventual release of Acacho who reportedly said, “We Shuar men are warriors. We are not easy prey. They can’t mess around with us” (Caselli, 2011). As of May 2011, this conflict has not been resolved. Shuar and other indigenous groups continue to resist the actions of the Correa government in an effort to preserve and protect their ancestral land. On May 6, 2011, Acacho was elected and formally inducted to the office of Vice President of CONAIE, largely based on his steadfast resistance to the actions of the Correa regime.

Market Integration across Shuar Territory: Basic Characteristics

Since the 1950s, non-Shuar migration into Shuar territory, specifically in the Upano Valley, has increasingly exposed Shuar to lifestyles and goods associated with MI. Although subsistence activities remain common throughout much of Shuar territory, lifeways are changing most rapidly in the Upano Valley, where a partially paved road runs the length of the valley and has facilitated access to nearby market centers, such as Sucua and Macas. Completion in 2009 of the paved road between Sucua and Puyo to the North further accelerated change in the area. Over the last decade, other infrastructural developments have become available to Upano Valley communities, including expansion of a dirt road system to smaller communities, access to the electrical grid, and development of a cellular phone network. Housing has shifted from a mix of traditional thatch roof dirt floor housing with some milled lumber houses as late as 1998, to almost entirely wood houses by 2005, to cinder block housing provided through a government sponsored development project in 2010.

Many Upano Valley Shuar villages have primary schools that are attended by most children. For most families, sending children to school is costly as they are required to purchase uniforms, shoes, backpacks, and school supplies. This can become a deterrent for some families and often, children do not pursue education beyond primary school. Fewer adolescents attend secondary school but the numbers are increasing—at present, approximately 11% of females and 31% of males from 25-34 years old report having attended secondary school (McSweeney and Jokisch, 2006). Most schooling is conducted in Spanish, with Spanish slowly becoming the primary language for many Shuar families.

Of 1,760 households surveyed across Shuar and Achuar territory in 2005, 52% spoke only Shuar, 28% were bilingual in Spanish and Shuar, and 20% only Spanish (McSweeney and Jokisch, 2006). The number of families who speak Shuar is almost certainly declining as more parents increasingly forego Shuar for Spanish when speaking to their children. My own interviews with Shuar suggest that at least in the Upano Valley, only among the oldest demographic is Shuar the exclusive language, while the younger generations are losing the ability to communicate in Shuar. A similar trend is reported by informants in the Trans-Cutucu region as well (Mankash, personal communication, 2011).

The degree of economic development and MI in the Upano Valley contrasts markedly with that across the Cutucu range. In this area, Shuar villages lack access to the electrical grid and there is no telephone access of any kind. A few families own generators but this is typically limited to health workers or teachers who have external government income. Housing is primarily traditional, with some wood structures. Travel to main market centers involves a motor canoe trip ranging from 1-16 hours, followed by at least an 8-9 hour bus ride almost entirely along dirt roads. With regard to schooling, little information is available regarding education in this region, although in most communities primary schools are available for Shuar children.

The level of MI is particularly accelerated among Shuar who live in larger regional centers, such as Sucua or Macas, and to a lesser extent, in communities located closest to the Upano Valley main road. In these areas, a significant portion of Shuar participate in wage labor (e.g., nurses, bakery workers, or waiters), have children who attend high school, own imported goods (e.g., televisions, DVD players, computers), and

engage in non-traditional celebrations such as quinceañeras, the birthday celebration and rite of passage for girls turning 15 years old similar to “Sweet Sixteens” in the US. E-mail and, more recently, Facebook, are becoming increasingly the preferred means of communication between friends and family living in Ecuador and abroad. Local restaurants that serve non-local cuisine (e.g., Chinese and Italian) create an abundance of food options for local Shuar and *Colonos* living in the largest towns in the region.

Because economic development and lifestyle change is not occurring uniformly between communities and among households within communities, an enormous spectrum of social, economic, environmental, and health effects related to MI are observable.

Effects of Market Integration on Health among Indigenous Groups

Increasing MI among indigenous populations can have profound social, economic effects on health, yet these are not uniform across populations (Fleming-Moran et al., 1991; Hawkes et al., 1997; Henrich, 1997; Huss-Ashmore et al., 1992; Gross et al., 1979; Godoy, 2000; Godoy and Cardenas, 2000; Shephard and Rode, 1996; Snodgrass et al., 2007). For those with poor nutrition and high infectious disease burden, increased MI can bring education, economic opportunities, better diet, and access to healthcare (Leonard et al., 1993; Santos and Coimbra, 1991; Strauss and Thomas, 1988; von Braun and Kennedy, 1994). However, it can also bring with it a rise in chronic conditions such as cardiovascular disease and type 2 diabetes, (e.g., Lindgärde et al., 2004; Pavan et al., 1999; Popkin, 2004). Limited research on market integration and health among the Shuar and other indigenous Amazonian groups (e.g., the Tsimane of Bolivia) demonstrate generally similar trends (Pavan et al., 1999; Lindegärde et al., 2004; Lu, 2007).

The mechanisms responsible for this health transition among increasingly market integrated populations remain incompletely understood, although factors usually implicated include physical inactivity, dietary changes, alcohol consumption, tobacco use, and chronic psychosocial stress (Durnin, 1990; Shephard and Rode, 1996; WHO/FAO, 2003). Declines in total energy expenditure (EE) may occur as a result of decreased participation in energy-intensive subsistence activities, such as hunting and hay cutting (Singh et al. 1989; Yao et al. 2002; Tataranni et al. 2003; Snodgrass et al., 2006). In addition to drops in EE, substantial dietary changes also typically occur including an overall increase in energy intake, heightened stability in food availability, and an increase in the consumption of refined carbohydrates, saturated fats, and alcohol (Shephard and Rode, 1996; WHO/FAO, 2003; Snodgrass et al., 2007). The process of MI may also present new sources of social status that diverge from indigenous systems and are more aligned with Western views. These status cues are often tied to ownership of Western commodities and, among indigenous populations, may introduce novel stressors due to the mismatch between aspirations and the resources necessary to obtain those goods (Dressler, 1991; McDade, 2001, 2002). This incongruence appears to lead to chronic psychosocial stress, which has effects on immune function, blood pressure regulation, and patterns of fat distribution (Chin-Hong and McGarvey, 1996; James and Brown, 1997; Sorenson et al., 2005). Among the Shuar, individuals in more market-dependent communities have poorer measures of insulin, body fat, and leptin than those in traditionally living communities (Lindgärde et al., 2004). MI may also have important effects on children. Blackwell and colleagues (2009) found that Shuar children have

higher prevalence of stunting than closely related but more traditional Shiwiar forager-horticulturalists.

Of course, neither the pace nor type of market changes, nor the local social, ecological or economic conditions are uniform across societies. Thus, neither the decisions or behaviors of those experiencing these changes nor their ultimate health outcomes should be uniform. Some peoples' diets and health may improve with MI, while others may decline, and this variation may occur even within a single community (Leatherman, 1994; Leonard et al., 1993). Such differences are likely the consequence of complex interactions between particular social and economic changes, and the social, economic, cultural, and individual circumstances in which these effects take place (Snodgrass et al., 2007).

SHUAR HEALTH AND LIFE HISTORY PROJECT

The studies presented in this dissertation are conducted as part of the Shuar Health and Life History Project (<http://www.bonesandbehavior.org/shuar>). This project is an ongoing, multi-year collaborative endeavor that integrates quantitative and qualitative techniques in order to examine life history trade-offs along a number of different dimensions of health, subsistence, economy, and demography. The project is co-directed by Drs. Lawrence Sugiyama and Josh Snodgrass, and is based out of the Department of Anthropology at the University of Oregon. Since 2005, our team has forged close relationships with several international collaborators including the *Federación Interprovincial de Centros Shuar* (FISCH) and *Instituto Ecuatoriano de Seguridad Social* (IESS).

ORGANIZATION OF THE DISSERTATION

This dissertation is organized into six chapters that examine Shuar energetics and reproduction with specific focus on reproductive effects on skeletal health and physical activity. Chapter II presents a review of the literature on bone anatomy and physiology as well as a discussion on the major determinants of bone mass. This chapter also includes a discussion on the current available techniques of assessing bone health and concludes by considering bone loss within an evolutionary framework.

Chapter III presents a normative bone health profile from bone density data collected for the Shuar and non-indigenous *Colono* population. *Colonos*, an unrelated group who live in the same geographical region as the Shuar, were included in the analysis for comparative purposes. Minimal data are available on bone health from populations living in developing countries, and no normative datasets are available for either sex in the populations addressed. Age-related declines in bone density were observed for both sexes of each population. However, the results demonstrate that Shuar bone density, particularly among males, is extremely high, when compared with *Colonos* and other global reference populations. Chapter III makes a compelling case for the need for more bone health data from similar subsistence-based populations, and calls for a reconsideration of what constitutes “normal” bone health references in Western countries. This chapter has been accepted for publication in *Archives of Osteoporosis*, and is co-authored with Josh Snodgrass, Melissa Liebert, Tara Cepon, Aaron Blackwell, and Lawrence Sugiyama.

Chapter IV presents unique research that investigates various hypotheses concerning reproductive variables and their influence on bone density. Reproductive-

related trade-offs in bone health are expected among women, who must rely on skeletal calcium stores to meet the energetic requirements of the growing fetal and neonatal skeleton during pregnancy and lactation, respectively. Few studies have explored the relationship between reproduction and skeletal health in a subsistence-based group from a non-clinical, non-Western field setting. Because Western lifestyles and fertility patterns are vastly different from those characteristic of our evolutionary past, our current understanding of the mechanisms of reproductive-related bone loss among females are obscure. Although cross-sectional rather than longitudinal in design, the results indicate the importance of early environmental and energetic conditions as well as the timing of developmental stages (e.g., age at menarche) for establishing bone status in advanced life. Chapter IV also demonstrates the utility of examining bone health within a life-course perspective in order to better conceptualize the complex interactions of developmental stages and their role in determining health outcomes later in life. This chapter presents unpublished co-authored material; it has been offered publication in the *American Journal of Human Biology* and will be submitted for review in the upcoming months.

Following Chapter IV's discussion of life-history trade-offs and bone health, Chapter V examines reproductive trade-offs and physical activity in one rural Shuar community. Studies with subsistence populations have reported that various bio-behavioral strategies are adopted by pregnant and lactating women as a means of compensating for their elevated metabolic costs. Reductions in energy expenditure, increased caloric intake, or a combination of the strategies are among some of the strategies available. These strategies are clearly dependent on the available energy and energetic resources from the surrounding environment. Results from this cross-sectional

study show that when compared to non-pregnant, non-lactating women, pregnant and lactating participants do not significantly alter their activity output. However, males with pregnant and lactating wives increase their energetic output appreciably when compared to other males. This suggests that Shuar males alleviate the higher costs of their partners' reproductive state by increasing their contribution to subsistence activities. Chapter V concludes by discussing the applicability of accelerometer devices to record physical activity patterns for future research in human biology. This chapter has recently been published in the *American Journal of Human Biology*, and is co-authored with Josh Snodgrass, Aaron Blackwell, Melissa Liebert, and Lawrence Sugiyama.

Finally, Chapter VI synthesizes the results of the dissertation as a whole and presents general conclusions and future avenues of research.

BRIDGE TO CHAPTER II

Despite the enormity of the problem of osteoporosis on global populations, the mechanisms of bone loss are still incompletely understood. Before presenting results on skeletal health in the subsequent two chapters, Chapter II presents an overview of the literature on bone anatomy and physiology and the major factors that contribute to bone loss. This chapter includes a discussion of the common techniques used to measure skeletal health and the motivations for employing a calcaneal ultrasonometer for this dissertation. In the final section of Chapter II, the skeletal system and the process of calcium mobilization are considered within an evolutionary framework.

CHAPTER II

BONE: ANATOMY, PHYSIOLOGY, AND THE FACTORS CONTRIBUTING TO SKELETAL HEALTH

Bone is dynamic tissue that serves four main purposes: 1) to provide a framework for locomotion and attachment sites for muscles; 2) to protect vital internal organs; 3) to store calcium and other minerals; and 4) to produce blood cells critical to immune protection. A composite tissue comprised of both organic and inorganic constituents, bone is highly vascularized and metabolically active and thus adapts to changing external circumstances through processes that balance formation and resorption.

With advancing age, resorptive activities typically begin to outpace bone formation, which causes a loss in strength and elasticity. The eventual result is compromised bone health characterized by a decreased bone mineral density (BMD) that may lead to the sub-clinical condition, osteopenia. According to World Health Organization (WHO) criteria, osteopenia is defined as 1 to 2.5 standard deviations (SDs) below the mean for one's age group. With continued bone loss, this may develop into the more severe condition, osteoporosis, differentiated by a deterioration of bone micro-architecture, increased bone fragility, and heightened fracture risk. Osteoporosis is diagnosed at more than 2.5 SD below the mean for one's age group (WHO, 2003).

The mechanisms of bone loss are incompletely understood, which is surprising given the enormous health effects of osteoporosis. Known as the “silent epidemic,” osteoporosis is now identified as a major public health issue affecting populations worldwide (Bartl and Frisch, 2004; IOF, 2010). In the United States, an estimated 30 million people suffer from osteoporosis, with equally high prevalence in Europe and other developed countries (IOF, 2010). Of the 1.3 million fractures reported annually among US patients over 45 years old, 70% can be attributed to low bone density (Bartl and Frisch, 2004). The problem is more severe for women, with osteoporotic fractures affecting more women than heart attacks, strokes, and all female cancers combined.

Epidemiological and clinical literature suggests that osteoporosis is a “natural” or “normal” component of the aging process, and emphasizes that intervention has little effect on this process. However, the assertion that bone loss is universal may be premature since most relevant data are exclusively from industrialized countries, such as the US. Because of less interest and limited diagnostic resources, relatively few data on bone health are available for developing nations and in particular, for subsistence, natural fertility populations. These latter populations are particularly interesting because their lifestyles, diets, and fertility patterns are more characteristic of our evolutionary past. This critical gap in the literature has important consequences for how bone loss is approached in clinical and public health contexts.

The evolutionary medicine approach and its emphasis on comparative studies across human populations have yielded some surprising findings. Research on health and aging in non-industrial populations has provided empirical evidence that some age-related changes observed in industrial societies are not universally experienced. For

example, many traditionally-living groups and rural populations do not show the age-related increases in blood pressure commonly seen in industrialized countries (Waldron et al., 1982). Similarly, Gurven and colleagues (2009) documented that the chronic, moderately elevated levels of inflammation typically associated with arterial aging and cardiovascular disease in industrialized populations do not appear to have the same effect among traditionally-living indigenous groups such as the Tsimane forager-horticulturalists of Bolivia. In this case, potentially damaging effects of inflammation appear to be offset by an active lifestyle, optimal body mass, lean diets, and low serum lipid levels. Cross-cultural studies of aging are in their infancy and many chronic conditions associated with aging have not been fully investigated. One of the goals of this dissertation is to address whether BMD changes with age are universal, or simply an artifact of the industrial/post-industrial condition.

In order to better understand the mechanisms of bone loss and establish the fundamental basis for the following chapters of this dissertation, the current chapter presents an overview of bone anatomy and physiology, followed by an outline of the main factors associated with bone loss and low BMD. The chapter concludes with a consideration of bone loss within an evolutionary context.

ANATOMY OF BONE

Bone is a composite tissue comprised of organic and inorganic compounds that primarily include collagen fibers and crystalline salts. These crystalline salts, consisting of calcium and phosphate, are deposited into the organic matrix and combine to form hydroxyapatite crystals. Together, collagen fibers and calcium salts are responsible for

the tremendous strength and integrity of bone. While collagen fibers contribute to tensile strength, allowing bone to endure stretching forces, calcium salts have compressional strength to endure squeezing forces. If growing bone is similar to constructing a brick wall, the “cement” would be composed of collagen and the other matrix constituents, while calcium crystals and phosphate would be “bricks” (Bartl and Frisch, 2004).

Bone formation, mineralization, and resorption are directed by cells that interact with bone and contribute to its dynamic quality. Formation of bone begins with an increase in the number of bone cells and fibers beginning with stem cells that eventually develop into osteoprogenitor cells. Because of their role in initiating bone formation, osteoprogenitor cells are often utilized in bone grafts. Osteoprogenitor cells are multipotential cells that may differentiate into osteoblasts (bone building cells) or chondroblasts (cartilage producing cells) (Forwood, 2001). The cells that differentiate into osteoblasts are responsible for the synthesis and mineralization of bone during both initial bone formation and bone remodeling later in life (Cowin, 2001).

Osteoblasts, which cling to the periosteum (the outer membrane of bone) and also reside in the endosteum (inner membrane of bone) of the marrow cavity, secrete vesicles that carry crystalline calcium and phosphate ions (Guyton and Hall, 2011). This secretion is called an osteoid, a cartilage-like material that will combine with calcium salts and then mineralize (Forwood, 2001). As the osteoid forms, some osteoblasts get entrapped within the osteoid matrix; these trapped cells are bone maintaining cells called osteocytes. Receptors for various hormones, including parathyroid hormone (PTH) and 1, 25-dihydroxyvitamin D and sex hormones (e.g., estrogens) are contained in osteocytes. Housed within spaces called lacunae, the osteocytes produce an elaborate network of

cellular extensions that connect them to each other and to the surface of bone via tiny tunnels called canaliculi (Bartl and Frisch, 2004) (Figure 2.1). The exact function of the osteocyte is unclear but it appears to maintain the matrix of the bone immediately surrounding it by producing a chemical signal that instructs osteoblasts to expand in response to strain (Cowin, 2001). Also, the number, or density, of osteocytes is a primary determinant of bone mass for both cortical and trabecular bone (Guyton and Hall, 2011).

Acting in opposition to osteoblasts are osteoclasts, or “bone breakers,” a critical cell type that functions to resorb or destroy bone. Osteoclasts exist in small, concentrated clusters that are typically found on the surface of the marrow cavity, a hematopoietic cell membrane that consists of a “ruffled border” (Forwood, 2001). Here, in between the ruffled membrane and bone, osteoclasts produce secretions such as citric and lactic acid, that serve to dissolve the bone matrix. Osteoclastic resorption of bone usually occurs at sites of greatest compression and torsion (Stini, 1995).

The rates of osteoblastic formation and osteoclastic resorption are typically equal to each other, except in growing bones. During the process leading to bone accretion, osteoclasts will first resorb bone and then disappear. The bone resorbing cells are then replaced by osteoblasts, which lay down new bone in successive layers of concentric lamellae on the inner surfaces of the marrow cavity until the hollow space is filled. Deposition of new bone is terminated when this new tissue begins to impinge on the blood vessels supplying the area (Njeh et al., 1999). The Haversian canal, through which these vessels run, is the only remnant of the original cavity. Each new area of bone deposited in this fashion is called an osteon, or Haversian system, the basic unit of compact bone (see Figure 2.1).

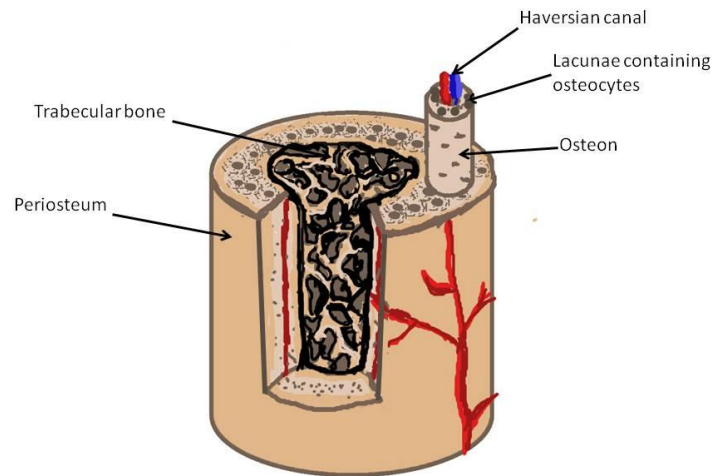


Figure 2.1. Trabecular bone and the Haversian system found in compact bone.
(Illustration by Tara J. Cepon)

Bone is continually turned over during life and also resorbed and deposited to adjust its strength to match the extent of bone stress. This process, in which the density of bone in a given locus is dependent upon the magnitude of the applied loads, is known as Wolff's law. When high levels of mechanical loading are experienced, new bone can be added or the existing bone may be redistributed to balance strains which alter diaphyseal size and strength. During the process of bone turnover, bone deposition and resorption replace old, brittle, and weak bone with new organic matrix. If the rate of bone resorption outpaces production, the total quantity of bone decreases and creates a negative "bone balance." In children, the rates of deposition and resorption are rapid, and bones exhibit minimal brittleness compared to those of older individuals, where rates of deposition and resorption are slow. The homeostasis of the skeletal system is an evolved mechanism that operates to prevent brittle bone and loss of tensile strength, both of which will inhibit at least two critical functions, support of locomotion and protection of vital internal organs.

Microscopic Constituents of Bone

At a histological level, two types of bone exist – immature (woven) bone and mature (lamellar) bone. The formation of new bone begins with a hyaline cartilage template, which is largely collagen-based and approximately 75% water (Frost, 1986). This structure is then replaced by unorganized woven bone, which is the initial osseous tissue produced by osteoblastic activity. Woven bone does not conform to mechanical load properties placed upon it but rather builds along the capillary paths that originally invaded the cartilaginous model at the onset of the bone formation process (Cowin, 2001). This initial type of ossification is found throughout fetal bone and in the epiphyseal plates of children. In adults, it exists at the subchondral articular cortex and at tendon attachment sites, between bone and calcified cartilage (Njeh et al., 1999). Woven bone is also the initial bone laid down at fracture sites during the early stages of remodeling.

Over time, woven bone is replaced by more organized mature bone, which is distinguished by its finer collagen bundles that give bone its greater mechanical strength. Additionally, the collagen bundles are housed within circumferential lamellae and run in opposite directions in alternating layers, which contributes to the bone's ability to resist torsional forces.

Macroscopic Constituents of Bone

Mature bone consists of two distinct architectural types—cortical bone and trabecular bone (Figure 2.2). Cortical bone is significantly denser and stronger than the trabecular variety, and forms the outer layer of bone and the shafts of long bones. In

contrast, trabecular bone has a weaker constitution and is typically found at the ends of long bones, proximal to joints and within the bodies of vertebrae. Cortical, compact bone makes up approximately 80% of bone. The remaining approximate 20% is trabecular, cancellous bone (Guyton and Hall, 2011).

Cortical bone is 90% mineral and 10% collagen (Cowin, 2001). It is densely packed (hence, “compact”) and has a slow metabolic rate so that bone turnover occurs at a slower rate than trabecular bone; however, there is considerable inter- and intra-individual variation in cortical bone turnover rates (Frost, 1960; Guyton and Hall, 2011). Cortical bone is composed of three surfaces, or envelopes, which are all sites where remodeling occurs: 1) the endosteal envelope, which lies adjacent to the marrow cavity and is characterized by a relatively high bone turnover; 2) the periosteal surface, which is the outer membrane of bone and the attachment site of tendons, ligaments, and muscles; and 3) the intracortical envelope, located inside the cortex within the Haversian systems (Guyton and Hall, 2011).

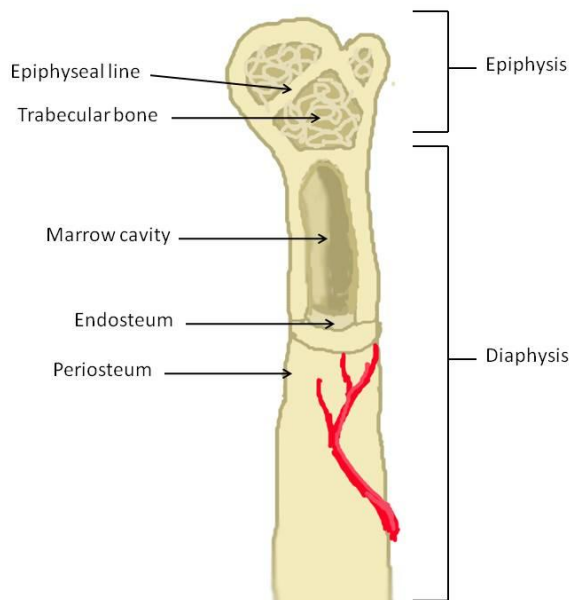


Figure 2.2. Macroscopic constituents of bone.
(Illustration by Tara J. Cepon)

Trabecular bone is organized by concentric sheets of bone and generally does not contain osteons. Each trabecula is a thin strut of bone surrounded by blood vessels and when clustered together form trabecular networks resembling lattice-like structures. The strength of trabecular bone depends both on the alignment and density of the trabeculae, and the degree of interconnectedness between trabecular struts (Merz and Schenk, 1970; Mosekilde et al., 1987; Weinstein and Hutson, 1987). Because trabecular bone has a greater surface to volume ratio and more surfaces adjacent to bone marrow, it is generally subject to greater metabolic activity and faster rates of turnover than cortical bone (Guyton and Hall, 2011). When a trabecula thins to a certain point, active osteoclasts may perforate the bone and disconnect it from the remaining trabecular network, which is irreversibly weakens that region of bone. A thinning trabecula, however, can thicken again if osteoblasts produce more new bone than is resorbed by osteoclasts (Mosekilde, 2000).

Modeling and Remodeling

Bone formation can be divided into processes that occur during growth (modeling) and repair of adult bone (remodeling). Modeling is a process of organized bone cell activity that allows bone to grow, as well as to adjust bone strength in adulthood. This process principally controls bone shape and size during growth and in adult bone is responsible for the expansion of the cortex and periosteum (Frost, 1986). During growth, bone has a greater capacity to react to external loads than during any other developmental stage. It is for this reason that this is a crucial period for establishing bone mass. Although peak bone mass (maximum bone density) is typically attained

between 25-30 years of age, early childhood and adolescent bone modeling play a critical part in determining peak bone mass, and also bone mass throughout adulthood.

Bones possess the ability to regenerate continuously in a process called remodeling, which is defined as the production and maintenance of metabolically sufficient bone in response to changing external circumstances. The loss of mineral and changes in the bone matrix in adult bone compromises its elasticity and strength, and heightens risk of fractures (Cho and Stout, 2003). Therefore, regular bone turnover must occur in order to mobilize calcium, replace old bone tissue, adapt the micro-architecture to different loads and stressors, and repair damaged or fractured bone (Guyton and Hall, 2011). The remodeling process takes on a new dimension during pregnancy and lactation when maternal skeletal calcium stores are drained to meet the requirements of the baby's growing skeleton. In addition to the positive dimensions of remodeling, this process can also have negative effects by removing trabeculae as well as causing an increase in cortical bone porosity, which are both actions that will lead to a reduction in bone strength. Remodeling processes continue throughout life but as adulthood progresses, bone resorption begins to occur at a greater rate than bone formation; this will result in a net loss of bone and a consequent reduction in trabeculae, thinner cortices, and expanded marrow cavities.

The Role of Calcium

The process and rate of remodeling relies on the ability of the skeletal system to efficiently mobilize calcium. Calcium is the most abundant mineral in the human body. Ninety-eight percent of body's calcium is contained in the skeleton, and it typically

accounts for approximately 1 kilogram (kg) of adult body weight (Guyton and Hall, 2011; Stini, 1995). Calcium is essential to many vital cellular functions, including maintenance of cell membrane permeability, nerve impulse conduction, regulation of muscle contraction, and hormonal regulation (Guyton and Hall, 2011). Loss of the mineral occurs on a daily basis through shedding of the skin, the hair, and the nails, and through excretion processes such as urine, feces, and sweat.

Calcium Homeostasis: In general, the system of calcium homeostasis has evolved efficient and reliable mechanisms to avoid insufficient or overabundant levels of circulating calcium by stimulating bone turnover and remodeling processes. However, breakdown of homeostasis can occur, which may lead to hypo- or hypocalcaemia states. In conditions when the exogenous supply of calcium is insufficient, bone tissue is resorbed in an effort to maintain constant serum calcium levels. Insufficient levels of circulating calcium, or hypocalcaemia, can produce uncontrollable muscle spasms (tetany) and can lead to death by cardiac arrhythmia. In contrast, an excess of calcium, or hypercalcaemia, can result in extra-osseous calcification, particularly in the kidney and can in some cases lead to renal failure (Stini, 1995). A key mechanism that helps avoid hypocalcaemia is the reduction of calcium absorption in the intestine. At most times during life, human calcium absorption operates at a relatively low efficiency. However, during certain times, such as during the adolescent growth spurt, pregnancy, and lactation, calcium absorptive efficiency improves dramatically (Agarwal and Glencross, 2011). This issue is explored further in Chapter IV. It is clear that the increased calcium requirements of the growing embryo or offspring typically mobilize maternal skeletal reserves, but that mother's improved ability during pregnancy and lactation to absorb

calcium from dietary sources helps to offset or reduce the magnitude of this problem. This shift in calcium homeostasis can have important implications for bone formation in the post-partum, post-lactation state.

ASSESSING SKELETAL HEALTH: BONE MINERAL DENSITY

Measures of Skeletal Health

Bone mineral density (BMD) is the most important diagnostic parameter of bone health and is commonly used in clinical settings as an objective and reliable predictor of future fracture risk (Bartl and Frisch, 2004; Levis and Altman, 1998; Ross et al., 1995). Whereas bone mineral content (BMC), measured in grams, is the absolute amount of calcium and other minerals in a bone, BMD is measured in terms of area (g/cm^2) or volume (g/cm^3) (Bartl and Frisch, 2004). BMD represents the amount of bone tissue present at the end of skeletal maturation (peak bone mass) minus any bone loss.

Bone loss is a heterogeneous process and can result in changes to the size and organization of cortical bone and/or trabecular bone. Progressive loss of bone density with age appears to be typical for both women and men but, as discussed earlier, variation in the timing of onset of bone loss and the rate of bone deterioration have not been extensively studied. In addition to BMD, fracture risk is also related to bone quality changes including overall bone geometry and micro-architecture (Njeh et al., 1999). It is possible that in two individuals with the same low bone density, one may exhibit thin trabeculae with a normal trabecular network and have a low fracture risk, whereas the other individual may display porous bone with a reduction of trabecular connections and have a high risk of fracture (Marcus, 1991).

Techniques for Assessing Skeletal Health

Dual-energy x-ray absorptiometry (DXA or DEXA) is the most widely used method for assessing BMD and diagnosing osteoporosis in clinical settings. The technique involves two x-ray beams positioned on a specific body site and the measurement of beam attenuation, a parameter related to bone mineral content; this then allows the calculation of BMD. DXA can measure central (hip and spine) and peripheral (forearm) sites, and also has the capacity to be used to perform a full body scan. Although considered the “gold standard” in bone density techniques, DXA scanners are extremely expensive, are not portable, and expose participants to ionizing radiation; these factors limit its use for non-clinical purposes.

Quantitative computed tomography (QCT) is a well-established technique that may be applied to the lumbar spine and appendicular skeleton in order to generate BMD values as well as cross-sectional images of trabecular and cortical bone (Bartl and Frisch, 2004). However, the presence of marrow fat in the vertebral bodies may cause an underestimation of BMD by up to 15% (Bartl and Frisch, 2004). As with the DXA technique, QCT methods also expose participants to radiation, which limits its use to clinical or laboratory settings.

Quantitative ultrasound (QUS) is a relatively new technique that allows for the measurement of skeletal health. QUS provides information on both cortical and trabecular bone, and its measurements are influenced by several factors, including bone micro-architecture, bone mineral constituents, and elastic modulus (Bartl and Frisch, 2004). QUS shows great promise for both research and population-level screening, in part because it is portable, relatively inexpensive, and does not expose participants to ionizing

radiation. Quantitative ultrasound involves the placement of transducers on either side of the bone of interest and use sound waves to generate three measures of bone: broadband ultrasound attenuation (BUA; dB/MHz), speed of sound (SOS; m/s), and heel BMD (hBMD). BUA is defined as the slope of this attenuation versus frequency curve. This parameter may be understood using the analogy of a slinky toy. When the toy is stretched and then released, the rings will oscillate for a period of time, with the oscillations subsiding in intensity before finally stopping. A similar process occurs with sound waves as they pass through bones; some of the energy is lost from the sound wave and the oscillations of the sound wave are diminished (Bonnick and Lewis, 2006). BUA reflects the amount of energy that is lost in relation to the density and micro-architecture of the bone. The second measure, SOS, is defined as the distance between the two opposing transducers divided by the time it takes for the signal to pass from one transducer, through the bone of interest, to the other transducer. Higher BUA and SOS values indicate greater bone density. While there is generally a linear relationship between BUA and SOS, differences in the rate of bone loss in these measures reflect the different qualities of bone being measured (Evans and Tavakoli, 1990).

QUS, particularly when applied to the heel bone (calcaneus), has proven to be an excellent screening tool for low BMD. Calcaneal ultrasound measures provide accurate and repeatable measures of bone density, and numerous studies have documented an excellent correlation between DXA measures and calcaneal ultrasound (Barkmann et al., 2007; Gerdhem et al., 2008; Nayak et al., 2006). The calcaneus is a weight-bearing site rich in trabecular bone and, although there is certainly variation in bone density among different skeletal regions, the bone is an ideal single-site measure of bone density.

Further, unlike x-ray measures, QUS-transmitted waves not only reflect the amount of bone but also the trabecular distribution and orientation (Barkmann et al., 2007). The calcaneal ultrasonometer remains one of the few techniques available for the assessment of BMD in remote field conditions.

FACTORS THAT AFFECT SKELETAL HEALTH

A number of factors shape skeletal health by influencing the processes of bone formation and resorption. These include hormones, diet, and various environmental factors, which affect bone growth, maintenance, and/or fragility. Genes also play a role in skeletal metabolism and specific genetic markers controlling bone quantity and quality have been isolated (Drake et al., 2001; Van der Weyden et al., 2006). However, genes only explain a small proportion of the variation in individual bone mass and do not determine the rate of bone loss over the lifespan (Cooper et al., 2002). Rather, epidemiological studies have demonstrated the importance of non-genetic factors, modifiable factors in influencing the accumulation of bone mineral across life. The following section discusses the role of these modifiable agents and focuses on dietary, physical activity, hormones, and reproductive factors in determining skeletal health.

Diet

Nutritional factors are important in determining peak bone mass and the rate of bone loss during adulthood. As discussed above, adequate dietary calcium intake is critical for maintaining bone integrity. Even more critical for diminishing fracture risk than the amount of calcium being ingested is the calcium absorptive abilities of the body

(Vieth, 2003). Key sources of dietary calcium include dairy products, broccoli, beans, green leafy vegetables, and fish. The National Osteoporosis Foundation (NOF) (2010) recommends a daily allowance of dietary calcium of at least 1,200 mg per day. The calcium recommendations for pregnant and lactating women are higher and the need for supplementation varies across populations depending on availability of reliable calcium sources. In general, the effect of calcium supplementation on bone density continues to be debated as some studies find no positive effect of calcium intake on bone loss rates with aging (Anderson et al., 2004; Dawson-Hughes, 2004). With regard to females and reproduction specifically, some studies show a positive association between calcium intake and bone density maintenance across the female lifespan, although most studies conclude that bone loss occurs despite calcium intake (Cumming and Nevitt, 1997; Looker, 2003; Wosje and Specker, 2000). Additional evidence that maternal intake is independent of changes in bone is provided by Prentice and colleagues (1995) who demonstrate that supplying calcium supplements to women with low calcium intake does not affect calcium levels in their breast milk or changes in bone metabolism. The loss of bone during lactation occurs in women with high and low calcium intake alike.

Vitamin D is a key regulator of intestinal calcium absorption and thus contributes to bone health. A daily vitamin D allowance of 800-1,000 IU is suggested although this quantity may be ingested through sources such as vitamin D-fortified milk, cereals, egg yolks, and liver (NOF, 2010). Current guidelines for prevention of osteoporosis strongly recommend taking calcium along with vitamin D supplements to obtain maximum nutrient benefits.

Vitamin C also appears to be an important contributor to skeletal health, as epidemiological studies have documented a positive association between vitamin C and bone mass (Bunker, 1994). This likely stems from the fact that vitamin C is necessary for intercellular substance secretion by all cells, including osteoid formation (Guyton and Hall, 2011). This nutrient also stimulates osteoblastic activity and improves calcium absorption. The recommended daily allowance of vitamin C for adults is 75-100 mg and can be primarily obtained by consuming citrus fruits.

Vitamin K may also contribute to increased bone mass because of its role in synthesizing osteocalcin, a major component of the non-collagenous proteins in bone, (Bartl and Frisch, 2004). This nutrient regulates the attachment of calcium to the bone matrix and is thus critical for healing fractures. In order to maximally benefit from the bone maintaining properties of vitamin K, the suggested daily vitamin K allowance is 100-300 mg, which may be acquired through supplementation or by consuming dark-green vegetables (e.g., spinach).

Several other dietary factors, including caffeine, alcohol, processed foods, carbonated cola beverages, and excessive protein have been negatively associated with bone mass (Bunker, 1994; Hernandez-Avila et al., 1993; Lazenby, 1997; Sampson, 2002). However, establishing causality between any one of these factors and bone loss can be problematic.

Physical Activity

Physical activity is a well-established factor that influences bone density. High activity levels, especially those that involve weight-bearing, lead to increased bone

formation and reduced bone loss (Proctor et al., 2000; Stini, 1995). Following Wolff's Law, bone adapts to stressors that are placed upon it and thus, bone will respond to low to moderate exercise through accretion of new bone. Aside from inhibiting bone loss in adulthood, physical activity promotes strength, flexibility, balance, and endurance, and thereby decreases the risk of falls in the elderly by up to 20-60% (Bartl and Frisch, 2004; Dargeant-Molina et al., 1996; Kemper et al., 2000). The optimal amount of exercise necessary to maintain bone density varies by sexes and among individuals. While high impact exercise appears to have the greatest positive effect, excessive exercise, particularly in women, can reduce circulating estrogens and lead to a concomitant decrease in bone density. Additionally, high levels of energy expenditure coupled with disordered eating patterns (e.g., anorexia or bulimia) can lead to negative energy balance and result in reductions in bone density, and heightened fracture risk (Zanker and Hind, 2007).

In addition to the types of mechanical loading on bone, the timing of physical activity is also a key factor determining fracture risk since the amount of bone mass gained during growth and development significantly contributes to adult bone density (Kemper et al., 2000; Streeter and Stout, 2003). Several studies have shown that bone mass increases dramatically during puberty; growing bones have a greater potential for periosteal expansion than aging bone, and therefore adapt more rapidly and efficiently to acute stressors in order to increase strength (Kröger et al., 1993; Matkovic et al., 1994; Theintz et al., 1992). Up to 60% of the variance in BMD at age 65 may be the result of the peak bone mass attained early in life (Kelly et al., 1995). This relationship emphasizes the importance of establishing high peak bone mass for limiting the age and

lifestyle-related influences on bone loss. Thus, the contributions of physical activity are arguably strongest during pubescent years when there is rapid response of bone to stressors that will result in accretion.

Systemic Regulation of Calcium

The equilibrium of the central calcium pool is maintained by calcitropic hormones such as parathyroid hormone, vitamin D, and calcitonin. As a result of their effect on calcium regulation, these hormones are agents that contribute to skeletal health.

Parathyroid Hormone (PTH): Secreted by the thyroid gland, PTH is the primary hormone that regulates calcium homeostasis. Its mechanism of action involves inciting osteoclastic activity in response to low serum calcium level to stimulate the release of calcium and phosphate from the skeleton (Forwood, 2001). PTH also synthesizes 1,25-dihydroxycholecalciferol, the principle active product of vitamin D, which functions to decrease the excretion of calcium by the kidneys (Guyton and Hall, 2011). Shifts in PTH occur in response to pregnancy, during which some women experience a reduction in PTH; PTH levels typically return to baseline following parturition.

Vitamin D: Vitamin D, or the hormone calciferol, plays an important role in both bone resorption and deposition. Vitamin D is available in two major physiologically relevant forms: Vitamin D₂ (ergocalciferol) and Vitamin D₃ (cholecalciferol). Vitamin D₃ is produced in the skin after exposure to sunlight or artificial sun sources, whereas D₂ is derived from plants that have been exposed to ultraviolet exposure. Often regarded as equivalent and interchangeable (Committee of Revision, 1997; Institute of Medicine, 1997), some controversy exists over whether Vitamin D₂ can fully substitute for the D₃

variety in the human diet. Recent research has found that Vitamin D₂ may be less effective in raising serum concentrations of Vitamin D, and identify Vitamin D₃ as the major endogenous, source of Vitamin D. The studies conclude that Vitamin D₂ should not be regarded as a nutrient suitable for supplementation (Adams and Hewison, 2010; Houghton and Vieth, 2006; Holick, 2008).

Vitamin D, in general, influences the transport of calcium through cellular membranes, affects renal and intestinal absorption of calcium, and enables mobilization of calcium from bones (Dawson-Hughes, 2004). Vitamin D also improves muscle strength, balance and leg function, which decreases the risk of falling and thereby reduces risk of future fractures. This hormone has many beneficial effects across age groups and developmental stages. Sufficient levels of vitamin D are especially important during childhood growth as deficiencies can result in the childhood disease, rickets. During pregnancy, vitamin D rises in serum concentration and remains elevated. This heightened presence appears to promote an increased efficiency in transporting of calcium into circulation. Vitamin D may be obtained through dietary sources or endogenously synthesized exposure to adequate amounts of sunlight (Vitamin D₃). Deficiencies of this nutrient are rarely found in equatorial populations (Mazess, 1978; da Rocha and Ribiero, 2003).

However, despite the availability of sunlight, cultural and religious elements can play a significant role in an individual's exposure to Vitamin D. For example, high rates of hypovitaminosis D are documented among Muslim women in the Middle East and the Indian subcontinent where many women maintain a conservative style of dress (e.g., burkas) that covers most of their bodies, including hands and faces and limits their

exposure to sunlight (El-Hajj Fuleihan and Deeb, 1999; El-Sonbaty and Abdul-Ghaffar, 1996; Ghannam et al., 1999; Gannage-Yared et al., 2000).

Calcitonin: Calcitonin, like PTH, is secreted by the thyroid gland but acts in opposition to PTH by reducing serum calcium concentrations (Galloway, 1997). This reduction is achieved by suppressing the resorption of bone by inhibiting osteoclastic activity, and thus limits the release of calcium into the blood. The effects of calcitonin on blood calcium ion concentration are relatively weak and, for this reason, the hormone is seldom considered (Bartl and Frisch, 2004). However, during pregnancy, calcitonin levels are relatively high and this may inhibit bone resorption while simultaneously allowing PTH and vitamin D to activate absorption of dietary calcium and kidney reabsorption (Galloway, 1997).

Other Hormones

Estrogens: Natural estrogens are steroid hormones that come in three major forms. The primary form that is present in human females from menarche to menopause is estradiol. The two other forms of estrogens are estrone, which is found in higher concentrations among postmenopausal women, and estriol, which is only produced in significant amounts during pregnancy. In general, studies that address the effects of estrogens on bone density refer to the most common estrogen form, estradiol.

Estrogens appear to play a central role during the course of the female lifespan by influencing bone and collagen formation, and increase intestinal absorption and retention of calcium. By limiting bone resorption, estrogens help to maintain bone mass. Deficiencies in estrogens precipitate production of osteoclastogenic factors, which results

in increased porosity of cortical bone and resorption of trabecular bone (Pacifci, 2007); this leads to overall bone loss. Estrogens are also responsible for epiphyseal fusion of long bones and are stronger than the effect of testosterone in males. As a result, female growth usually ceases before males.

A large body of research suggests that changes in bone mass coincide with levels of estrogen during pregnancy, lactation, and menopause (Agarwal and Stuart-Macadam, 2003; Galloway, 1988; Galloway, 1997; Guyton and Hall, 2011; Pacifci, 2007; Pearce, 2006). Estrogen levels fluctuate throughout the female life cycle, with monthly fluctuations starting at the onset of menarche. Following menopause, no estrogens are secreted by the ovaries and, consequently, osteoclastic activity is no longer inhibited. The result is a decrease in the deposition of bone calcium and phosphate and therefore, a reduced bone matrix (Guyton and Hall, 2011). Because of their role in bone maintenance, estrogen hormone replacement therapy (HRT) may be offered as treatment to inhibit bone loss in postmenopausal women. However, it is important to note that several studies have found that HRT can have severe health risks for women including increased risk of breast cancer, heart attack, stroke, and blood clots (Lindsay et al., 1996).

Due to the hypoestrogenic states of premenopausal removal of ovaries (oophorectomy), premature ovarian failure, anorexia nervosa, and exercise-induced amenorrhea, these conditions are characterized by low bone density and an increase osteoporosis risk, a relationship that is well-documented in the literature (Meczekalski et al., 2010; Popat et al., 2009).

Progesterone: Progesterone also plays a key role in promoting bone accrual through its influence on proliferating osteoblastic activity. Several studies have reported

that increased progesterone levels in postmenopausal women result in new bone formation with greater effects than even estrogen therapy (Munk-Jensen et al., 1988). That is, while estrogens will prevent further bone loss, progesterone may function to accumulate new bone. Fluctuations in progesterone are documented during pregnancy and lactation and the hormone may be continually secreted before parturition thereby initiating birth contractions (Galloway, 1997).

Growth Hormone (GH) and Insulin-Like Growth Factors (IGFs): GH stimulates growth and cell reproduction in humans and other animals. Its effect on the regulation of longitudinal bone growth and bone metabolism has only been recently recognized (Ohlsson et al., 1998). A number of in vivo and in vitro studies have shown that GH controls aspects of bone formation and resorption and plays an important role in the process of bone growth until peak bone mass is achieved (Bouillon, 1991; Eriksen et al., 1993; Eriksen et al., 1996; Sloomweg, 1993; Wuster, 1993). Several studies have demonstrated that GH increases the local production of IGF-I, one of the most abundant growth factors present in bone (Isaksson et al., 1987; Isgaard et al., 1988). IGF-I has demonstrated a role in stimulating osteoblastic activity, which results in the formation of bone and inhibition of resorption. Studies on genetically modified mice have established the importance of IGF-I in the acquisition and maintenance of trabecular bone mass (Bikle et al., 2001).

GH and IGFs affect the epiphyseal growth plate, and some abnormalities in skeletal growth have been associated with an excess or deficiency in these hormones (Khan et al., 2001). They may also contribute to the accretion and/or maintenance of bone mass during young adulthood and with progressing age, GH and IGF declines may

reduce bone formation. Because of their role in stimulating bone accretion, GH and IGFs have been proposed in therapy for osteoporosis.

Reproductive History

Reproductive patterns among women appear to influence BMD across the lifespan. Reproductive parameters linked to changes in skeletal health include parity, patterns of breast-feeding, and age at first pregnancy. This relationship is unsurprising given that circulating hormones associated with pregnancy, lactation, and menopause have been shown to have an effect on bone resorptive and formation processes (see earlier section: *Other Hormones*). For this reason, reproductive biology accounts for substantial part of the disparity in bone density between females and males. For example, one in five men experience osteoporotic fractures, whereas one-third of women over age 50 are at risk for future fractures. However, it is important to note that rates of osteoporosis-related fractures are increasing among both sexes (IOF, 2010).

During certain periods of the female life, regulatory hormone levels alter according to the variable calcium requirements of the body, with the greatest “cost” occurring during pregnancy and lactation. Unsurprisingly, male reproductive patterns have less of an effect on skeletal health than women, although men are susceptible to increased bone resorption due to testosterone deficiency that occurs in the later stages of life.

The following section reviews evidence for the effects of different female reproductive states on skeletal health.

Menarche: Menarche, the onset of menses, marks the beginning of female reproductive life. Menarche may have a stimulating effect on the development of bone by increasing the osteoblastic activity that accompanies the unsettled estrogen (Ito et al., 1995; Jaffe and Dell'Acqua, 1985). Because of circulating estrogens that are introduced at menarche and their role in bone accretion processes, an earlier age at first menses (<16 years old) has been linked to higher BMD values (Ito et al., 1995; Roy et al., 2003). Additionally, early menarche is typically related to larger body size, which increases mechanical load on the skeleton, and to greater adiposity, which leads to increased production of estrogens. Thus, both larger body size and greater fat content theoretically contribute to increases in bone quantity (Eastell, 2005).

Pregnancy: Pregnancy is characterized by heightened and continual maternal bone turnover, independent of the calcium demands of the fetus (Cerroni et al., 1993). During pregnancy, the mother develops mechanisms to balance calcium requirements of the developing fetus in order to allow for fetal bone formation. The average newborn skeleton contains a total of 25-30 grams of calcium, and 80% of this calcium crosses the placenta in the last trimester (Stini, 1995). If maternal bone mineral were the sole source of calcium for the fetus, the mother would lose approximately 3% of her skeletal mass with each pregnancy (Khan et al., 2001). Mechanisms of calcium absorption appear to have evolved to minimize the deleterious effects on maternal skeleton.

Several studies have documented positive associations between the number of full-term pregnancies and bone density. This protective effect of parity against bone loss has been linked to pregnancy-related weight gain (which involves increased mechanical loading), increased intestinal calcium absorption, heightened cumulative exposure to

estrogens, and possible delays in age at menopause (Nguyen et al., 1995; Streeten et al., 2005). Further, studies comparing bone density in nulliparous and multiparous women have generally documented lower BMD in females with no children (Sowers et al., 1992). However, studies of pregnancy and bone health have not been entirely consistent. Several studies have documented negative associations with BMD with increasing parity (Allali et al., 2007; Gur et al., 2003), while others documented no significant correlations (Ensom et al., 2002; Melton et al., 1993). One potential explanation for these contradictory results is that the majority of these studies were conducted among Western populations that are typically characterized by low fertility rates; therefore, the effects of multiparity may be obscured by relatively small offspring quantities.

Age at First Parturition: Another reproductive factor that appears to shape bone health is age at first parturition. As bone mineralization typically continues until the mid-twenties, pregnancy and lactation may disrupt bone formation in younger females that give birth and lead to impacts on long-term bone mass (Sowers, 1996). One recent epidemiological study found that women who were 27 years or older at the time of their first pregnancy had a significantly lower risk of developing osteoporosis in postmenopausal life (Schnatz et al., 2010). In addition to long-term effects, a younger age at first pregnancy may have negative impacts during pre-menopausal years. In several epidemiological studies, women who were younger at first parturition (<20 years old) demonstrated an impaired ability to gain in height when compared to other women, indicating disruptions during early bone development (Allal et al, 2004; Gigante et al., 2006; Sear et al., 2004). However, the effect of the timing of first pregnancy on bone

mass is not consistent across studies with some research finding no significant relationship at all (e.g., Sowers et al., 1985).

Lactation: Lactation is a hypoestrogenic state where the demand for calcium by the infant influences the rate of maternal bone turnover (Cerroni et al., 2003). This period is characterized by altered levels of calciotropic hormones (e.g., PTH, calcitonin) that respond to breast milk production, fluctuating levels of ovarian hormones, and elevated levels of hormones associated with breastfeeding (e.g., prolactin and oxytocin). Prolactin is involved with the initiation and continuation of lactation, and during pregnancy this hormone may increase intestinal calcium absorption (Galloway, 1997; Guyton and Hall, 2011). The high levels of prolactin associated with repeated suckling appear to deplete maternal bone mass. In a six month period of exclusive lactation, approximately 7% of maternal bone mass may be lost (Sowers, 2001). After weaning, this process typically reverses and bone accretion occurs over the course of a few months.

Several longitudinal studies that document changes in bone status during post-partum and weaning periods typically report that bone loss is transient and later restored to pre-pregnancy values (Pearce, 2006; Sowers, 1996). The general consensus among research studies is that BMD recovers after weaning, although there is debate regarding exactly when this recovery occurs—data suggest anywhere from 6-24 months post-parturition (Ensom et al., 2002; Kalkwarf and Specker, 1995; Matsumoto et al., 1995; Sowers et al., 1993).

Studies on the effects of lactation on bone mass in the long-term, postmenopausal life are more replete with inconsistencies. Associations between the duration and intensity of lactation on bone density have been documented, but with considerable

variation in findings. Several studies have shown that long-term breastfeeding is associated with increased bone loss in the postmenopausal years (Chowdhury et al., 2002; Grimes and Wimalawansa, 2003; Lopez et al., 1996; Melton et al., 1993; Popivanov and Boianov, 2002; Sowers, 1996), whereas some research has found no such association (Feldblum et al., 1992; Johnell and Nilsson, 1984), and yet others demonstrate a decrease in maternal bone density due to lactation history (Lissner et al., 1991; Wardlaw and Pike, 1986).

The lack of consensus regarding the long-term effects of lactation may be largely due to the heterogeneity in lactation behaviors on a cross-cultural and inter/intra-population level. Women can resume menstruation as early as two months after parturition or as late as 3 years postpartum, with a broad range of lactational amenorrhea lengths along the spectrum (Konner and Worthman, 1980; Jones, 1989; Wood, 1994).

Birth Spacing: Studies on the frequency and spacing of pregnancies suggest that the greatest effects of bone mass occur during the first reproductive cycle (Peng et al., 1987). Further reductions occur with subsequent pregnancy/lactation periods. However, after the third cycle, little additional bone loss is observed, which suggests a threshold effect on bone integrity (Peng et al., 1987). Again, results of different studies are not entirely consistent, and there may be an effect of the total number of births.

Short spacing between births may be a risk factor for low BMD in pre- and postmenopausal years, due to the cumulative periods of BMD loss in quick succession (Affinito et al., 1996). However, most longitudinal studies that have investigated intervals between childbirth and lactation periods have found that bone health is not compromised among women with shorter birth spacing (Laskey and Prentice, 1997; Sowers et al.,

1995). In some high fertility populations, repeated and closely spaced pregnancies with extended lactation, such as the Omani and grand multiparous Finish American women, had no significant effect bone density (Bererhi et al., 1996; Henderson et al., 2000).

Menopause: According to the World Health Organization (WHO), the risk for osteoporosis in postmenopausal women is approximately three times higher than in older men. The cessation of ovarian function at menopause results in reductions in estrogen and progesterone levels, and these declines are associated with precipitous reductions in bone mass (Black and Lane, 2002; Bartl and Frisch, 2004). Coupled with changes in the levels of female sex steroids are declines in the levels of calcium-regulating hormones with advanced age.

Postmenopausal (or Type I) osteoporosis is characterized by an exaggerated loss of trabecular bone, the predominant constituent in vertebrae and the proximal femur. This increases the susceptibility of these sites to fracture. Early menopause is widely believed to be a risk factor for reduced BMD (Gallagher, 2007; Kritz-Silverstein and Barrett-Connor, 1993; Ohta et al., 1996; Pouillès et al., 1994). Because of menopause-related declines in this sex steroid, women who experience earlier menopause spend more time in a hypoestrogenic state, which may place them at greater risk for poor bone health (Gallagher, 2007; Kritz-Silverstein and Barrett-Connor, 1993; Ohta et al., 1996; Pouillès et al., 1994). However, studies show no consensus on what constitutes ‘early.’ Sioka et al. (2010) found that osteoporosis was higher among Greek women who reported menopause onset between 40 and 45 years, when compared to those at an older age. Among Iranian and Indian women, Keramat and colleagues (2008) report that women with an age at menopause less than 45 years are at higher risk for osteoporosis. In

contrast, several studies suggest that age at menopause is unrelated to increased fracture risk (Kritz-Silverstein and Barrett-Connor, 1993; Melton et al., 1993; Vico et al., 1991). As with most reproductive variables, the relationship between menopausal age and skeletal health remains unclear.

BONE LOSS IN AN EVOLUTIONARY PERSPECTIVE

Evolutionary Basis for Bone Loss

Bone is a living, dynamic tissue that, like other aspects of our anatomy and physiology, is shaped by evolutionary pressures. As discussed earlier in the chapter, bone serves multiple purposes and, in order to better understand the evolutionary origins of these functions, they must be examined, not only within a strictly adaptationist perspective, but also one that considers compromises and trade-offs. The system of calcium homeostasis evolved with the dual function of maintaining the mechanical properties necessary for bone's structural role (e.g., tensile strength), and for the skeleton to store calcium for basic bodily functions (e.g., regulation of muscle contractions, nerve conduction, and modulation of cell membrane permeability) when adequate dietary sources are not available.

The diet of the earliest hominins was likely similar in many ways to modern chimpanzees and bonobos (*Pan spp.*), with a diet characterized by foods with high calcium density (2-2.5 millimoles/liter [mmol] per 100 kcal) (Eaton and Nelson, 1991). Contemporary and historically-known hunter-gatherer populations in Africa, South America, and New Guinea also have diets that are nearly as high in calcium (1.75-2 mmol per 100 kcal) (Eaton and Nelson, 1991; Stini, 1995). This contrasts markedly with

calcium intake in contemporary industrialized countries. For example, the median calcium intake North American women, as well as populations in many European countries, is extremely low (<15 mmol per day per 100 kcal). Based on this contrast, Stini (1995) posits that calcium homeostasis among humans evolved in a context of a diet extremely high in calcium.

Excessive serum calcium levels, or hypocalcaemia, can have a number of negative effects including kidney dysfunction, dementia, vomiting, and may even be lethal. Selection appears to have shaped human physiology primarily to protect against hypercalcemic states. General human inefficiency to absorb calcium may have evolved to include mechanisms that protect against serum calcium concentrations that were too high. Thus, while beneficial in some contexts, the evolved inefficiency to mobilize calcium, coupled with novel environments that are characterized by lifestyle and reproductive patterns strikingly different than those of our evolutionary past, has resulted in the high rates of osteoporosis among contemporary, Western populations.

With senescence, calcium absorptive efficiency decreases with advancing age resulting in greater bone resorption relative to formation. There may also be life-history tradeoffs in bone formation with age. It seems possible that the selective benefits of continued high investment in bone formation are reduced with age, while the need to maintain calcium resorptive abilities for various non-skeletal cellular functions declines at a slower rate. The result is that the balance between formation and resorption shifts, emphasizing resorption with age, and results in reductions in the structural integrity of the skeleton. This loss in structural integrity with age is further reduced by the relatively

sedentary habits and calcium-deficient diets of many Western populations (see: *Lifestyle Change and Skeletal Health* below).

The evolution of calcium homeostasis among females has also been shaped by the energy and resource-expensive reproductive functions, such as pregnancy and lactation. Furthermore, females must balance fetal and neonatal requirements while simultaneously maintaining maternal calcium homeostasis and skeletal integrity. Since most female bone loss is postmenopausal, selection to maintain bone later in lifetime would be reduced. During this life stage, bone physiology would have been decoupled from direct fitness benefits, although it may be maintained by other factors such as inclusive fitness benefits of somatic maintenance (e.g., Hawkes et al., 1998). Galloway (1997) argues that postmenopausal bone loss is an example of antagonistic pleiotropy, which suggests that traits beneficial to early reproductive life, including relatively low absorptive efficiency but highly effective mechanisms for the mobilization of calcium for offspring, become deleterious when coupled with other age-related physiological changes.

The gradual expansion over the past century of the average life expectancy beyond reproductive years appears to have exposed the hidden costs of reproduction. In this respect, accelerated bone loss in postmenopausal life can be seen as an epiphenomenon related to the rapid depletion of estrogen that occurs at menopause. Conversely, however, the pattern of accelerated postmenopausal bone loss may reflect the evolution of an optimal age-related trade-off between the benefits of continued calcium availability from bone, the need to maintain sufficient structural functions of bone across the remaining lifespan, and the parental investment or inclusive fitness benefits of continued work and life effort across the later lifespan.

Lifestyle Change and Skeletal Health

While bone density decline with age has been observed in past populations (Agarwal and Stuart-Macadam, 2003; Karasik et al., 1998; Mays, 2000), clinically recognized osteoporosis and postmenopausal osteoporosis is far more prevalent in contemporary, Western populations (Mays, 2000). Few fragility-related hip fractures related to age-related bone loss have been reported for archaeological populations. The paucity of these findings in skeletal assemblages may be explained by a few factors. Firstly, while in past societies there would have been a significant number of individuals who reached old age, the proportion of the population living beyond the seventh and eighth decades of life would have been substantially smaller than contemporary times (Brickley and Agarwal, 2003). Therefore, increases in life expectancy may partially explain the differences in rates of low bone mass between modern and past populations. However, data from extant hunter-gatherers suggest that even though reproduction ceases by 42 years of age or less, over half the women who make it to that age will live another 20 years (e.g., Hill and Kaplan, 1999; Kaplan et al., 2000). Additionally, even when the increased age of Western populations is taken into account, fracture risk is not tied exclusively to life expectancy and moreover, rates of osteoporosis continue to rise (Brickley and Agarwal, 2003; Mosekilde, 2000). So, as with many chronic diseases such as type 2 diabetes and metabolic syndrome, recent increases in the rates of osteopenia and osteoporosis may best be explained as the deleterious health consequences of lifestyle change associated with market integration. That is, we are paying the price for living an evolutionarily novel lifestyle.

While the processes of evolution and changed environments have led to increases to human longevity, there has been a simultaneous rise in the frequency of diseases associated with Westernization. In archaeological populations, the effects of dietary and lifestyle change on health are most often exemplified with the comparison of hunter-gatherers and agriculturalists.

With the transition to reliance on food production (e.g., agriculture and/or animal domestication) as early in some areas as 10,000-12,000 years ago, skeletal populations in archaeological assemblages show various negative impacts of these evolutionarily novel lifeways. Dietary and other lifestyle factors, including sedentism, increased proximity to domesticated animals (and their diseases), and increased population density, produced an evolutionarily novel environmental condition. Bioarchaeology research suggests that changes in bone integrity accompanied the transitions from hunter-gatherer to agriculturalist lifestyle, although because agricultural production can yield calcium-abundant foods such as dairy products and green vegetables (e.g., broccoli), one might expect that bone loss rates would be higher among pre-historic hunter-gatherers than early agriculturalists. Further, because high levels of protein can have a negative effect on bone health by increasing urinary excretion of calcium, hunter-gatherers who had greater animal protein in their diets, could theoretically be expected to exhibit greater risk of osteoporosis. Additionally, Roberts and Manchester (2005) argue that farmers are relatively less active than hunter-gatherers, such that the regular bouts of weight-bearing physical activity among the latter group counteract any effects of bone loss. Yet, early agriculturalists were hardly sedentary and agriculture is arguably a more arduous and time-consuming activity than hunting and gathering (Bridges, 1989).

Comparisons of skeletal assemblages from hunter-gatherer and agricultural populations have demonstrated contradictory conclusions regarding the prevalence of low bone quantity and quality in past populations. Cohen and Armelagos (1984) documented a greater prevalence of cortical bone thinning along with a decrease in bone circumference in early agricultural skeletal populations, leading Roberts and Manchester (2005) to suggest that these may be indicators of osteoporosis. In a prehistoric, pre-Hispanic skeletal population from Gran Canaria (Canary Islands), a high prevalence of osteopenia was found, with rates of low bone density increasing with age (González-Reimers et al., 2001). The Gran Canaria sample engaged in a primarily agriculture-based economy, with some fishing and herding practices. In contrast, Kelley (1980) found that hunter-gatherers from the archaic Indian Knoll skeletal sample exhibited osteoporotic bone lesions and fractures, which was attributed to a lack of calcium in diet and water resources. While the few findings from bioarchaeology are conflicting and often limited for our understanding of osteoporosis prevalence in the past, the transition to agriculture did mark a critical period of dietary and lifestyle change that resulted in significant and unprecedented health outcomes that continue to reverberate into contemporary times.

The evolution of human calcium homeostasis is critical for understanding the ultimate causes of osteoporosis. Although expansions of the human lifespan may partially explain the rise in bone fragility and fractures observed in developed countries, key proximate mechanisms of bone loss may provide better insight into this epidemic. In Western populations, nutritionally-deficient diets and evolutionarily uncharacteristic patterns of physical activity over the life course, coupled with changes in reproductive behaviors from high to low fertility, have demonstrated major effects on contemporary

health. The rise of many chronic diseases, including osteoporosis, illustrates that the ultimate factor underlying chronic diseases in Westernized nations is the mismatch between the ancestral *Homo* genome and the lifestyles of affluent nations (Cordain et al., 2005; Williams and Nesse, 1991). This highlights the compelling need for bone health studies from subsistence-based populations with vastly different lifestyles and reproductive patterns than Western groups in order to illuminate on the factors affecting bone loss and potentially inform global osteoporosis prevention and treatment efforts.

BRIDGE TO CHAPTER III

Chapter II establishes the fundamental basis to understand the mechanisms of bone loss that will be further discussed in Chapters III and IV. The next chapter presents a normative dataset for skeletal health in the Indigenous Shuar and non-Shuar *Colonos* of Amazonian Ecuador. *Colonos* were included in the analysis for comparative purposes as they live in communities close to Shuar, but are not biologically related. Minimal data are available on bone health from populations living in developing countries, and no other normative datasets are available for either sex in the populations addressed. Chapter III concludes with a discussion on the significance of documenting more skeletal health data from similar subsistence-based populations.

CHAPTER III

NORMATIVE CALCANEAL QUANTITATIVE

ULTRASOUND DATA FOR THE INDIGENOUS SHUAR

AND NON-SHUAR *COLONOS* OF THE ECUADORIAN

AMAZON

This chapter has been published as Madimenos FC, Snodgrass JJ, Blackwell AD, Liebert MA, Cepen TJ, and Sugiyama LS in Archives of Osteoporosis (pp. 1-11). All data collection for bone density measures, data analyses, and write-up were conducted by the author of the dissertation. Additional anthropometrics including stature and weight were gathered with the assistance of Blackwell, Liebert, and Cepen. Sugiyama and Snodgrass provided editorial assistance.

INTRODUCTION

Bone mineral density (BMD) data are limited for non-industrialized, natural fertility populations, a gap that obscures our current understanding of the prevalence of osteoporosis in developing countries. In part, the lack of epidemiological data on osteoporosis in developing countries reflects the limited availability of diagnostic resources in these regions. However, relatively non-invasive and portable techniques have recently facilitated the collection of BMD data in remote field settings. Calcaneal quantitative ultrasound (QUS) techniques in particular have proven to be clinically useful as a screening tool for early signs of low BMD, and are highly correlated with dual X-ray

absorptiometry (DXA) measures (Barkmann et al., 2007; Gerdhem et al., 2008; Nayak et al., 2006). Additional reasons for the lack of attention to bone health in developing countries are assumptions that osteoporosis is a disease of developed nations and an inevitable consequence of aging that cannot be effectively treated or prevented (Handa et al., 2008). Indeed, key environmental risk factors for the development of osteoporosis are typically associated with a Westernized lifestyle, characterized by low levels of physical activity and poor diets that include processed foods, coffee, and carbonated soda beverages. Yet demographic transition and the aging of the population, as well as increasing global trends towards Westernization, appear to be contributing to a dramatic rise in the burden of osteoporosis prevalence in developing countries (Woolf and Pfleger, 2005). The paucity of data on bone density in these countries limits our ability to assess osteoporosis prevalence and risk, and complicates prevention and treatment efforts. In order to begin addressing these issues, it is critical to gather and present population-specific normative data on BMD from non-urbanized groups living in developing countries.

The current study was designed to assess bone quality, using calcaneal QUS techniques, among the indigenous Shuar and non-Shuar mestizo colonists (*Colonos*) of Ecuadorian Amazonia. As with other indigenous populations, BMD information for the Shuar is not presently available. Further, published data are virtually non-existent for any subsistence-based, natural fertility population. These type of data are critical because diet, activity, and reproductive patterns all contribute to BMD, but it is currently unclear whether the BMD trajectories observed in low-fertility Western populations will be similar to those of subsistence-based groups. Further, an investigation of the key factors

that structure BMD variation with age among subsistence groups can provide valuable information on the myriad of factors that shape bone health and disease risk in industrialized populations. Inclusion of the *Colono* population was intended to provide a comparative sample of individuals from the same geographical region who also participate in agricultural-based activities but who are not closely related to the Shuar. The cross-sectional data reported here are part of a larger, multi-year study on health and market integration, the Shuar Health and Life History Project.

MATERIALS AND METHODS

Study Populations

Shuar: From an epidemiological perspective, the Shuar in the current study are not a particularly high-risk group for the development of osteoporosis. In the participant communities, Shuar engage in consistent levels of moderate subsistence-based activities year-round (see Chapter V) and consume a diet that is rich in yuca, plantains, and rice with minimal inclusion of foods that are typically associated with poor bone health (e.g., caffeine, alcohol, processed foods, excessive protein) (Bunker, 1994; Hernandez-Avila et al., 1993; Lazenby, 1997; Sampson, 2002). Additionally, deficiencies in vitamin D, a key nutrient for promoting intestinal calcium absorption, are rarely found in equatorial populations because of year-round exposure to sunlight (e.g., Mazess, 1978; da Rocha and Ribiero, 2003). While the Upano Valley Shuar are currently shifting towards an increased reliance on a market economy, including economic development and lifestyle changes similar to those observed in Western countries, the participant communities continue to be highly dependent on a subsistence-based lifestyle.

Colonos: The settlement of non-Shuar *Colonos* into Morona Santiago began during the mid-20th century at the height of the oil boom. While Upano River Valley *Colonos* and Shuar often reside in communities that are in close proximity, they are largely socially and politically divided, a schism that can be attributed to traditional Shuar antagonism toward outside colonization of their land and recurrent land feuds (Rubinstein, 2001).

Similar to the Shuar, *Colonos* in the Upano River Valley live in communities with varying degrees of market dependence. *Colonos* who live in town, particularly in the Sucua canton, typically engage in wage labor, attend school, and have minimal participation in the subsistence economy. Conversely, in rural areas, many *Colonos* are strongly dependent on household-level animal husbandry and agricultural production. The *Colonos* included in this study are primarily from small rural communities scattered across the Upano River Valley where varying degrees of dependence on an agriculture-based lifestyle exist across individuals.

Participants

Typically, there is a high degree of inter-relatedness within rural Shuar communities. Potential issues can arise in assessing bone health for a highly related group of individuals because this can increase frequencies of population-specific alleles responsible for maintaining bone integrity. To avoid these issues, Shuar participants were recruited from four Upano Valley communities. Rural *Colono* communities do not exhibit the same degree of relatedness as the Shuar. Nevertheless, *Colono* participants

were recruited from across a large canton in the Upano Valley comprised of several smaller rural communities. In total, 227 Shuar (136 females and 91 males; 15-86 years old) and 261 *Colonos* (157 females and 104 males; 15-91 years old) participated in this study.

Although conditions in this remote part of Ecuador prevented us from obtaining a random sample of participants, we made every effort possible to enroll a sample of participants that was representative of each community in terms of age and sociodemographics. This study used a cross-sectional design and while this approach is inherently limited for assessing individual bone loss through time, it is useful for determining prevalence of low bone density across a population.

All participants gave individual informed verbal consent, with both parental consent and child assent for individuals under 18 years old. The study protocol was approved by community leaders, the Instituto Ecuatoriano de Seguridad Social (IESS), the Federación Interprovincial de Centro Shuar (FISCH) as appropriate for FICSH-associated communities, and the Office for Protection of Human Subjects at the University of Oregon.

Bone Mineral Density Measurements

Calcaneal QUS parameters were collected on the right heel of each participant using the gel-based Sahara® bone ultrasonometer (Hologic, Inc., Waltham, MA). Two primary QUS values were used for this analysis: broadband ultrasound attenuation (BUA; decibels per megahertz), which is the slope of the ultrasonic attenuation versus frequency as it passes through bone; and speed of sound (SOS; meters per second), a value

determined by the width of the heel and time delay between initial transmission and subsequent receipt of sound waves. BUA is more closely related to the micro-architecture of bone such as trabecular connectivity, while SOS is greatly influenced by the elasticity and mineral constituents of the bone matrix (Bartl and Frisch, 2004; Lee et al., 2010). A third parameter, heel bone mineral density (hBMD), a device-generated measure calculated from BUA and SOS values, was also used. Instrumental quality control scans of the manufacturer-provided phantoms were performed daily.

Additional Measures of Body Size and Reproduction

Participant stature (measured to the nearest millimeter [mm]) and weight (measured to the nearest 0.1 kilogram [kg]) were recorded using a field stadiometer (Seca, Hanover, MD) and digital scale (Tanita BF-558 electronic scale, Tokyo, Japan) respectively. Body mass index (BMI) was calculated as weight (kg)/height (in meters)².

Female participants were asked additional questions regarding current reproductive status. Women were considered post-menopausal if they reported not having experienced a menstrual cycle within the last 12 months and were neither pregnant nor lactating.

Statistical Analyses

Participants were divided into six groups based on age (15-20, 21-30, 31-40, 41-50, 51-60, > 60 years) with a 10-year age-span within each group (except in the 15-20 and over 60 age intervals). Two-tailed Student's t-tests were used to assess differences between ethnic groups and sexes for bone density measures and anthropometric variables.

Stepwise multiple linear regression analyses were conducted to estimate the variation of QUS explained by ethnicity, age, body weight, height, and BMI. All statistical analyses were conducted using SPSS 17.0 (Chicago, IL).

Issues with Normative Data Sets in Developing Countries

Reproductive patterns among women, including parity, breast-feeding behaviors, and age at first pregnancy are known to contribute to bone density changes throughout the lifespan (e.g., Murphy et al., 1994; Nguyen et al., 1995; Streeten et al., 2005). For example, physiological changes characteristic of various reproductive states such as pregnancy-related weight gain, cumulative estrogen exposure, and fluctuations in bone turnover rates invariably influence bone integrity. As a result, pregnant and/or lactating women are typically excluded from normative datasets on bone health. However, presenting normative data on a natural fertility, subsistence-dependent population in a developing country without including women who are pregnant and/or lactating is problematic for several reasons. First, women in many developing countries are pregnant and/or lactating for much of their reproductive life spans (Eaton et al., 1994; Sperling and Beyene, 1997; Weaver, 1998). Exclusion of these women dramatically reduces the size of the participant pool, specifically for critical age intervals when peak bone mass is realized. Further, in developing countries, particularly in the type of rural settings described here, nulliparous women of reproductive age do not represent the norm, as they may be suffering from health or fertility problems. Therefore, presenting normative data on the indigenous Shuar and *Colono* populations without inclusion of this female demographic is problematic. However, in order to maximize our ability to compare

across populations, data on the subset of females who were not pregnant and/or lactating at the time of this study are also presented separately.

RESULTS

Characteristics of Study Participants

Anthropometric and QUS measures by ethnic group and sex, and menopausal status are presented in Table 3.1. Although height, weight, and BMI of *Colono* participants are higher than for Shuar participants, QUS values are significantly higher among the Shuar ($p < 0.001$). While this trend is consistent across the Shuar group when compared to *Colonos*, the most pronounced difference is found between pre-menopausal Shuar and *Colono* females (BUA: $p < 0.05$; SOS and hBMD: $p < 0.001$); the only exception is shown among post-menopausal women where there is no discernable difference in bone health measures between ethnic groups. In both populations, all QUS values are progressively lower in advanced age groups irrespective of sex. Figure 3.1 shows a scatter plot of combined sexes for the Shuar and *Colono* populations compared to a US reference population. Shuar exhibit consistently higher bone density values with age when compared to both populations.

Table 3.1. Anthropometric and QUS data by group, sex, and menopausal status (mean [SD]).

	Shuar	Colono	<i>p</i>
<i>Sexes Combined</i>			
N	227	261	
Age (years)	34.97 (14.72)	47.43 (19.53)	**
Height (cm)	152.09 (7.23)	153.83 (10.91)	*
Weight (kg)	58.49 (10.50)	64.72 (12.50)	**
BMI (kg/m²)	25.19 (3.51)	27.28 (5.18)	**
BUA (dB/MHz)	82.0 (19.3)	69.4 (19.0)	**
SOS (m/s)	1571.4 (33.2)	1545.1 (32.9)	**
hBMD	0.600 (0.131)	0.499 (0.130)	**
<i>Males</i>			
N	91	104	
Age (years)	37.26 (14.76)	47.30 (20.15)	**
Height (cm)	157.99 (6.27)	161.13 (8.45)	*
Weight (kg)	63.19 (9.32)	69.49 (13.38)	**
BMI (kg/m²)	25.20 (2.73)	26.90 (5.99)	*
BUA (dB/MHz)	87.5 (19.54)	74.1 (20.38)	**
SOS (m/s)	1574.4 (34.65)	1550.8 (33.96)	**
hBMD	0.622 (0.137)	0.527 (0.134)	**
<i>Females</i>			
N	136	157	
Age (years)	33.44 (14.54)	47.52 (19.18)	**
Height (cm)	148.32 (4.9)	149.00 (9.6)	**
Weight (kg)	55.51 (10.14)	61.57 (10.8)	**
BMI (kg/m²)	25.19 (3.95)	27.53 (4.6)	**
BUA (dB/MHz)	78.4 (18.4)	66.2 (17.43)	**
SOS (m/s)	1569.4 (32.14)	1541.3 (31.7)	**
hBMD	0.586 (0.126)	0.481 (.124)	**
<i>Pre-menopausal women</i>			
N	118	81	
Age (years)	29.52 (10.55)	31.75 (10.44)	<i>Ns</i>
Height (cm)	148.86 (11.31)	152.34 (6.07)	**
Weight (kg)	56.17 (10.57)	61.76 (11.13)	**
BMI (kg/m²)	25.33 (4.51)	26.58 (4.43)	*

Table 3.1. (continued)

BUA (dB/MHz)	80.6 (15.3)	74.3 (15.3)	*
SOS (m/s)	1574.2 (27.9)	1559.2 (23.6)	**
hBMD	0.605 (0.110)	0.548 (0.095)	**
<i>Post-menopausal women</i>			
N	13	76	
Age (years)	61.60 (11.35)	64.32 (9.68)	<i>Ns</i>
Height (cm)	144.66 (6.23)	145.40 (11.31)	<i>Ns</i>
Weight (kg)	49.63 (8.75)	61.37 (10.57)	**
BMI (kg/m²)	23.57 (2.85)	28.56 (4.51)	**
BUA (dB/MHz)	58.7 (21.2)	57.5 (15.3)	<i>Ns</i>
SOS (m/s)	1532.7 (35.8)	1522.2 (27.9)	<i>Ns</i>
hBMD	0.439 (0.144)	0.409 (0.110)	<i>Ns</i>

Ns = not significant

* $p < 0.05$; ** $p < 0.001$ (significant difference between ethnic groups)

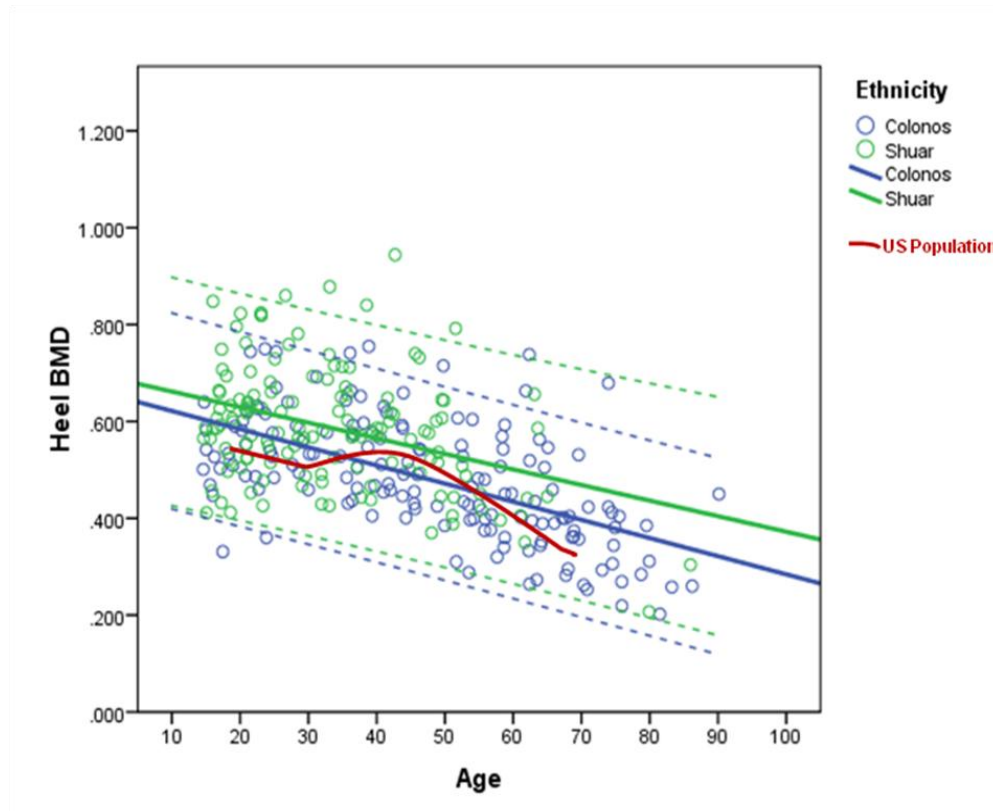


Figure 3.1. Scatter plot of hBMD for Shuar and *Colono* combined sexes compared to values from a US reference population with linear best of fit line. Dotted lines indicate 99% confidence intervals.

Shuar

Overall, Shuar males exhibit significantly higher bone density parameters than their female counterparts for all bone health measures except SOS (BUA: $p < 0.001$; hBMD: $p < 0.05$). Table 3.2 illustrates Shuar anthropometric and calcaneal QUS results by age cohort and sex. For both sexes, all bone health measures are highest in the 21-30 age cohort and are progressively lower in each subsequent age group. One exception is found in males from the 51-60 year old cohort where all QUS parameters are greater than among males in the preceding 41-50 age interval. This difference may be attributed to small sample size in the older age interval, biased by one outlier with extremely high bone density (BUA: 164.2 dB/MHz; SOS: 1712.0 m/s hBMD: 1.177).

Figure 3.2 (a-c) presents clustered scatterplots of BUA, SOS, and hBMD by age for Shuar females and males. In both sexes, there is a significant inverse relationship between age and each of the QUS parameters ($p < 0.001$). From ages 21 to 86, females show a cumulative decrease of 34% in BUA, 3.1% in SOS, and 32% in hBMD. Although males begin with higher peak bone mass values than females, Shuar males from ages 21 to 80 show greater total decrease in bone density: 37% in BUA, 3.8% in SOS, and 38% in hBMD. Among females, the most pronounced decrease in BUA, SOS, and hBMD by age group occurs after 50 years of age, whereas among males the greatest decline is seen among individuals over 60.

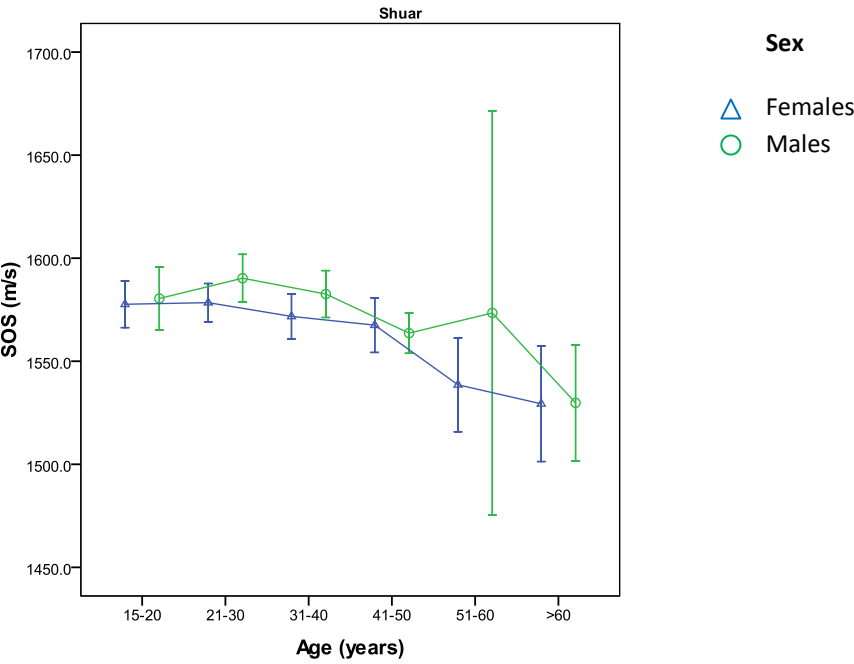
Table 3.2. Age-specific anthropometric and QUS parameters by ethnic group and sex (mean [SD]).

Ethnic Group	Age	Sex	n	Height (cm)	Weight (kg)	BMI (kg/m²)	BUA (dB/MHz)	SOS (m/s)	hBMD
Shuar	15-20	Female	28	148.77 (4.94)	50.52 (5.03)	22.80 (1.77)	77.5 (17.0)	1577.6 (29.2)	0.604 (0.115)
		Male	12	154.83 (11.6)	54.05 (12.32)	22.20 (2.56)	77.9 (15.6)	1580.4 (23.9)	0.612 (0.098)
	21-30	Female	38	149.63 (4.51)	55.49 (11.52)	24.81 (4.64)	83.0 (17.2)	1578.4 (28.3)	0.620 (0.112)
		Male	21	158.93 (5.16)	62.53 (6.22)	24.69 (1.21)	95.7 (12.8)	1590.3 (25.4)	0.684 (0.096)
	31-40	Female	30	149.31 (3.63)	61.44 (9.44)	27.53 (3.81)	80.4 (17.9)	1571.8 (29.2)	0.599 (0.117)
		Male	25	158.38 (3.99)	66.16 (5.97)	26.35 (1.85)	96.1 (15.7)	1582.6 (27.5)	0.665 (0.110)
	41-50	Female	23	146.86 (4.69)	57.66 (10.44)	26.60 (3.61)	81.2 (14.7)	1567.5 (30.6)	0.591 (0.114)
		Male	19	159.07 (4.02)	68.14 (9.84)	26.85 (3.13)	83.3 (11.5)	1563.6 (20.3)	0.583 (0.077)
	51-60	Female	9	145.79 (5.57)	55.71 (7.54)	26.15 (2.57)	67.4 (19.8)	1538.6 (29.7)	0.476 (0.125)
		Male	5	162.40 (4.58)	63.97 (3.56)	24.33 (2.25)	86.1 (44.4)	1573.4 (79.0)	0.615 (0.319)
	> 60	Female	8	143.14 (7.06)	45.79 (6.32)	22.23 (1.20)	56.4 (23.1)	1529.4 (33.5)	0.424 (0.145)
		Male	9	153.38 (4.58)	58.30 (10.45)	24.68 (3.58)	66.5 (20.9)	1529.8 (36.6)	0.452 (0.149)
Colono	15-20	Female	16	151.76 (5.66)	54.73 (13.0)	23.66 (5.04)	66.7 (9.2)	1557.9 (22.5)	0.525 (0.732)
		Male	13	164.41 (7.72)	61.18 (10.39)	22.68 (3.94)	69.6 (21.6)	1556.5 (30.4)	0.535 (0.135)
	21-30	Female	22	153.96 (5.68)	62.23 (10.22)	26.20 (3.77)	79.5 (16.3)	1563.9 (24.6)	0.574 (0.101)
		Male	13	165.67 (5.85)	68.95 (8.85)	25.17 (3.47)	86.1 (21.0)	1583.9 (36.7)	0.641 (0.141)
	31-40	Female	22	152.52 (6.52)	63.86 (9.32)	27.57 (4.55)	77.3 (15.1)	1562.8 (24.2)	0.565 (0.981)
		Male	14	159.98 (14.24)	72.46 (11.99)	29.49 (10.98)	91.0 (16.8)	1580.9 (24.7)	0.647 (0.106)

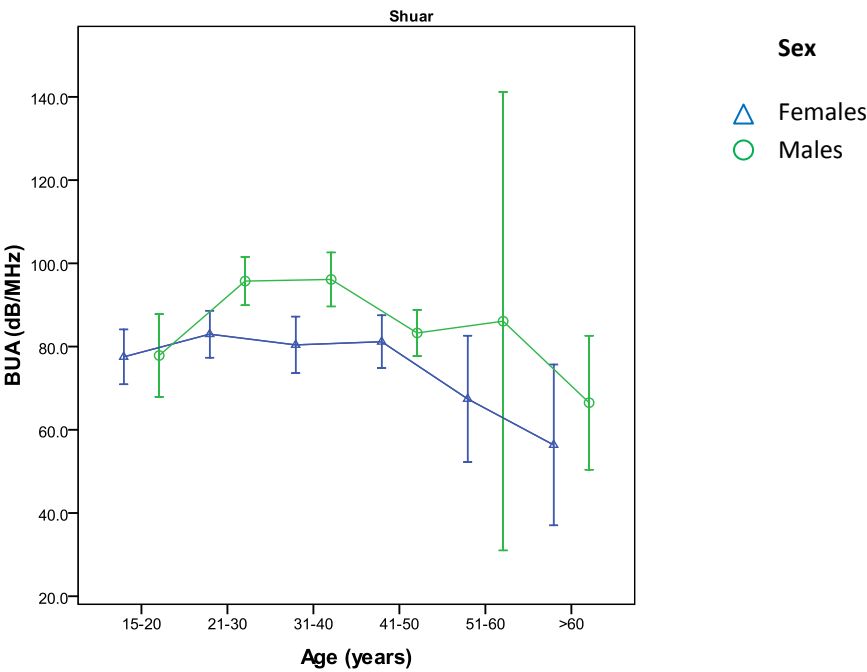
Table 3.2. (continued)

41-50	Female	23	149.96 (6.91)	63.89 (10.37)	28.30 (3.19)	70.7 (14.5)	1549.4 (21.5)	0.513 (0.089)
	Male	21	163.19 (5.39)	80.16 (14.90)	30.04 (5.03)	73.8 (15.5)	1545.5 (19.5)	0.511 (0.086)
51-60	Female	27	145.48 (17.75)	67.60 (10.39)	30.65 (4.44)	63.1 (14.0)	1530.9 (22.6)	0.448 (0.092)
	Male	13	160.40 (6.42)	68.86 (13.31)	26.70 (4.48)	70.9 (12.1)	1538.8 (19.6)	0.488 (0.754)
> 60	Female	47	145.58 (5.67)	58.04 (9.34)	27.41 (4.31)	54.3 (16.1)	1516.9 (30.3)	0.386 (0.118)
	Male	30	157.23 (8.45)	65.10 (13.38)	26.28 (4.0)	64.8 (21.0)	1528.8 (30.6)	0.448 (0.123)

a)



b)



c)

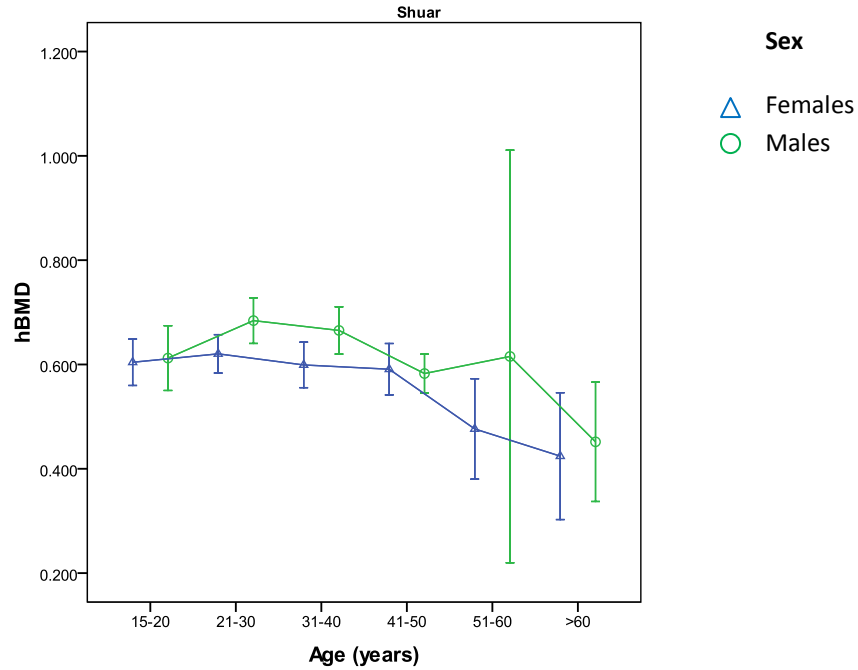


Figure 3.2 (a-c). Sex differences in the Shuar for SOS (a), BUA (b), and hBMD (c) values based on age. Values represent means for the age category with 99% confidence intervals.

In the stepwise multiple linear regression analyses (Table 3.3), weight was a less important factor for calcaneal QUS than age in both Shuar men and women. In men, all QUS parameters were significantly related to age (BUA: $\beta = -0.543$, $p < 0.001$; SOS: $\beta = -1.288$, $p < 0.001$; hBMD: $\beta = -0.005$, $p < 0.001$) and BMI (BUA: $\beta = 2.768$, $p < 0.001$; SOS: $\beta = 2.682$, $p < 0.05$; hBMD: $\beta = 0.014$, $p < 0.01$). However, in women, SOS was dependent only on age ($\beta = -0.889$, $p < 0.001$), while BUA was dependent on age ($\beta = -0.284$, $p < 0.05$), height ($\beta = 0.740$, $p < 0.05$), and BMI ($\beta = 1.288$, $p < 0.05$). Height was not a predictive variable for hBMD, although age ($\beta = -0.003$, $p < 0.001$) and BMI ($\beta = 0.006$, $p < 0.05$) were significant. Weight alone was not a significant predictor for any QUS value among the Shuar.

Table 3.3. Stepwise multivariate linear regression model between QUS values and age (years), weight (kg), height (cm), and BMI (kg/m²).

Ethnicity	QUS	Stepwise multiple regression model*	R ²	Adjusted R ²	p
Shuar	Male				
	BUA	38.04-0.543*age+2.768*BMI	.224	.204	.000
	SOS	1554.9-1.288*age+2.682*BMI	.269	.250	.040
	hBMD	0.443-0.005*age+0.014*BMI	.252	.233	.007
	Female				
	BUA	-53.7-0.284*age+0.74*height +1.288*BMI	.192	.172	.029
	SOS	1560.0-0.889*age	.159	.152	.000
	hBMD	0.56-0.003*age+0.006*BMI	.151	.138	.042
	Total				
	BUA	-63.1-0.35*age +0.8*height+1.431*BMI	.236	.224	.000
	SOS	1497.5-0.94*age+0.705*height	.198	.190	.016
	hBMD	0.03-0.003*age+0.004*height+0.006*BMI	.202	.191	.017
Colono	Male				
	BUA	62.7-0.289*age+0.363*weight	.143	.126	.012
	SOS	1590.2-0.832*age	.244	.236	.000
	hBMD	0.175-0.003*age+0.003*height	.226	.210	.049
	Female				
	BUA	68.1-0.486*age+0.773*BMI	.282	.272	.005
	SOS	1588.9-1.001*age	.370	.366	.000
	hBMD	0.567-0.004*age+0.004*BMI	.357	.348	.043
	Total				

Table 3.3. (continued)

BUA	$-29.0 - 0.321 * \text{age} + 0.609 * \text{height} + 0.726 * \text{BMI}$.253	.244	.000
SOS	$1489.7 - 0.837 * \text{age} + 0.617 * \text{height}$.335	.330	.001
hBMD	$0.195 - 0.003 * \text{age} + 0.002 * \text{height} + 0.001 * \text{weight}$.322	.314	.039

*QUS parameters that are not in the regression equation are not significant in the model.

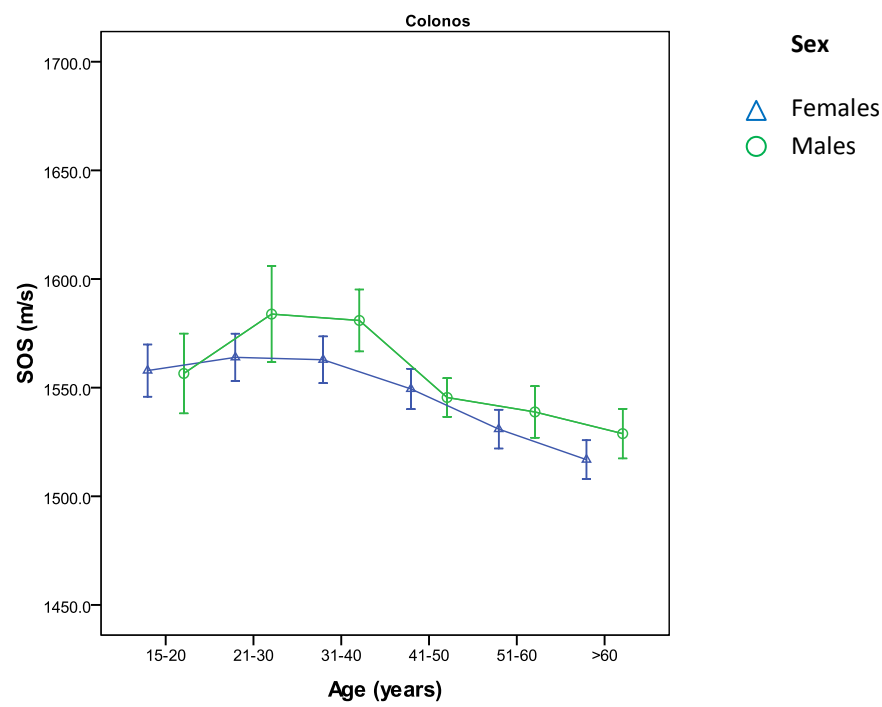
Colonos

Among *Colonos*, sex differences for all QUS measures are more pronounced than among Shuar, and higher bone density measures are found among *Colono* males than females (BUA: $p = 0.001$; SOS: $p < 0.05$; hBMD: $p < 0.01$). Analysis of QUS values by age cohort (Table 3.2) shows that similar to Shuar, *Colono* females display the highest ultrasound measures in the 21-30 year old cohort, with lower values in each successive age cohort. *Colono* males also have highest SOS in the 21-30 year old cohort, although 31-40 year olds exhibit the highest BUA and hBMD values. Among *Colono* females, as with their Shuar counterparts, the most pronounced reduction in BUA, SOS, and hBMD is apparent after 50 years of age. In contrast, for *Colono* males, all QUS parameters are sharply lower after 40 years of age.

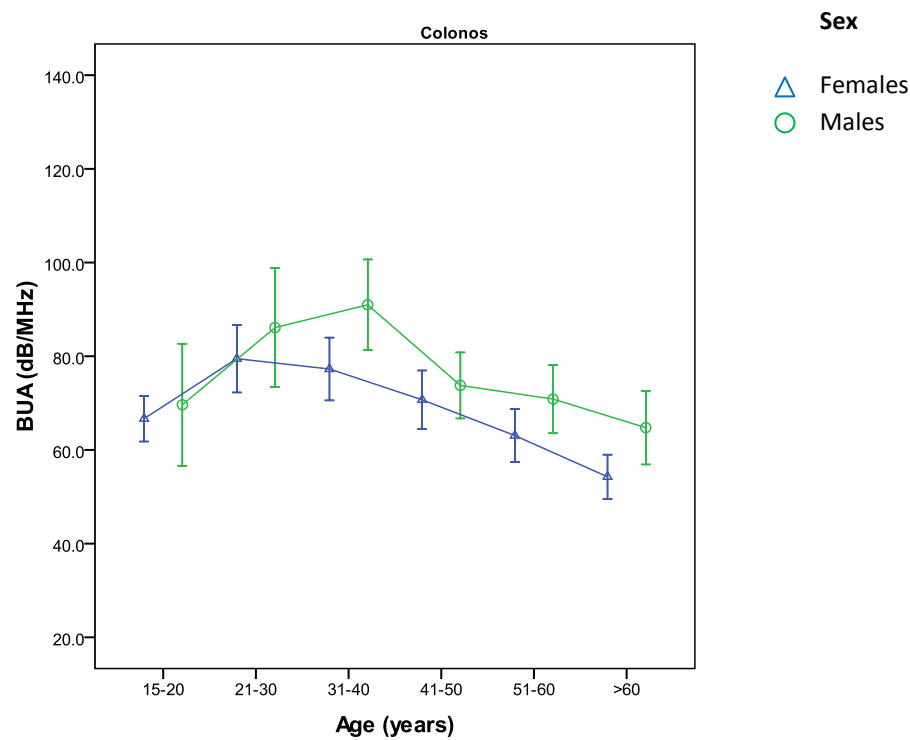
Clustered scatterplots for BUA, SOS, and hBMD by age for *Colono* females and males (Figure 3.3 [a-c]) show a significant negative relationship between age and QUS parameters for both sexes ($p < 0.05$). Among *Colono* females between 21 and 90 years old, total decline in QUS values with age are 38%, 3.0%, 36% for BUA, SOS, and BMD respectively; among *Colono* males 21 to 91 years old, there is a 31% decrease in BUA, 3.5% decrease in SOS, and 36% decrease in hBMD.

The stepwise multiple regression analysis in Table 3.3 shows that age is the sole predictive factor for SOS in *Colono* males ($\beta = -0.832$, $p < 0.001$) and females ($\beta = -1.001$, $p < 0.001$). In *Colono* females, BUA and hBMD were a function of both age (BUA: $\beta = -0.486$, $p < 0.001$; hBMD: $\beta = -0.004$, $p < 0.001$) and BMI (BUA: $\beta = 0.773$, $p < 0.01$; hBMD: $\beta = 0.004$, $p < 0.05$). In *Colono* males, BUA was a function of age

a)



b)



c)

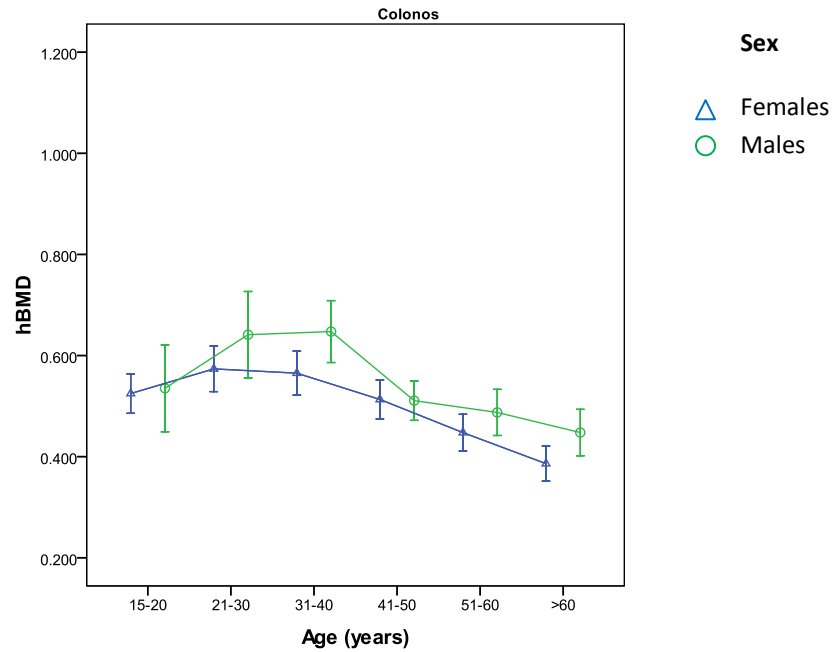


Figure 3.3 (a-c). Sex differences in the *Colonos* for SOS (a), BUA (b), and hBMD (c) values based on age. Values represent means for the age category with 99% confidence intervals.

($\beta = -0.289$, $p < 0.01$) and weight ($\beta = 0.363$, $p < 0.05$) while hBMD was dependent on age ($\beta = -0.003$, $p < 0.001$) and height ($\beta = 0.003$, $p < 0.05$).

Reproductive Status

Table 3.4 shows descriptive statistics of QUS and anthropometric measures for Shuar and *Colono* females who were neither pregnant nor lactating at the time of the study. Among the Shuar, nine participants (4%) were pregnant, thirty (13.2%) were lactating, and one (0.4%) was both pregnant and lactating at the time of the study. Half of Shuar women aged 21-30 years reported being either pregnant and/or lactating at the time

of the bone scan; all women who reported being in a lactating state were from this age cohort.

Among *Colono* women, four (1.5%) were pregnant and eight (3.1%) were lactating at the time of the study. Student's t-tests were performed comparing QUS parameters between Shuar and *Colono* women based on reproductive status--that is, whether they were pregnant, lactating, both, or neither. Significant ethnic group differences in bone health measures were found only for non-pregnant/non-lactating participants, with Shuar women showing significantly higher bone density than *Colono* women for BUA, SOS, and hBMD ($p < 0.001$).

Table 3. 4. Descriptive statistics for non-pregnant and non-lactating women by ethnic group (mean [SD]).

	Shuar (n=96)	Colono (n=145)	<i>p</i>
Age (years)	35.81 (15.37)	48.50 (19.17)	**
Height (cm)	148.17 (5.19)	148.84 (9.68)	<i>Ns</i>
Weight (kg)	55.30 (9.17)	61.51 (10.92)	**
BMI (kg/m²)	25.13 (3.55)	27.54 (4.61)	**
BUA (dB/MHz)	77.5 (19.4)	64.8 (16.7)	**
SOS (m/s)	1566.4 (32.6)	1539.1 (30.8)	**
hBMD	0.576 (0.130)	0.472 (0.119)	**

Ns = not significant; ** $P < 0.001$ (significant difference between ethnic groups)

Menopausal Status

Analysis by menopausal status revealed that among pre-menopausal women there are significant ethnic differences for all QUS parameters ($p < 0.001$). Menopausal status was not reported for four Shuar women (aged 52-55 years old) and thus could not be

included in these analyses. Thirteen Shuar (9.6%) and seventy-six *Colono* women (48.4%) were post-menopausal at the time of the study (Table 3.1). Among *Colono* and Shuar post-menopausal women, there is no significant difference in BUA, SOS, and hBMD values, which suggests that the highest degree of variation in bone health between ethnic groups is present in the pre-menopausal lifespan.

DISCUSSION

The main purpose of this paper was to present population-specific normative data for bone density in a non-industrialized, natural fertility population from a developing country. Minimal bone health data are available from these contexts and this has inhibited efforts to quantify the burden of osteoporosis in developing nations, especially among rural groups (Handa et al., 2008; Woolf and Pflieger, 2005). Additionally, bone health information from diverse populations is necessary to determine the degree to which normative data from developed countries may reflect sub-optimal bone density levels. The skeletal system and mechanisms of bone turnover evolved under subsistence-based, natural fertility conditions with dietary, activity, and reproductive patterns that likely produce a vastly different representation of normative bone health than data garnered from industrialized, urban populations. The paucity of epidemiological data from non-urbanized populations in developing countries can be partly attributed to the lack of diagnostic resources in these regions. However, portable QUS technology now enables the collection of BMD data from remote field settings. Calcaneal ultrasonometers, in particular, are increasingly being utilized to obtain data from populations worldwide and to compare this information with existing reference populations. The current study

focused on the calcaneus in order to maximize future inter-population comparisons, and because the calcaneus is a weight-bearing site rich in trabecular bone that is therefore an ideal single-site measure of bone density parameters.

The present study found that *Colonos* are taller, heavier, and have greater BMI values than the Shuar. However, despite this result, and the finding that age-related bone loss occurs in both groups, values for BUA, SOS, and hBMD are consistently higher in the Shuar population. Higher peak bone mass among the Shuar may partially explain the higher QUS values exhibited in all age groups when compared to *Colonos*. Although both ethnic groups participate to a varying degree in an agriculture-based subsistence regime, population-specific variability in dietary and lifestyle factors may account for the significant QUS differences reported here. Individual and community-level variation in diet, activity levels, and smoking/drinking behaviors have been reported elsewhere for these populations (see Chapter V) (Liebert et al., 2010) and future research will explore the specific effects of these factors on Shuar and *Colono* bone health. Genetic differences between ethnic groups have been established (e.g., African-American versus Caucasian-American populations) and these factors may also explain the lower BMD documented across *Colono* age cohorts (e.g., Anderson and Pollitzer, 1994; Hinkley et al., 2004; Naganathan et al., 2002; Thomas, 2007).

The most pronounced ethnic group difference in QUS values was documented between Shuar and *Colono* pre-menopausal women who were not pregnant or lactating. In post-menopausal women, no significant difference was found, suggesting that menopause onset has similar negative effects on bone integrity across ethnic groups. For both Shuar and *Colono* females, the lowest bone health values were found after 50 years

old. This finding is consistent with other studies (e.g., Pluskiewicz, 1998; Trovas et al., 2009) that show that the most profound bone loss occurs for similar age intervals, typically at the onset of menopause.

We also observed the effect of age and body size (height, weight, and BMI) on QUS by ethnic group using a stepwise multiple regression analysis. Age is a consistent predictor of all QUS parameters, with declining values in older age groups. Among female cohorts, the second most reliable predictor of higher QUS values is greater BMI. In the current study, the highest QUS values among females are found among 21-30 year olds, which coincides with a period when peak bone mass is generally achieved. It is also important to recognize that half of the Shuar women in this age cohort reported being pregnant and/or lactating which may be variously contributing to calcaneal QUS values. However, when comparing females who are not pregnant and lactating, differences between the two ethnic groups are maintained.

Males in both populations show consistently higher QUS parameters than females and, similar to women, calcaneal QUS values are a primary function of age. BMI also predicts SOS, BUA, and hBMD among Shuar males. However, in *Colono* males QUS values, specifically BUA and hBMD, are not a function of BMI and are better predicted by the independent variables, weight and height, respectively. Furthermore, one of the earliest and more pronounced declines in QUS values occurs in *Colono* males after 40 years old. In contrast, Shuar male QUS data do not show this steep decline until after 60 years old, more typical of the pattern documented in other studies (e.g., Takeda et al., 1996; Liu et al., 2006). While the *Colono* result does not follow the typical trend, a similar decrease in heel BMD is reported among Roma males from Croatia after 35 years

old (Skarić-Jurić et al., 2006). Potential causes for this apparently early decrease in QUS parameters are not addressed in the Roma study, but the authors state that increased sedentism and diminutive body size are possible explanations for the low BMD characteristic of the entire group. The reason for an early decline among *Colono* males is unclear, but may be related to sampling biases. A larger sample size for each age interval is necessary to address this issue.

Shuar men and women both display comparatively greater QUS parameters than other populations whose reference values were determined using the same calcaneal QUS instrument. In Korea, in a study by Rhee and colleagues (2009), all QUS values are lower than the Shuar population but higher than the *Colono* group. In a German study on an exclusively female cohort (Alenfeld et al., 2002), BUA and SOS parameters are again lower than Shuar women but higher than *Colono* females in our study for similar age intervals. Normative data from the Croatian Roma population discussed earlier show markedly lower hBMD values when compared to Ecuadorian data; however this disparity is minimized after 40 years old for both female cohorts and for *Colono* males. Shuar males maintain among the highest ultrasound values for any comparative population presented here. From this cross-study comparison, the Shuar exhibit some of the highest QUS values, while *Colonos* as a group show slightly lower bone density measures than other global reference populations. The high values among the Shuar are likely related to a suite of lifestyle and activity patterns that are reflective of greater dependency on a mixed agriculture and subsistence-based economy than the *Colono* participants.

Study Limitations

The present study has several important limitations. While field-based research is a necessary and critical approach to gathering information from populations in remote settings, this type of research presents issues when aiming to establish a controlled setting for data collection. One study limitation is that participant sample sizes for certain age intervals are small, particularly among older cohorts, an issue that is more pronounced in the Shuar group. The small sample sizes and non-random selection of participants within age categories may inhibit our interpretations of the age effects on BMD that have been reported for other reference populations. Also, participants in the current study were recruited from a sample pool of individuals who may have been inclined to seek out health information provided by this study. Although this may inherently bias the dataset towards individuals who demonstrate greater health-related issues, this recruitment strategy is not atypical for field-based research in remote settings. Also, as previously mentioned, normative data on bone health generally exclude pregnant and lactating females, because the modifiable (e.g., diet, activity) and non-modifiable (e.g., hormones) factors affecting bone integrity are constantly fluctuating. Yet in many developing countries most females are pregnant and lactating for much of their reproductive life spans, so inclusion of this demographic was an effort to present a normative representation of bone health in these two groups. This inclusion, however, can present problems with cross-population comparative studies. Further research on bone health in developing countries is necessary in order to determine the most effective approach to remedying this issue. Finally, cross-sectional data prevent us from making definitive

statements on age-related changes in skeletal health, but is a necessary first step in determining patterns of bone loss.

CONCLUSIONS

The current study established a preliminary dataset for three calcaneal QUS parameters in the Shuar and non-Shuar *Colono* populations of Amazonian Ecuador. It investigated ethnic and sex differences for the effect of age and body size variables on QUS parameters. The study also addressed relevant issues in gathering bone health data in a remote field-based setting within a developing country. The Shuar and *Colonos* represent two populations from the same geographical region who have a range of dependency on mixed agricultural and subsistence-based economy. Differences in QUS values between the two groups may be attributed to genetics as well as to individual- and community-level variation in diet and lifestyle. Future research will examine the specific ways in which dietary composition and lifestyle behaviors contribute to Shuar and *Colono* bone health profiles.

BRIDGE TO CHAPTER IV

This chapter has examined bone health among the Shuar and *Colonos* and found that Shuar bone health values are significantly higher than that of *Colonos* and a US reference population. While Chapter III presents normative data for Ecuadorian populations, Chapter IV examines the association between various reproductive variables and skeletal health in Shuar females from four rural Upano Valley communities. Specifically, the study applies a retrospective and cross-sectional design to examine

BMD and self-report reproductive patterns among 131 Shuar women aged between 14 and 86 years old. Chapter IV also presents fertility information for the Shuar participants, data that are not previously reported elsewhere for this indigenous population. The following chapter includes unpublished co-authored material.

CHAPTER IV

REPRODUCTIVE EFFECTS ON SKELETAL HEALTH

IN SHUAR WOMEN OF AMAZONIAN ECUADOR:

A LIFE-COURSE PERSPECTIVE

This chapter contains unpublished co-authored material and has been prepared with some assistance from Snodgrass JJ, Liebert MA, Cepen TJ, and Sugiyama LS. All data collection for bone density measures, data analyses, and write-up were conducted by the author of the dissertation. Additional anthropometrics including stature and weight were gathered with the assistance of Liebert, and Cepen. Sugiyama and Snodgrass provided editorial assistance.

INTRODUCTION

Low bone mineral density (BMD) is a multi-factorial, chronic condition that can progress into the debilitating disease, osteoporosis. The factors contributing to bone loss are incompletely understood, which is surprising given the enormous health effects of osteoporosis. Known as the “silent epidemic,” osteoporosis is now identified as a major public health problem that affects populations worldwide (Bartl and Frisch, 2004; International Osteoporosis Foundation [IOF], 2010). In the United States, an estimated 30 million people suffer from osteoporosis, with an equally high prevalence in Europe and other developed countries (IOF, 2010). Of the 1.3 million fractures reported annually among U.S. individuals over 45 years old, at least 70% can be attributed to low bone density (Bartl and Frisch, 2004). The problem is more severe for women, with

osteoporotic fractures affecting more women than heart attacks, strokes, and all female cancers combined. According to the International Osteoporosis Foundation (2010), an estimated 54% of postmenopausal Caucasian women in the U.S. have low bone density (osteopenia) and 30% have osteoporosis.

Although advancing age is a key contributor to bone loss in both sexes (Frost, 2003), a number of genetic and specific environmental factors have been implicated in osteoporosis. While peak bone mass is generally accepted to be strongly influenced by genetic factors (e.g., Ferrari et al., 1998), these factors only explain a small proportion of the variation in individual BMD (Cooper et al., 2001), and age-related bone loss appears to be more sensitive to the effects of individual lifestyles (Jouanny et al., 1995; Krall and Dawson-Hughes, 1993). In particular, physical activity, especially of the weight bearing variety, has been linked to accretion of new bone and maintenance of current bone via muscle contractions that stimulate osteoblastic processes (Dargeant-Molina et al., 1996; Kemper et al., 2000; Proctor et al., 2000). Numerous studies have also documented the contribution of several nutritional factors to bone mass, including calcium, vitamin C, and vitamin D, whether through increasing intestinal calcium absorption or by stimulating bone formation (Anderson et al., 2004; Bunker, 1994; Dawson-Hughes, 2004). Additional components of lifestyle that may deleteriously impact bone mass include caffeine, alcohol, and excessive protein consumption, as well as a history of smoking. Because of the multi-etiological nature of bone loss, establishing a causal relationship with bone loss for any one of these factors is challenging (Bunker, 1994; Hernandez-Avila et al., 1993; Lazenby, 1997; Sampson, 2002).

Among women, the skeletal effects of age and lifestyle are influenced by reproductive factors, primarily as a result of the mother's role in providing calcium to the growing fetus during pregnancy and to the neonate during lactation. Clinical and epidemiological studies suggest that reproductive factors such as parity, patterns of breastfeeding, and age at first pregnancy can significantly impact bone mass changes throughout the lifetime as a result of heightened mobilization of calcium and hormone shifts, especially estrogens (see review below) (Agarwal and Stuart-Macadam, 2003; Galloway, 1988; Galloway, 1997; Guyton and Hall, 2011; Pacifici, 2007; Pearce, 2006). Unraveling the skeletal effects of variation in female life history is complicated by the inter-relatedness (i.e., non-independence) of various developmental and reproductive stages. Each reproductive factor is not an isolated event occurring without connection to the series of developmental stages (Agarwal and Glencross, 2010; Leidy, 1996; Pike, 2001). The timing of pregnancy and duration of lactation, for instance, are potentially linked to other factors such as age at menarche and inter-birth interval, which, themselves, are shaped by ecological and environmental variables. The influence on skeletal health of various developmental and reproductive factors and their potentially cumulative effects across the lifespan remains unclear despite decades of research. However, this information is critical for the development of clinical guidelines and public health policies for osteoporosis prevention.

One of the most important limitations to understanding the effects of reproductive factors on skeletal health is that few studies to date have examined maternal bone status in non-Western, non-clinical populations. Even fewer data are available for subsistence-based, natural fertility groups. This is unfortunate since studies from these populations

may allow insight into the complexities of bone loss without the confounding effects of sedentary lifestyles and Westernized diets that characterize industrialized groups. In addition, fertility patterns in past and present subsistence populations are more characteristic of those that would have existed for most of our evolutionary past. As a result, research on skeletal health in these populations has the potential to improve our current understanding of the conditions under which reproductive trade-offs with skeletal health evolved. The following section reviews the published literature on the effects of reproduction on skeletal health.

REPRODUCTION AND BONE HEALTH: LITERATURE REVIEW

The influence of reproductive factors on skeletal health is primarily shaped by fluctuations in sex steroids such as estrogens and progesterone (Galloway, 1997). Estrogens play a central role during the course of the female lifespan by influencing bone and collagen formation, and increasing intestinal absorption and retention of calcium. These hormones also inhibit bone remodeling by reducing the number of osteoclasts and osteoblasts that are responsible for bone resorption and formation, respectively (Agarwal and Stuart-Macadam, 2003; Galloway, 1997; Guyton and Hall, 2011). Similar benefits are afforded by progesterone, which has shown to promote bone accrual through proliferation of osteoblastic activity (Galloway, 1997). Additional hormones, including parathyroid hormone, vitamin D, and calcitonin help to maintain the equilibrium of the central calcium pool, and during various reproductive phases the levels of these hormones undergo fluctuations that invariably contribute to bone integrity (Bartl and Frisch, 2004; Dawson-Hughes, 2004; Forwood, 2001). The female reproductive factors

most often linked to bone integrity in clinical and epidemiological literature are: age at menarche; age at first pregnancy; patterns of breastfeeding (e.g., intensity and duration); and age at menopause.

Age at Menarche

Several studies have documented associations between age at menarche and bone density, as well as with risk of osteoporosis later in life. A later age at menarche may be related to a heightened risk of osteoporosis in the postmenopausal period, whereas an earlier menarche age may reduce this probability by increasing the peak bone mass achieved earlier in life. Earlier menarcheal age may have a stimulating effect on the early development of bone by increasing the osteoblastic activity that coincides with estrogens, thereby establishing higher bone density values at peak bone mass attainment (Jaffe and Dell'Acqua, 1985). Studies have documented the relationship between better skeletal health and early menarcheal age for pre- and postmenopausal women. Ito and colleagues (1995) found that delayed menarche, particularly when it occurred after 16 years of age, was associated with lower long-term bone mass. Similarly, among postmenopausal women, Roy and colleagues (2003) observed that participants with an age at menarche after age 16 were eight times more likely to experience vertebral fractures.

Two explanations have been posited for the relationship between earlier age at menarche and higher bone mass. First, delayed menarche results in lower lifetime exposure to estrogens and, as described earlier, these hormones are typically associated with bone accumulation. Second, early menarche is related to larger body size and greater adiposity which, in turn, can increase bone quantity (Eastell, 2005). For these reasons, menarcheal age may be more strongly related to postmenopausal bone mass than age at

menopause (e.g., Gerdhem and Obrant, 2004; Roy et al., 2003; Silman, 2003), although other studies have shown no such relationship (Ito et al., 1995; Ozdemir et al., 2005; Sioka et al., 2010; Varenna et al. 1999).

Age at First Parturition

A later age at first birth has been linked with better skeletal health in both pre- and postmenopausal life. Bone density typically continues to increase into the mid-twenties when peak bone mass is achieved, but pregnancy and lactation during this time have been shown to disrupt bone formation and negatively influence long-term bone mass (Hayslip et al., 1989; Kent et al., 1990, 1993; Sowers et al., 1993). A study by Schnatz and colleagues (2010) demonstrates that women who were 27 years or older at the time of their first pregnancy had a significantly lower risk of developing osteoporosis in postmenopausal life. In addition to long-term effects, a younger age at first pregnancy may have negative impacts during the pre-menopausal years. In several studies, women who were younger at first parturition (<20 years old) demonstrated an impaired ability to gain in height when compared to other women, suggesting disruptions during early bone development (Allal et al, 2004; Gigante et al., 2006; Sear et al., 2004). These findings for height have been documented for Western, industrialized groups and non-Western, rural populations alike. However, the effect of the timing of first pregnancy on bone mass is not consistent across studies with some research finding no significant relationship at all (e.g., Sowers et al., 1985).

The relationship between age at first parturition and skeletal health is rooted in the fundamental nature of life history trade-offs between reproduction and growth. Following

the basic tenets of this theoretical approach, in a resource-limited environment, energy and energetic resources are allocated to one physiological demand at the expense of others. The high costs of pregnancy and the subsequent period of lactation generally necessitates directing energy and nutrients away from other somatic functions. Devotion of energy to reproduction can theoretically limit allocation to growth, thereby resulting in trade-offs between early parturition and longitudinal skeletal gain.

Pregnancy and Parity

The literature on the long-term effects of pregnancy and parity do not show consistent results. Several studies comparing bone status of nulliparous and multiparous pre- and perimenopausal women have demonstrated that nulliparous females have lower bone values (e.g., Sowers et al., 1992). Similarly, Forsmo and colleagues (2001) found that among early postmenopausal women nulliparity predicted lower bone density values. However, several studies have reached an opposite conclusion, documenting a negative association between bone density and multiple pregnancies (Allali et al., 2007; Gur et al., 2003). Others have not found any significant associations with number of pregnancies (Ensom et al., 2002; Hillier et al., 2003; Lenora et al., 2009; Melton et al., 1993). Even in studies of groups characterized by repeated and closely spaced pregnancies, such as among Omani women (Bererhi et al., 1996) and multiparous Finish American women (Henderson et al., 2000), no significant relationship was documented between number of children and bone density.

The inconsistencies in the literature are largely due to the multiple, complex pathways of calcium turnover during pregnancy. For example, pregnancy can decrease

maternal bone density through the heightened calcium demand associated with fetal growth. During pregnancy, the growing fetus requires approximately 25-30 grams of calcium from the mother to develop its skeleton (Holmberg-Marttila et al., 1999); this marks a critical period during which significant change in maternal serum calcium concentration occurs. However, despite this draw on calcium, the maternal skeleton typically exhibits an increase in bone mass during pregnancy, which is most likely due to increased estrogen levels that inhibit bone loss, and in some cases, promote bone accretion (e.g., Lees et al., 1998). Pregnancy-related weight gain (i.e., increased loading), as well as greater intestinal calcium absorption, also contributes to the protective effect that being pregnant has on bone mass (Nguyen et al., 1995; Streeten et al., 2005). It is unclear whether sheer offspring quantity may shape bone density, and the influence of pregnancy and parity may be obscured by the effects of lactation.

Lactation

The duration, timing, and intensity of lactation all appear to influence bone density, yet some studies show a protective effect of breastfeeding (Hreschchyshyn et al., 1988; Pearce, 2006), and others show a negative impact (Affinito et al., 1996; Drinkwater and Chestnut, 1991; Kent et al., 1993; Lamke et al., 1977; Sowers, 1996). These seemingly contradictory results appear to reflect the length of time that participants spent lactating, and how the studies were designed in order to determine the effects of lactation on bone health. For instance, several longitudinal studies that document changes in bone status during post-partum and weaning periods typically report that bone loss is transient and later restored to pre-pregnancy values (Pearce, 2006; Sowers, 1996). However, other

studies have shown complete bone recovery does not occur and that long-term breastfeeding leads to progressive bone loss in the pre- and postmenopausal states (Chowdhury et al., 2002; Grimes and Wimalawansa, 2003; Lopez et al., 1996; Melton et al., 1993; Popivanov and Boianov, 2002; Sowers, 1996); some research has found no such association (Feldblum et al., 1992; Johnell and Nilsson, 1984).

The intensity of nursing may also influence the extent of bone density loss. Studies have shown that lactational decrease in bone density correlates with the amount of calcium lost in the breast milk output (Laskey et al., 1998). For this reason, women who lactate more intensively (i.e., the infant is exclusively breastfed and has frequent feedings) may be expected to have greater net calcium losses and lose more bone (Kovacs, 2001). However, the few studies that have investigated this relationship have found that greater breastfeeding intensity, particularly in the immediate post-partum amenorrheic months, may in fact serve to maintain bone integrity (Pearce, 2006).

During lactation, nursing women typically experience prolonged periods of amenorrhea and low levels of circulating estrogens (Sowers et al., 1993). This condition, coupled with a loss of approximately 300-400 mg of calcium to breast milk, enhances maternal bone loss. This generally lasts until weaning when bone mass is restored (Galloway, 1997; Kovacs, 2001; Stuart-Macadam, 1995). In a six month period of exclusive lactation, approximately 7% of maternal bone mass may be lost (Sowers, 2001). If breastfeeding is terminated before nine months, there appears to be a full recovery of bone mass within the 18 months postpartum period; this indicates the critical contribution of lactation duration to bone status. However, the extent to which multiple reproductive cycles, coupled with other characteristics of lactation (e.g., intensity),

impact long-term skeletal health is still largely unknown; the differences in results from the aforementioned studies may be largely attributed to the heterogeneity of lactation behaviors across study populations.

Inter-Birth Interval

Short spacing between births may be a risk factor for low BMD in pre- and postmenopausal years, due to the cumulative periods of bone loss in quick succession (Affinito et al., 1996). Yet, most longitudinal studies that have investigated intervals between childbirth and lactation periods have found that bone health is not compromised among women with shorter birth spacing (Bererhi et al., 1996; Henderson et al., 2000; Laskey and Prentice, 1997; Sowers et al., 1995).

In natural fertility populations, the period between reproductive cycles (inter-birth interval [IBI]) is shaped largely by lactation patterns. The period of lactational amenorrhea following parturition serves to inhibit ovulation and reduces the probability of immediate conception. This state of reduced fecundity allows the child to benefit from breast milk without having to compete with a gestating sibling for maternal energy stores, and it simultaneously protects the mother from the additional energetic stress of metabolizing for three (Valeggia and Ellison, 2001). A shortened IBI may theoretically compromise the ability of maternal calcium stores to rebound to pre-pregnancy, pre-lactation values, and can lead to a substantial drain of bone from the maternal skeleton. This phenomenon follows the pattern of the maternal depletion syndrome, where repeated cycles of pregnancy and lactation in energy-restricted environments can result in a deterioration of fat and lean muscle stores. Adequate spacing between reproductive

cycles may permit a woman time to replace her fat reserves and return to pre-pregnancy weight (Miller et al., 1994). While the maternal depletion syndrome is typically used to explain the effects of repeated pregnancy and lactation cycles on soft tissue reserves, it can provide a useful framework for examining the relationship between reproduction and bone density. Surprisingly, the few available studies that consider IBI suggest otherwise (Henderson et al., 2000; Sowers et al., 1993).

Age at Menopause

Several research studies have documented a connection between age at menopause and postmenopausal bone health, with an earlier menopausal age linked to lower BMD (Sioka et al., 2010). This is unsurprising since menopause is marked by a cessation of ovarian function and a consequent reduction in the production of estrogens. Because of menopause-related declines in this sex steroid, women who experience earlier menopause spend more time in a hypoestrogenic state that may place them at greater risk for poor bone health (Gallagher, 2007; Kritz-Silverstein and Barrett-Connor, 1993; Ohta et al., 1996; Pouillès et al., 1994).

The present study uses a life-course perspective to investigate effects of the reproductive factors outlined above on bone health in pre- and postmenopausal women from an Indigenous Amazonian population from Ecuador.

RESEARCH OBJECTIVES AND HYPOTHESES

The dynamic nature of bone and the functional system of calcium turnover evolved under vastly different environmental and life conditions than experienced by

Western populations. Research among subsistence groups therefore provides an interesting lens to examine bone health, and has the potential to shed new light on the evolution of calcium regulatory mechanisms. The current study focuses on the Indigenous Shuar women of Ecuadorian Amazonia and evaluates four hypotheses:

1) *Women who experienced an earlier menarche will have higher bone density values.*

This hypothesis is based on the logic that an early menarcheal age increases the lifetime exposure to estrogens.

2) *Women with an older age at first parturition will have higher bone density values.* This hypothesis is based on older age at first parturition delaying any disruption of bone formation prior to achieving peak bone mass.

3) *Women with more protracted periods of lactation will have lower bone density values.*

This hypothesis is predicated on the knowledge that rates of calcium turnover are high during lactation and thus, extended periods of breastfeeding will result in lower bone density values.

4) *Women with longer IBIs will have greater bone density values.* This hypothesis is based on similar processes observed with the maternal depletion syndrome, in which longer IBIs allow for a greater period during which maternal bone density can restore to pre-pregnancy values.

MATERIALS AND METHODS

Study Population

The Shuar are a large indigenous population concentrated in the southeastern region of the Ecuadorian Amazon. Shuar were traditionally forager-horticulturalists although within the last thirty years many communities, particularly in the Upano River Valley (Figures 1.1 and 1.2), have experienced the nutritional, demographic, and economic repercussions of integration into a market economy. The degree of market integration among the Shuar varies widely at an intra- and inter-community level (see Chapter I).

Shuar participants in the present study came from four small, rural Upano Valley communities located approximately 40 minutes to one hour by truck and 3-4 hours by foot from the nearest market center (Sucua). Population size across participating communities ranged from 50 to 150 adults. Members of these communities continue to depend on subsistence horticulture for daily dietary needs, while also engaging in a mix of small scale agro-pastoralist production for market sale.

Participants

Participants in this cross-sectional study included 141 Shuar women between 14-86 years old. Women who were pregnant at the time of the study ($n = 11$) were not included in the statistical analyses since bone density would likely reflect the acute influence of their pregnant state. Further, the small size of the pregnant cohort did not significantly reduce the size of the participant sample. The analyses presented here include a total of 130 women.

Currently lactating women (n = 41) were considered in the statistical analyses as they comprised half the reproductive age women in the sample. However, several analyses treated them as a separate group. Participants were also separated into pre-menopausal or postmenopausal cohorts for analyses. Because many of the reproductive variables are influenced by current age (e.g., older women typically have had more births), pre-menopausal women were further divided into approximately ten-year age sub-categories (14-24, 25-34, 35-44, >45) based on standard age divisions in the clinical and epidemiological literature. Participants were considered postmenopausal (n = 22) if they were not pregnant or lactating at the time of the study and they reported not experiencing a menstrual cycle within the last year.

All participants gave individual informed verbal consent, with both parental consent and child assent for individuals under 18 years old. The study protocol was approved by community leaders, the *Federación Interprovincial de Centro Shuar* (FISCH), and the Office for Protection of Human Subjects at the University of Oregon.

Calcaneal Ultrasound: Bone Mineral Density Measurements

Bone mineral density (BMD) measures were obtained from the right heel of each participant using a gel-based Sahara® bone ultrasonometer (Hologic, Inc., Waltham, MA). Instrumental quality control scans of the manufacturer-provided phantoms were performed daily. The device generates three skeletal health parameters: 1) broadband ultrasound attenuation (BUA; decibels per megahertz), which is the slope of the ultrasonic attenuation versus frequency as it passes through bone; 2) speed of sound (SOS; meters per second), a value determined by the width of the heel and time delay

between initial transmission and subsequent receipt of sound waves; and 3) calculated heel bone mineral density (BMD), which is a device-generated measure determined by combined BUA and SOS values. BUA is more closely related to the micro-architecture of bone (e.g., trabecular connectivity) and, because trabecular bone has a greater surface to volume ratio, it is generally subject to greater metabolic activity and faster rates of turnover than cortical bone. SOS, on the other hand, is greatly influenced by the elasticity and mineral constituents of the bone matrix (Bartl and Frisch, 2004; Lee et al., 2010). All three parameters are presented here separately as they represent different components of bone density and health.

Anthropometric Variables

Participant stature (measured to the nearest millimeter [mm]) and weight (measured to the nearest 0.1 kilogram [kg]) were recorded using a field stadiometer (Seca, Hanover, MD) and digital scale (Tanita BF-558 electronic scale, Tokyo, Japan), respectively. Body mass index (BMI) was calculated as weight (kg)/height (in meters²).

Reproductive History Questions

Retrospective information on participant reproductive patterns was obtained through structured interviews. Each participant answered a series of questions regarding their reproductive histories. These included: a) age at menarche (age at first menses); b) age at first parturition; c) number of parturitions; d) number of offspring breastfed (in rare instances, not all children were breastfed, which accounts for the difference between number of parturitions and number of offspring breastfed); e) average duration of lactation per birth (women provided either a specific estimate of lactation duration for

each child or an average estimate for all children); and f) total lifetime lactation months (this variable represents a sum total of months spent lactating during a participant's lifetime but does not account for the intensity of lactation).

Only five postmenopausal women were able to recall their age at menopause onset; this was too small a participant sample to perform meaningful statistical analyses.

Statistical Analyses

A one-way ANOVA was conducted to determine differences in anthropometric and bone health values by menopausal status. Since many of the reproductive variables are associated with age, comparing the effects of these variables on skeletal parameters from females of different age groups is problematic. Further, controlling for age would remove the effects of age-related bone loss trends. For this reason, statistical analyses were performed within age-defined categories unless otherwise stated. For each age cohort, a stepwise multiple linear regression analysis was performed in order to investigate the relationships between bone density values, anthropometrics, and reproductive variables and to test the hypotheses. Each bone density parameter (BUA, SOS, and BMD) was entered separately as a dependent variable in regression models and age, height, and weight were used as the independent variables. Reproductive variables used in multivariate models included age at menarche, age at first parturition, number of live parturitions, average duration of lactation per birth, total lifetime months spent lactating, and IBI. BMI was not included in the multivariate models because of its collinearity with body weight and stature and because a univariate correlation with BMD was less strong than that of body weight and stature separately.

RESULTS

Table 4.1 presents the proportion of nulliparous and multiparous women by pre- and postmenopausal status. Parity information for one pre-menopausal woman was not available and is not included in this table. A total of 89 (82.4%) pre-menopausal and 21 (95.5%) postmenopausal women reported having at least one child.

Table 4.1. Frequency of nulliparous and multiparous women by menopausal status

	Pre-menopausal	Post-menopausal
Nulliparous	18 (16.7%)	1 (4.5%)
Multiparous	89 (82.4%)	21 (95.5%)

An ANOVA comparing BMD between nulliparous and parous women by menopausal status, controlling for age, indicates no significant differences in these values.

Table 4.2 presents the descriptive statistics and reproductive profiles for participants by menopausal status, including lactating women. Most anthropometric dimensions and all bone density measures are significantly higher among pre-menopausal women than postmenopausal women. Anthropometric, BMD trends, and reproductive profiles for pre-menopausal women are shown in Tables 4.3 and 4.4, with data presented by age groups. A decline in height is also noted with age. However, all other body size and most bone health values show a gradual increase until the mid-30s; weight, BMI, SOS and BMD peak in the 25-34 year age category. BUA is the sole bone measure that does not show a similar trend and rather, peaks in the 35-44 year age category.

Table 4.2. Anthropometric, bone density values, and reproductive profiles for pre- and postmenopausal Shuar women (including lactating women).

	Pre-Menopausal n=108				Post-Menopausal n = 22				P
	Mean	SD	Min	Max	Mean	SD	Min	Max	
Age (years)	29.82	10.80	14.07	52.55	58.34	10.35	48.04	85.98	**
Height (cm)	148.74	4.53	136.00	158.67	144.32	6.07	134.50	153.10	**
Weight (kg)	56.01	10.13	24.90	97.40	52.64	11.16	36.70	81.20	Ns
BMI (kg/m ²)	25.30	4.05	10.62	41.07	25.11	4.07	20.29	34.64	Ns
BUA (dB/MHz)	81.3	16.9	37.4	123.0	58.9	16.1	26.0	89.1	**
SOS (m/sec)	1574.8	28.2	1524.7	1661.9	1527.6	20.7	1475.9	1567.1	**
Heel BMD	0.608	0.112	0.411	0.944	0.427	0.095	0.207	0.607	**
Age at Menarche (years)	13.07	1.16	9.00	16.00	13.29	0.92	12.00	15.00	Ns
Age at First Parturition (years)	17.45	3.33	12.00	35.00	17.64	5.68	14.00	39.00	Ns
Number of Parturitions	4.07	3.58	0.00	13.00	8.81	3.40	0.00	15.00	**
Inter-birth Interval (months)	31.10	12.96	12.00	73.64	31.58	8.02	20.90	49.07	Ns
Total Number of Offspring Breastfed	3.94	3.52	0.00	13.00	8.81	3.40	0.00	15.00	**
Average Duration of Lactation Per Birth (months)	15.11	7.09	0.00	44.00	16.05	5.28	8.57	25.33	Ns
Total Duration of Lactation over Lifetime (months)	66.14	67.73	0.00	312.00	137.24	72.15	0.00	240.00	**

Ns = not significant; ** $p < 0.001$ (significant difference between menopausal states)

Table 4.3. Anthropometric and bone density values for pre-menopausal women by age cohort (including lactating women).

14-24 (n=47)				25-34 (n=25)			35-44 (n=22)			45> (n=14)		
	Mean (SD)	Min	Max	Mean (SD)	Min	Max	Mean (SD)	Min	Max	Mean (SD)	Min	Max
Height (cm)	149.19 (4.94)	136.0	158.67	148.9 (4.11)	140.55	155.1	148.01 (4.36)	139.8	156.4	147.95 (4.13)	140.6	155.0
Weight (kg)	51.23 (7.34)	24.9	65.5	61.65 (12.42)	49.4	97.4	58.28 (6.82)	49.7	76.1	60.01 (11.38)	47.6	90.9
BMI (kg/m²)	22.99 (2.81)	10.62	28.87	27.96 (4.74)	22.77	41.07	26.59 (2.77)	22.66	33.38	27.26 (3.87)	23.26	37.84
BUA (dB/MHz)	79.4 (17.0)	44.7	120.9	82.5 (18.6)	37.4	122.5	83.8 (17.3)	55.3	123.0	82.1 (13.7)	64.2	113.8
SOS (m/sec)	1575.9 (27.0)	1524.9	1629.3	1576.9 (31.3)	1524.7	1649.6	1575.3 (27.8)	1535.8	1661.9	1567.0 (28.6)	1529.7	1613.9
Heel BMD	0.604 (0.108)	0.411	0.848	0.618 (0.125)	0.426	0.878	0.614 (0.115)	0.438	0.944	0.592 (0.104)	0.445	0.792

Table 4.4. Reproductive profile of Shuar women by age cohort.

	14-24 (n=47)			25-34 (n=25)			35-44 (n=22)			45> (n=14)		
	Mean (SD)	Min	Max	Mean (SD)	Min	Max	Mean (SD)	Min	Max	Mean (SD)	Min	Max
Age at Menarche (years)	13.24 (1.23)	10.0	16.0	13.05 (1.05)	11.0	15.0	12.77 (1.19)	9.0	14.0	13.08 (1.08)	11.0	15.0
Age at First Parturition (years)	17.07 (2.21)	13.0	22.0	16.90 (2.47)	14.0	22.0	18.10 (4.46)	13.0	35.0	18.14 (4.44)	12.0	28.0
Number of Parturitions	1.17 (1.18)	0	4.0	4.96 (2.75)	0	10.0	7.09 (3.26)	1.0	13.0	7.29 (3.29)	1.0	13.0
Inter-birth Interval (months)	25.27 (10.22)	14.92	48.0	26.13 (9.39)	12.0	51.93	35.25 (14.61)	12.89	66.18	39.10 (12.59)	21.85	73.64
Total Number of Offspring Breastfed	1.15 (1.19)	0	4.0	4.72 (2.56)	0	10.0	7.05 (3.24)	1.0	13.0	6.86 (3.57)	1.0	13.0
Average Duration of Lactation Per Birth (months)	13.54 (6.97)	0	28.0	16.10 (7.22)	9.0	44.0	15.32 (6.12)	1.0	24.0	15.90 (8.73)	4.0	36.0
Total Duration of Lactation over Lifetime (months)	17.44 (22.23)	0	84.0	69.43 (32.84)	0	132.0	116.64 (75.0)	1.0	288.0	117.04 (87.34)	8.0	312.0

No significant secular trend in age at menarche or age at first parturition is seen in this sample. Across all Shuar participants, IBI varies from 1-6 years, with the longest average duration between births reported among the oldest pre-menopausal cohort (3.25 years [39 months]). While most women report breastfeeding all of their children, the total number of offspring breastfed is not necessarily identical to the total number of parturitions since some infants died immediately following birth and, in one instance, a teenage mother did not produce breast milk. On average, children are completely weaned at approximately 1 year, 3 months old, although some participants continue to lactate up to 3.6 years (44 months). The total duration of lactation across the individual's lifetime is age-dependent as it is related both to number of offspring and breastfeeding practices. Older women, therefore, tend to have higher total lifetime lactation, with one 45-year old participant nursing for approximately 26 years (312 months).

Pre-menopausal lactating women were analyzed as a separate group from non-lactating individuals in the stepwise multiple regression analyses. Among lactating women, anthropometric and reproductive parameters were only significantly predictive of BUA, SOS, and BMD in 14-24 year olds. Tables 4.5-4.10 show the model parameters and coefficients tables from the regressions for each skeletal parameter that was significantly related to anthropometrics and/or reproductive variables. When body size variables are considered separately in the model, there are significant positive associations between stature and BMD (Tables 4.5 and 4.6), SOS (Tables 4.7 and 4.8), and BUA (Tables 4.9 and 4.10) with approximately 41%, 37%, and 44% of the variation in these values attributable to height, respectively. Weight shows no relationship to any bone parameter. When reproductive variables are included in the model, height is no

Table 4.5. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on heel BMD among pre-menopausal lactating women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
14-24	1	.643 ^a	.414	.349	.103145	.414	6.357	1	9	.033
	2	.832 ^b	.692	.615	.079289	.278	7.231	1	8	.028

a. Predictors: (Constant), Height

b. Predictors: (Constant), Height, Age at First Parturition

Table 4.6. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on heel BMD among pre-menopausal lactating women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
14-24	1	(Constant)	-1.769	.947	-1.867	.095
		Height	.016	.006	.643	.033
	2	(Constant)	-.655	.838	-.782	.457
		Height	.002	.007	.069	.818
		Age at First Parturition	.061	.023	.780	.028

Dependent Variable: Heel BMD

Table 4.7. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on SOS among pre-menopausal lactating women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
14-24	1	.608 ^a	.369	.299	26.6084	.369	5.273	1	9	.047
	2	.804 ^b	.646	.558	21.1379	.277	6.261	1	8	.037

a. Predictors: (Constant), Height

b. Predictors: (Constant), Height, Age at First Parturition

Table 4.8. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on SOS among pre-menopausal lactating women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
14-24	1	(Constant)	1016.301	244.385	4.159	.002
		Height	3.774	1.644	.608	.047
	2	(Constant)	1292.523	223.331	5.787	.000
		Height	.216	1.930	.035	.914
		Age at First Parturition	15.178	6.066	.778	.037

Dependent Variable: SOS

Table 4.9. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on BUA among pre-menopausal lactating women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
14-24	1	.664 ^a	.441	.379	14.7201	.441	7.105	1	9	.026
	2	.828 ^b	.686	.608	11.7029	.245	6.239	1	8	.037

a. Predictors: (Constant), Height

b. Predictors: (Constant), Height, Age at First Parturition

Table 4.10. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on BUA among pre-menopausal lactating women

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
14-24	1	(Constant)	-276.579	135.197	-2.046	.071
		Height	2.424	.909	.664	.026
		(Constant)	-123.923	123.646	-1.002	.346
	2	Height	.457	1.069	.125	.428
		Age at First Parturition	8.388	3.358	.732	.037

Dependent Variable: BUA

longer a significant predictor of bone density; BMD, SOS, and BUA values are more strongly associated with age at first parturition. Females who were older at age of first parturition have significantly higher bone density values (BMD, SOS, BUA: $p < 0.005$). No similar trends in bone density for pre-menopausal lactating women in other age cohorts were found. A graphical illustration of the relationship between BMD and height by lactation status among 14-24 year olds is shown in Figure 4.1. Similar results are seen with BUA and SOS but are not presented here. The positive association between BMD and age at first parturition among lactating 14-24 year olds is shown in Figure 4.2.

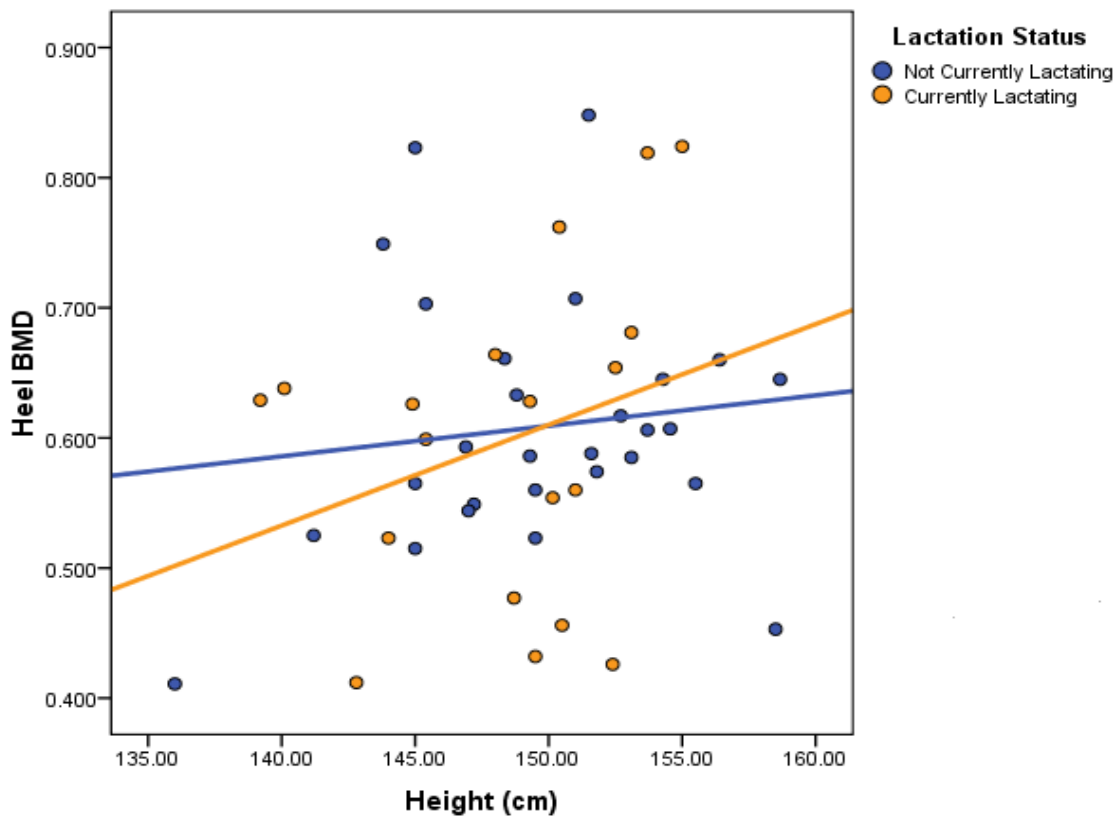


Figure 4.1. Heel BMD and height by lactation status among 14-24 year old pre-menopausal women. A significant relationship between heel BMD and lactation was determined for women who were lactating at the time of the study. (Not Currently Lactating: $R^2 = 0.016$; Currently Lactating: $R^2 = 0.079$)

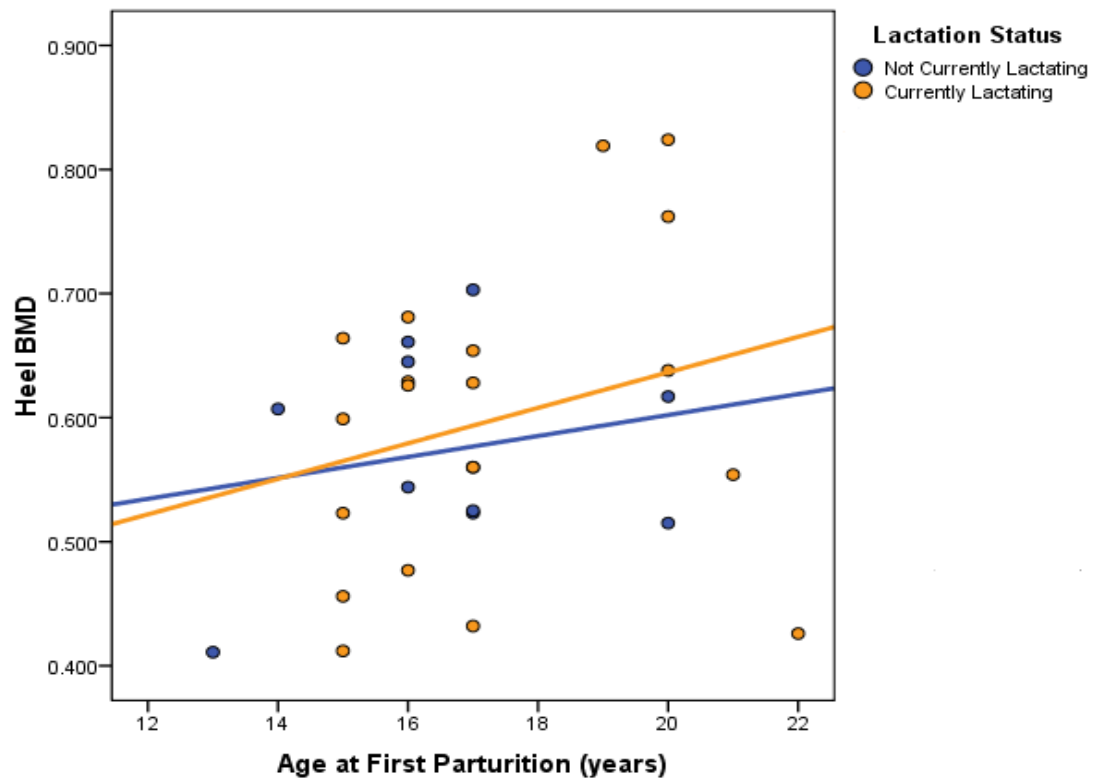


Figure 4.2. Heel BMD and age at first parturition by lactation status among pre-menopausal women. A significant relationship was determined for women who were lactating at the time of the study. (Not Currently Lactating: $R^2 = 0.046$; Currently Lactating: $R^2 = 0.069$)

In the non-lactating, pre-menopausal group, the only significant predictor of bone health parameters is average duration of lactation per birth and this relationship is only found among 35-44 year olds. Women from this age group who had longer bouts of nursing per child had significantly higher BUA ($p = 0.005$), SOS ($p = 0.001$), and BMD ($p = 0.001$) values. Model parameters are shown in Tables 4.11-4.16. No significant relationship between anthropometrics, reproductive variables and bone density were documented for any other age group of non-lactating women.

Table 4.11. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on heel BMD among pre-menopausal non-lactating women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
35-44	1	.832 ^a	.692	.658	.074117	.692	20.259	1	9	.001

a. Predictors: (Constant), Average Weaned Months (per birth)

Table 4.12. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on heel BMD among pre-menopausal non-lactating women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
35-44	(Constant)	.381	.068		5.581	.000
	Average Weaned Months (per birth)	.020	.004	.832	4.501	.001

Dependent Variable: Heel BMD

Table 4.13. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on SOS among pre-menopausal non-lactating women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
35-44	1	.847 ^a	.717	.685	17.7680	.717	22.786	1	9	.001

a. Predictors: (Constant), Average Weaned Months (per birth)

Table 4.14. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on SOS among pre-menopausal non-lactating women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
35-44	1	(Constant)	1514.644	16.354	92.618	.000
		Average Weaned Months (per birth)	5.080	1.064	.847	4.773

Dependent Variable: SOS

Table 4.15. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on BUA among pre-menopausal non-lactating women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
35-44	1	.774 ^a	.599	.555	11.7314	.599	13.448	1	9	.005

a. Predictors: (Constant), Average Weaned Months (per birth)

Table 4.16. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on BUA among pre-menopausal non-lactating women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
35-44	1	(Constant)	54.965	10.798	5.090	.001
		Average Weaned Months (per birth)	2.577	.703	.774	3.667

Dependent Variable: BUA

Stepwise regression analyses of reproductive variables and bone health among postmenopausal women indicate that the only reproductive variable that significantly predicts bone health values later in life is age at menarche (BUA: $p < 0.01$; BMD: $p < 0.05$). The younger the age at first menses, the higher the bone density values in post-reproductive life, specifically with regard to BUA and BMD. Model parameters are shown in Tables 4.17-4.20. Figures 4.3 and 4.4 illustrate the significant relationship between BMD and age at menarche, and BUA and age at menarche, respectively. Age at first menses is not predictive of postmenopausal SOS values in this sample. Of the anthropometric variables, current height, but not weight, was significantly related to postmenopausal BUA. Taller women who were postmenopausal had higher BUA values, but this relationship was not maintained for SOS or BMD. Approximately 36% of the variation in BUA could be attributed to current height (Figure 4.5). While taller women were also the youngest of the postmenopausal age cohort, the relationship between greater height and higher BUA values were maintained when controlling for age.

Table 4.17. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on heel BMD among post-menopausal women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
Post-Menopausal	1	.576 ^a	.332	.281	.077310	.332	6.463	1	13	.025

a. Predictors: (Constant), Age at Menarche

Table 4.18. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on heel BMD among post-menopausal women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
Post-Menopausal	(Constant)	1.227	.307		3.997	.002
	Age at Menarche	-.058	.023	-.576	-2.542	.025

Dependent Variable: Heel BMD

Table 4.19. Model parameters from stepwise linear regression indicating the association between anthropometric and reproductive variables on BUA among post-menopausal women.

Age Cohort	Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics				
						R Square Change	F Change	df1	df2	Sig. F Change
Post-Menopausal	1	.514 ^a	.264	.208	14.1929	.264	4.668	1	13	.050
	2	.793 ^b	.630	.568	10.4822	.365	11.833	1	12	.005

a. Predictors: (Constant), Height

b. Predictors: (Constant), Height, Age at Menarche

Table 4.20. Coefficients chart from stepwise linear regression indicating the association between anthropometric and reproductive variables on BUA among post-menopausal women.

Age Cohort	Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
Post-Menopausal	1	(Constant)	-158.358	102.274	-1.548	.146
		Height	1.511	.699	.514	.050
		(Constant)	3.453	88.984	.039	.970
	2	Height	1.384	.518	.471	.020
		Age at Menarche	-10.738	3.122	-.606	.005

Dependent Variable: BUA

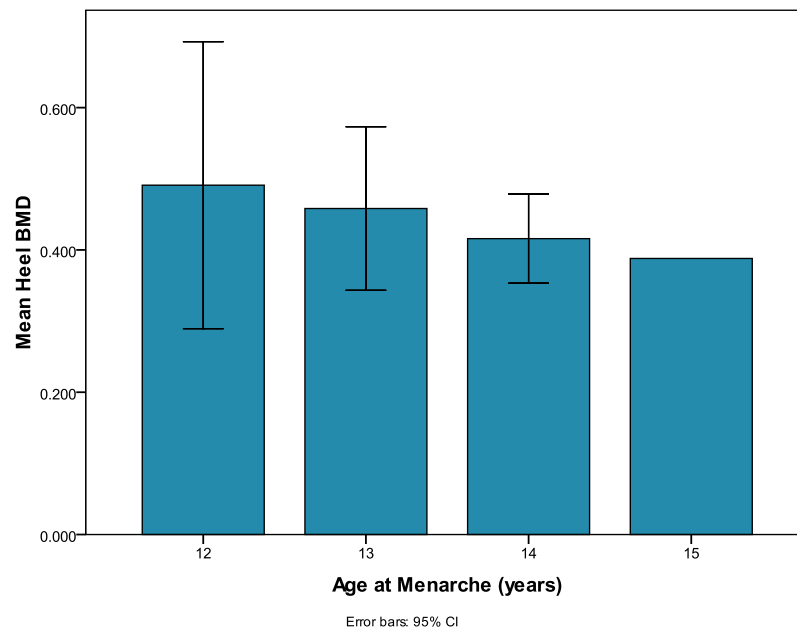


Figure 4.3. Heel BMD by age at menarche among post-menopausal women with error bars.

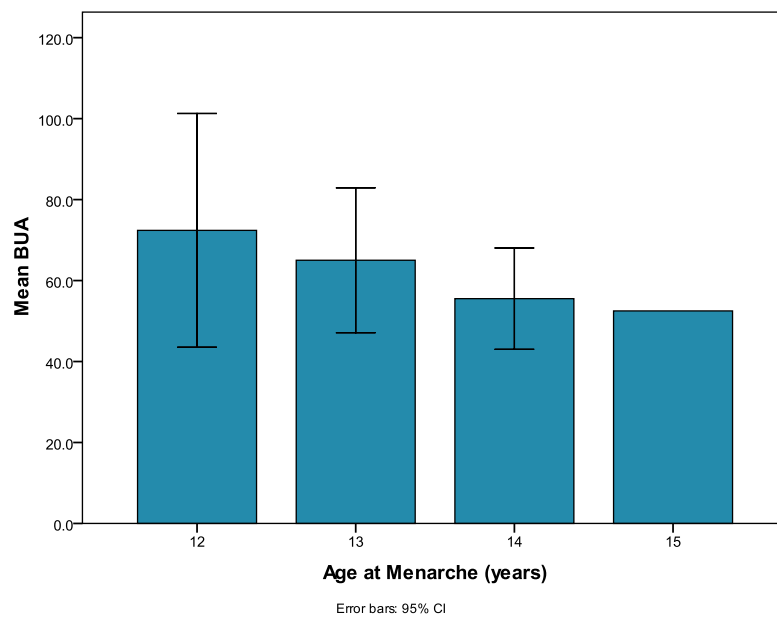


Figure 4.4. Mean BUA by age at menarche among post-menopausal women with error bars.

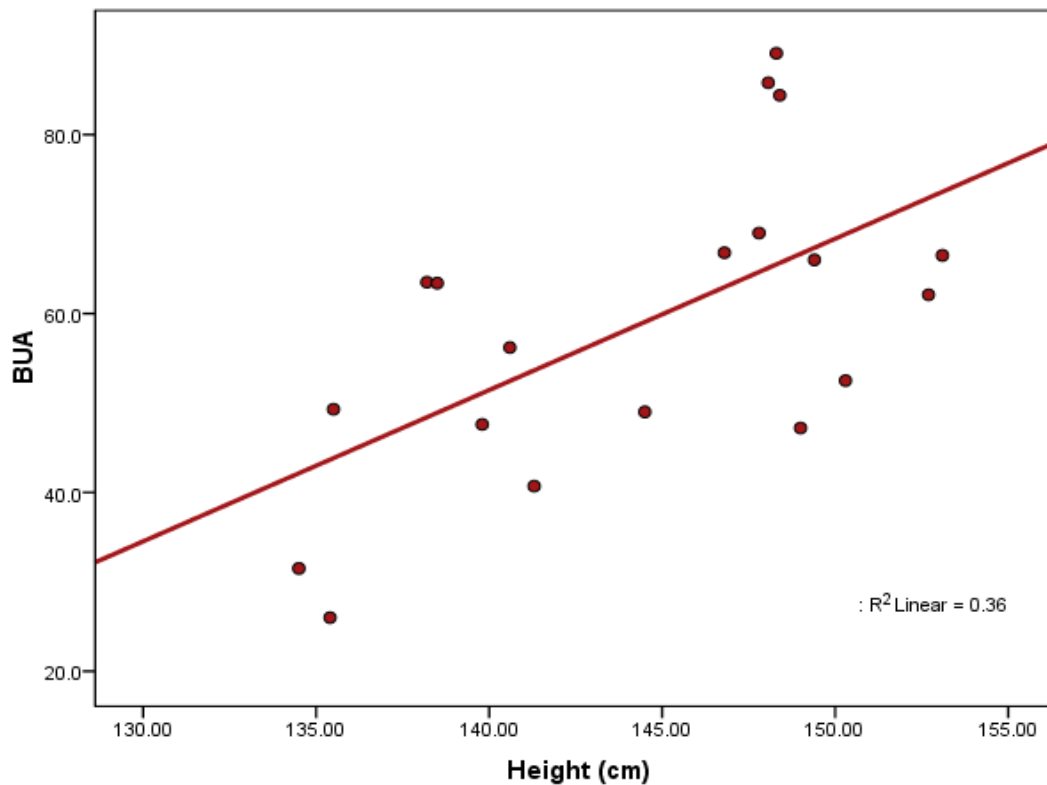


Figure 4.5. Scatter plot of BUA by height among post-menopausal women with linear best fit line ($R^2 = 0.36$)

DISCUSSION

The primary goal of the present study was to test several hypotheses related to the association between bone health and reproduction among Shuar women from Ecuadorian Amazonia. Shuar females, overall, have modestly higher pre-menopausal bone density values than United States reference population (Chapter III; Figure 3.1). However, bone density among postmenopausal women are similar to the reference range from the U.S. population.

Age at Menarche

Hypothesis 1. Women who experienced an earlier menarche will have higher bone density values.

Among Shuar postmenopausal women, earlier menarche was associated with higher bone density values, although a similar trend was not found for pre-menopausal women. Postmenopausal women who were younger at first menses (12 or 13 years old) had significantly greater skeletal health values than women who reported first menses at 14 or 15 years old. Earlier menarcheal age was the primary predictor of postmenopausal bone mass, suggesting that the timing of this early developmental stage canalizes bone density phenotype into later life, despite effects of other reproductive factors such as number of live births or inter-birth interval.

From a mechanistic perspective, the influx of estrogens at menarche contributes to bone accretion processes and thus, an early first menses expands the female reproductive life and increases cumulative exposure to these hormones. Additionally, early menarcheal age is related to larger body size, which increases mechanical loading of the skeleton (especially in the lower limbs), and to greater adiposity, the latter of which proliferates the production of estrogen. Thus, both larger body size and greater fat content can contribute to increases in bone quantity (Eastell, 2005).

From an evolutionary perspective, the age at which a female experiences her first menses reflects a suite of early environmental and social factors that influence nutrition and disease (Sloboda et al., 2010). According to life history theory, a developing organism will allocate energetic resources to growth, and these resources will only become essential for reproduction at the time of sexual maturity. In an energy-deficient

environment, organisms will delay reproductive maturation until a time when resources are adequate and reproductive activities will be successful (Coall and Chisholm, 2010; Ellison, 1982; Garn, 1987). In this study, a younger age at first menses appears to indicate better early environmental and energetic states. Postmenopausal Shuar women who experienced early menarche were arguably in better phenotypic condition than those who experienced first menses later – i.e., they were within a “healthy” range for the timing of this developmental stage at an earlier age. Women who are older at menarche may reflect more compromised early phenotypic quality that then extends throughout life and into postmenopausal life. This finding that key developmental events can serve as an indirect proxy for early energetic conditions with effects on phenotypic quality in later life is also supported by the positive association between height and skeletal health.

Height is frequently used as a marker of early environmental and nutritional conditions (Benefice et al., 2006; Bogin, 1999; Bogin and Loucky, 1997; Bronte-Tinkew and DeJong, 2004), and it is the one anthropometric variable that recurrently demonstrates relationships with bone health across pre- and postmenopausal women. As with the mechanisms governing menarche onset, shorter stature can reflect poor nutrition or adverse circumstances that lead to the devotion of energy towards a competing physiological domain, such as immune function, and inhibit skeletal growth in the process (Ellison et al., 1993; Gluckman and Hanson, 2006; Jasienska et al., 2006). Results from the current study indicate that both greater stature and earlier age at menarche may serve as indicators of early energetic conditions, and they remain important predictors of phenotypic quality, specifically bone quality and quantity, across the lifespan.

Age at First Parturition

Hypothesis 2. Women with an older age at first parturition will have higher bone density values.

The present study found partial support for the second hypothesis. That is, only among young, pre-menopausal lactating women were the positive effects of age at first parturition on BMD documented; however, a similar relationship was not found for non-lactating women. Pregnancy and lactation are periods of extensive bone turnover and, if normal growth patterns are disrupted to accommodate reproduction, this may result in lower peak bone mass. A later age at first parturition, therefore, may provide a protective effect for early bone health. Being young (<18 years) at first pregnancy is also linked to poorer pregnancy outcomes and greater risk of fetal deaths (Kline et al., 1989; Kramer, 1987; Wood, 1994), an association that may suggest an evolutionary advantage to a later age at first parturition for both optimal maternal and offspring fitness. However, one should also consider that a later age at first pregnancy shortens the reproductive life span, which reduces the potential number of offspring a female may have and can lower her completed fertility. Therefore, there appears to be a quantity/quality trade-off between maternal condition and the number or quality of her offspring.

Lactation

Hypothesis 3. Women with more protracted periods of lactation will have lower bone density values.

This study did not document significant relationships between bone density and number of offspring breastfed or total lifetime lactation months, for either pre- or

postmenopausal cohorts. The findings did show that, among the Shuar, average weaning is relatively short (~15 months) compared to many other subsistence populations including the Yanomamo (24-36 months) (Early and Peters, 1990), and the !Kung (36-48 months) (Lee, 1979). The only significant and positive association between any lactation variable and BMD was found among 35-44 year old women; women who reported longer periods of breastfeeding per birth had greater BMD values.

These findings are not entirely surprising given data from several studies that suggest the system of calcium homeostasis is efficient at restoring calcium stores to pre-pregnancy and pre-lactation values (e.g., Sowers et al., 1993). The evolved ability to remove and rapidly restore calcium from the skeleton during reproductive states allows for frequent cycles of pregnancy and lactation and in the long-term, improves maternal and offspring fitness (Stini, 1995). Several longitudinal studies have demonstrated that breastfeeding women lose bone mass during the first 6 months, and this typically coincides with the period of lactational amenorrhea. With the resumption of menses, women who breastfeed typically regain their bone mass (Kalkwarf and Specker, 1995; Kolthoff et al., 1998; Lopez et al., 1996; Sowers et al., 1996). Studies have also shown that women who do not breastfeed do not experience post-partum bone loss but they do not benefit by gaining in bone mass either (Hayslip et al., 1989; Laskey et al., 1998). Cross-sectional studies generally do not note this pattern of post-partum bone recovery as these women are not followed beyond six months postpartum and thus, only a partial picture of lactation-induced bone changes was provided by these studies.

While the current study was cross-sectional, a scatter plot of BMD by years since last birth can offer insights into how BMD fluctuates through time with lactation status

(Figure 4.6). Among non-lactating women, BMD appears to be relatively stable through time; conversely, among lactating women, BMD is highly variable. Although an ANOVA did not reveal significant differences in bone density values between lactating and non-lactating women by age cohort, the relatively stable nature of BMD in non-lactating women is noteworthy and adheres to predictive trends suggesting that post-partum changes in bone density are transient and effectively restored with the cessation of lactation.

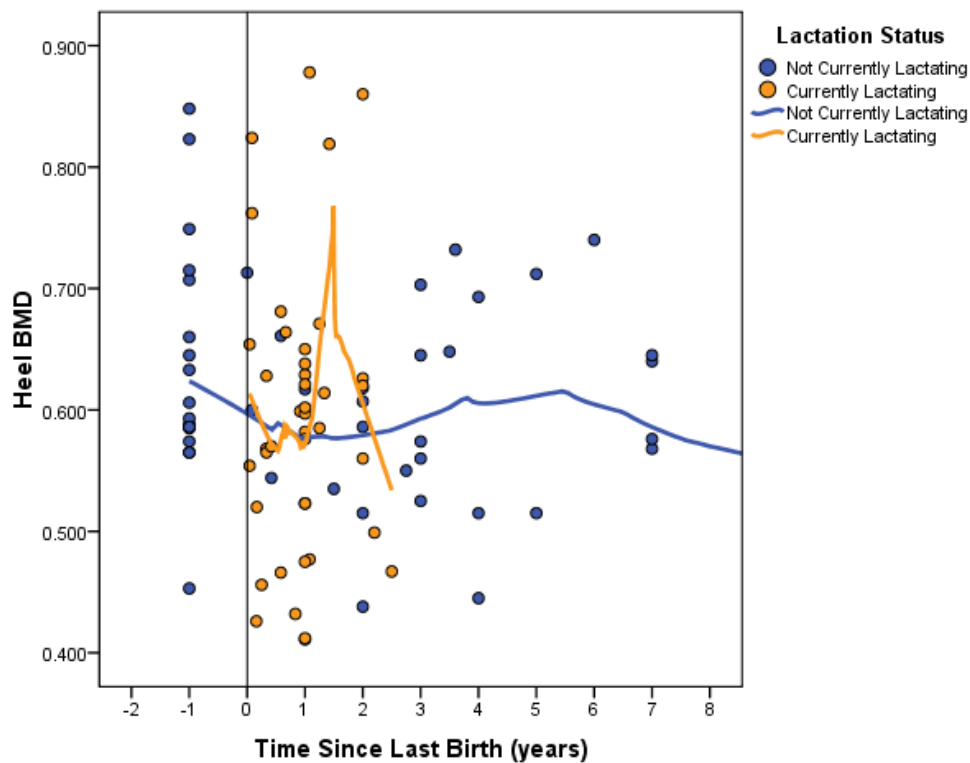


Figure 4.6. Scatter plot of heel BMD and time since last birth (years) by lactation status among pre-menopausal women with Loess smoothing lines. Individuals to the left of the black line (zero) are nulliparous. (Not Currently Lactating: $R^2 = 0.046$; Currently Lactating: $R^2 = 0.069$)

The only significant association between bone density and lactation was found among pre-menopausal, non-lactating women; the findings indicate that longer periods of breastfeeding per child may confer a protective effect on BMD in the 35-44 year old cohort. While several studies report a negative association between number of months of recalled lactation and bone status (Lissner et al., 1991), the vast majority of epidemiological studies of pre- and postmenopausal women have found no adverse effect of lactational history on BMD (Feldblum et al., 1992; Kovacs and Kronenberg, 1997; Johnell and Nilsson, 1984; Sowers, 1996). The precise way that bone loss is experienced and subsequently restored during lactation is not clear, and our finding that longer nursing periods are related to higher BMD in the 35-44 year cohort, independent of quantity of offspring, is somewhat surprising. While speculative at this point, one possible explanation is that women who are able to breastfeed their offspring for longer duration are essentially those with an enhanced ability to store and mobilize calcium, and this in turn reflects better overall bone phenotype. Put simply, having healthier bones may allow for better calcium mobilization and thus, an ability to increase lactation time per child.

Few associations between lactation variables and bone mass were documented in this study. Because of the heterogeneity in lactation duration and intensity within and across populations, the long-term effects of lactation on skeletal health remain unclear. Additional research into this relationship is necessary and should also consider the social and ecological factors that shape the decisions about lactation duration and intensity (e.g., availability of social support) and the implications for skeletal health (e.g., Piperata, 2009).

Inter-Birth Interval

Hypothesis 4. Women with longer IBIs will have greater bone density values.

The current study found no significant association between birth spacing and bone BMD for pre- or postmenopausal women. In this study as well as others (e.g., Bererhi et al., 1996; Sowers et al., 1995), the effects of the maternal depletion syndrome have not been demonstrated for skeletal reserves. One hypothesis for this pattern involves the contribution of maternal fat stores to satisfy costs of offspring brain development. The mobilization of larger amounts of fat during pregnancy and lactation serves an adaptive function that meets the developmental needs of the fetal and neonatal brain (Lassek and Gaulin, 2006). Replenishing fat stores rapidly may therefore not be possible because of the continuous and critical need to satisfy the requirements of an ever-expanding and metabolically expensive human brain. While maternal skeletal calcium stores are drawn upon to support offspring skeletal growth in an analogous way, it may be the case that replenishing calcium stores is more easily achieved than maternal fat reserves, perhaps because of greater availability of calcium from the environment (see Chapter II), thereby facilitating the recovery of bone loss between reproductive cycles.

Developmental Origins of Adult Bone Density

The present study suggests that the timing of early developmental events, specifically age at menarche and age at first parturition, may serve as a proxy for exogenous conditions that are important for establishing a phenotype of bone density. This bone density phenotype appears to persist throughout life despite transient perturbations during the reproductive years. This should not be surprising since early

developmental origins, especially growth in utero and infancy, have demonstrated an important role in structuring adult health outcomes such as obesity, heart disease, hypertension, and several cancers (Barker, 1995a, 1995b; Gluckman and Hanson, 2004). Other recent studies have suggested similar links between early developmental factors, particularly those experienced during intrauterine life, and bone mass and osteoporosis risk in adulthood (Agarwal and Glencross, 2011; Cooper et al., 1997; Cooper et al., 2001; Oreffo, 2003).

Some support for early origins of adult bone density has been garnered from epidemiology studies of maternal anthropometric and lifestyle effects on bone mineral density of their newborn offspring. Godfrey and colleagues (2001) found that neonatal bone mass is positively associated with birth weight, birth length, and placental weight. Additional determinants include maternal birth weight and tricep skinfold thickness at 28 weeks gestation. Maternal smoking and high maternal physical activity are negatively associated with neonatal bone mass. The study demonstrates that maternal nutrition, body size, and lifestyle appear to modify the fetal nutrient supply and subsequent bone accumulation (Godfrey et al., 2001). Additionally, longitudinal research on cohorts with available birth and childhood growth records and adult fracture information have found that low birth weight and poor childhood growth (e.g., height- and weight-for-age) are associated with greater risk of fractures in adulthood (Cooper et al., 1997; Cooper et al., 2000, 2001).

Several physiological studies have also examined the role of early endocrine programming in establishing bone status. Findings suggest that birth and infancy weight may predict basal levels of growth hormone (GH) and cortisol in adulthood and these

hormones, in turn, determine the rate of bone loss in adult life (Dennison, et al., 1999; Fall et al., 1998; Phillips et al., 1998). These data are compatible with hypotheses that stressors on the maternal body during intrauterine life alter the sensitivity of the epiphyseal plate to GH and cortisol. The effects of endocrine programming could lead to lower peak bone mass, potentially reduce bone mineralization, and predispose one to an accelerated rate of bone loss later in life (Cooper et al., 2002).

Study Limitations

The present study has several key limitations. First, participant sample sizes are small, particularly in the postmenopausal cohort, which limit statistical power; therefore, these data should be considered preliminary. Future data collection will augment the size of this cohort.

Second, a number of postmenopausal women could not recall their age at menopause, which reduced the sample size and illustrates the limitation of retrospective studies. The current study is limited by a reliance on memory for information about past reproductive patterns. Relatively close birth spacing, high fertility rates, and infant/childhood mortality can make it difficult for participants to accurately recall information such as number of births, offspring age, and lactation patterns. These are common problems in human biology research that the current study attempted to control by corroborating information with relatives.

Third, the present study was cross-sectional, which, although useful for obtaining a relatively large participant sample, provides only a snapshot of bone health. This

approach also impairs the ability to establish causality and to discern long-term changes in BMD.

Finally, the influences of diet and physical activity were not addressed in the current study.

CONCLUSIONS

The present study demonstrated that early menarche and greater stature are associated with higher BMD among postmenopausal Shuar women. This result highlights the importance of early life history events and conditions for establishing phenotypic quality appear to extend late into adult life. Further, results suggest that the effects of lactation on skeletal health are transient during pre-menopausal years, and are effectively restored during weaning. The fertility patterns of Shuar women are vastly different than those of Western females, from whom most of current understanding of the relationship between reproduction and skeletal health are based. This study represents one of the few to investigate the reproductive effects on patterns of bone loss in a non-Western, subsistence based population. However, additional data from the Shuar are necessary to examine the trade-offs in this relationship in greater detail, and more cross-cultural studies approaching skeletal health within a life course perspective are needed.

BRIDGE TO CHAPTER V

Using a life-course perspective, Chapter IV presented data on Shuar reproductive patterns and examined their effects on physiology, specifically bone health. As discussed in the previous section, social and environmental conditions motivate the decisions

women make regarding the timing and duration of various reproductive factors, such as lactation. For pregnant and lactating women, access to social networks and allo-parents will invariably impact the decisions made about time allocation and participation in economic activities. The next chapter investigates another aspect of reproductive energetics in one Upano Valley community and documents the effects of women's reproductive status on their own activity levels and those of their mates. Chapter V is predicated on the principle that several bio-behavioral strategies may be adopted by females in order to compensate for the high costs of reproduction and these strategies are ecologically and environmentally determined. This research employs accelerometry, a relatively recent technique developed for objectively measuring physical activity in free-living populations. Chapter V concludes with a consideration of the utility of accelerometry technology to examine activity patterns and energy use regulation in human biology research.

CHAPTER V

PHYSICAL ACTIVITY IN AN INDIGENOUS FORAGER-HORTICULTURALIST POPULATION AS MEASURED USING ACCELEROMETRY

This chapter is published as Madimenos FC, Snodgrass JJ, Blackwell AD, Liebert MA, and Sugiyama LS in the American Journal of Human Biology 23(4):488-497. Activity data collection was collected primarily by Madimenos in the Summer 2008 and 2009 field seasons, with some assistance from Liebert in the Winter 2009 field season. Data analysis and write-up were conducted by the author of the dissertation. Blackwell, Sugiyama, and Snodgrass provided editorial assistance.

INTRODUCTION

In indigenous societies, increased market integration, defined as the commoditization of material, food, and labor (Lu, 2007), has been linked to changes in health outcomes such as obesity, type 2 diabetes, and hypertension (Baker et al., 1986; Friedlander et al., 1987; Huss-Ashmore et al., 1992; Snodgrass et al., 2007). Changes in energy dynamics, or more specifically, increased energy intake and reductions in energy expenditure, have been implicated in this health shift as populations transition from traditional subsistence economies to increasingly sedentary occupations. Numerous studies have documented dietary transitions (e.g., Monteiro et al., 1995; Popkin, 2006), yet relatively few studies have systematically measured physical activity in populations

undergoing economic development and lifestyle change. This is unfortunate given the importance of physical activity data for understanding the health effects of market integration and the increasing global prevalence of overweight and obesity.

The relative lack of available data on activity patterns in subsistence-level populations partially reflects the difficulty of accurately quantifying habitual or “free-living” physical activity under field conditions. Physical activity is typically measured using time allocation, doubly-labeled water (DLW), or heart rate (HR) monitors, but all these methods have distinct advantages and disadvantages (Snodgrass, 2011; Westerterp, 2009). Most population-level studies of subsistence groups have used time allocation data, but this technique can underestimate energy costs, especially at high levels of physical activity (Leonard et al., 1997; Spurr et al., 1996). DLW, generally considered the “gold standard” for measuring free-living energy expenditure, estimates activity over a relatively long period of time (typically 7-10 days), is expensive and its high cost has limited its use in non-Western contexts (Coward, 1998). HR monitoring, on the other hand, is far less costly, but requires individual calibration of heart rate/energy expenditure relationships, which is time consuming and adds a significant measurement burden for participants.

Recently, technological advances in accelerometry have given researchers a new tool for estimating total daily energy expenditure (TDEE) and physical activity level (PAL) under field conditions although, as with all methods, there are advantages and disadvantages to their use (Chen and Bassett, 2005; Plasqui and Westerterp, 2007; Snodgrass, 2011; Swartz et al., 2000). Accelerometers are electronic motion sensors typically worn at the waist that can objectively measure the intensity, frequency, and

duration of body movement. The devices are also durable, fairly non-intrusive, reasonably affordable, and allow for the estimation of activity variation on shorter timescales than possible using time allocation studies or DLW. Several models of accelerometers have been extensively validated (Blanton et al., 2005; Chen and Bassett, 2005; Heil, 2006; Hoos et al., 2003; Plasqui and Westerterp, 2007), with data quality from accelerometers generally considered second only to DLW (Westerterp, 2009). While accelerometry has been widely applied in Western research contexts, particularly in clinical studies and among athletes, this technology has rarely been used in non-Western field settings.

An area of research where accelerometry could be particularly useful is in determining the effects of increasing market integration on patterns of energy expenditure and sexual division of labor. Most research to date suggests that a transition from a subsistence-based to market economy typically leads to reductions in female activity levels. For example, among Inuit from Igloolik, Canada, who were experiencing the initial stages of economic development, both males and females demonstrated a reduction in subsistence activities, although males maintained relatively higher activity levels than females (Shephard and Rode, 1996). In the Yakut (Sakha) of Siberia, a herding population that has experienced rapid economic development over the past decade, overall population activity levels were generally low, but they were considerably lower among females than males (Snodgrass et al., 2006). These sex differences in physical activity patterns are shaped by variation in subsistence participation and by other cultural factors. Even subsistence groups living in broadly comparable environments and with similar subsistence economies can have distinct physical activity levels. For example, the

Tukanoan women of Colombia have significantly higher activity levels than Brazilian *Ribeirinha* women (Dufour and Piperata, 2008) despite both populations living in Amazonian neo-tropical forest environments. This difference reflects inter-cultural variability in household sexual divisions of labor (i.e., male participation in harvesting activities), settlement patterns (i.e., proximity to gardens), and minor dietary differences (i.e., amount of processing of the cassava staple).

Since energy is a fundamental limiting resource that must be allocated to different somatic functions, an accelerometry-based energetics approach can provide a useful tool to investigate life history trade-offs such as the energy available for reproduction. An evolutionary life history framework has been useful in understanding the socio-behavioral strategies adopted by subsistence populations for economizing energy expenditure during costly reproductive states such as pregnancy and lactation (Hill and Hurtado, 1996; Stearns, 1992). A small number of studies have addressed the various strategies used by rural women in subsistence-based economies, whose energy conserving options are more restricted than women from urban settings (e.g., Guillermo-Tuazon et al., 1992; Piperata and Dufour, 2007). Among subsistence-based populations in Nepal (Panter-Brick, 1993) and Brazil (Piperata and Dufour, 2007), women decreased energy expenditure for subsistence activities, presumably as a means of offsetting their elevated energy needs for reproduction, even at the cost of reducing contributions to food production. In some contexts, a decrease in female economic contribution has been shown to prompt a compensatory increase in familial support or, more specifically, male provisioning. For example, Marlowe (2003) observed greater food returns from Hadza

hunter-gatherer men with pregnant and lactating wives, a socio-behavioral strategy that extends until the time of weaning.

The present study was designed as a preliminary step towards addressing the data gaps outlined above by using accelerometry to document energetic patterns in the Shuar, an indigenous Ecuadorian forager-horticulturalist population. The goals of this study are to: 1) compare physical activity levels among the Shuar with other populations for which data are available; 2) investigate potential sex differences in physical activity and examine the effects of reproductive status on activity; and 3) discuss the advantages and disadvantages of accelerometer use in human biology research.

MATERIALS AND METHODS

Participants

This study was conducted in one Ecuadorian Upano River Valley community located approximately forty minutes by truck from Sucua, the nearest major town center.

Participants included 49 Shuar adult (14-66 years of age; 23 males, 26 females) volunteers, representing approximately 50% of the adult residents in the study community. Among the female participants, five were pregnant (P), ten were lactating (L), and eleven were non-pregnant/non-lactating (NPNL). Among the male participants, 16 did not have pregnant or lactating wives while the remaining seven males did.

All participants gave individual informed verbal consent, with both parental consent and child consent for subjects under 15 years old. Research was conducted from a Ministerio de Salud health center, and the study design was approved by village leaders,

the *Federación Interprovincial de Centro Shuar* (FISCH), and the Office for Protection of Human Subjects at the University of Oregon.

Seasonality

Data were collected over the course of three field seasons: August-September 2008, February 2009, and August-September 2009. Activity data were recorded for each participant for one of the three field seasons. The months of data collection are all considered “dry” months (i.e., periods of moderate rainfall of approximately 300-400mm) (Sirén, 2007). Furthermore, although neo-tropical in flora and fauna, the Upano River Valley region (located 1000 meters above sea level at the base of the Andes) experiences less rainfall and temperature variation across the year than in the Amazonian lowlands east of the Cutucu and, thus, our expectation is that activity will not vary as much seasonally as in some subsistence populations in other regions.

Anthropometry

Participant stature (measured to the nearest mm) and weight (measured to the nearest 0.1 kilogram [kg]) were recorded using a field stadiometer (Seca, Hanover, MD) and digital scale (Tanita BF-558 electronic scale, Tokyo, Japan) respectively, according to established procedures (Lohman et al., 1988). Body mass index (BMI) was calculated by dividing body mass (in kg) by height (in meters²).

Physical Activity

Physical activity was estimated using Actical accelerometers (Respironics, Bend, OR), one of the more widely validated monitoring devices (Plasqui and Westerterp, 2007). Each monitor was initialized before placing it on the participant, which involves entering a start date and time into the Actical 2.1 software platform. During this initialization process, the participant's sex, age, height, and weight were also entered. The Actical begins recording automatically at the designated start time and continues to record activity until the data are downloaded or the device memory reaches its limit (ranging from 11-45 days, depending on the user-defined epoch length selected). Each participant wore an Actical at the waist, positioned over their right iliac crest for at least two consecutive days irrespective of weekend or weekday. Participants engaged in their normal activities or those already planned, and kept the monitor on while bathing and sleeping. Following the activity recording period, the data from the Actical were downloaded to a computer.

A shortcoming of the present study is that the number of days the device was worn, the time the device was initialized, and when the device was removed varied between participants. This limitation in the study design reflects the constraints of standardizing the collection of activity data from free-living populations living in remote environments. Participants lived various distances from the health center and although all individuals wore the device for at least two days, a few individuals were not available to remove the device at a precise time and continued to wear the monitor for several additional days. No participant wore the device for more than four days. The average number of days in which a device was worn was 2.51 days (SD = 0.681). Data were

averaged over standardized 24-hour periods, so the specific time the device was initialized or removed is unlikely to enter systematic bias in the data.

The accelerometer devices record Activity Counts (AC), which represent the frequency and intensity of acceleration events that occur during user-defined epochs. The epoch length is the period of time the device will accumulate and record ACs, and then reset the counter to zero. Depending on the user preference, Actical epoch lengths may vary between 15 seconds and one minute; this option does not affect the actual accumulation of activity counts, only the detail of the data output. The significance of accelerometer epoch lengths will vary depending on the type of research being implemented. For example, shorter epochs are useful for studies examining short, sporadic bouts of activity as with research on children (e.g., Stone et al., 2009). Since the purpose of this study was to examine activity patterns over a period of a few days, the precise length of the epoch is not critical and therefore, the default setting, a 25-second epoch, was used.

Although the Actical software converts ACs into caloric energy expenditure, two additional techniques are employed to estimate energetic parameters from raw accelerometry data: 1) a two-regression model by Heil (2006) to determine activity energy expenditure (AEE) based on ACs (averaged for the recorded days); and 2) total daily energy expenditure (TDEE) calculated as AEE + basal metabolic rate (BMR). BMRs for males and NPNL women were estimated using the Oxford predictive equations according to weight and age (Henry, 2005). BMR among pregnant participants was determined using an appropriate multiplication factor depending on trimester (Prentice et al., 1996). Using FAO/UNU/WHO (2004) guidelines, BMR changes in lactating women

were determined as approximately 675 kcal above their non-lactating metabolic requirements.

Currently, there is no consensus on the best method for adjusting for the effects of body size and composition in energetics, although physical activity level (PAL; TDEE/BMR) is the most commonly used method of comparing activity data in human biology research. However, using PALs as a measure for pregnant women may be complicated by a progressively increasing BMR that occurs throughout the course of pregnancy. Thus, a woman participating in the same activities with similar duration and intensity would show a decline in PAL throughout her pregnant state. For this reason, we also present data for the activity variables AEE and AC in order to examine population-level differences in activity.

Statistical Analyses

Pair-wise comparisons were conducted using independent samples t-tests, with two-tailed *P*-values and equal variance assumed. Variance components were assessed in R using the *lme* and *VarCorr* procedures from the package *nlme*. Age patterns were fit with thin-plate splines using generalized additive modeling (GAM) by using the *gam* procedure in the R package *mgcv*. ANOVAs were used to assess interactions between sex and the reproductive state of females in a household (i.e., pregnant, lactating, NPNL). Following ANOVAs, post-hoc comparisons were done using two-tailed t-tests. Statistical analyses were performed using a combination of SPSS 17.0 (SPSS, Inc.) and R 2.10.1 (www.r-project.org).

RESULTS

Descriptive statistics for age, anthropometric, and activity data are presented in Table 5.1. Men had significantly higher average AEEs (762 kcal/d vs. 573 kcal/d, $t = 2.56$, $P = 0.01$), PALs (1.54 vs. 1.42, $t = 2.24$, $P = 0.03$), and ACs (290,064 vs. 224,900, $t = 1.97$, $P = 0.05$) than women. However, differences between men and women in TDEE (2176 kcal/d for men vs. 2033 kcal/d for women) and estimated BMR were non-significant (1403 kcal/d for men vs. 1444 kcal/d for women). Controlling for sex, we tested for differences between activity parameters collected during August/September field seasons compared to the field season in February, and between activity levels from weekends (two day averages starting on Friday, Saturday, or Sunday) compared to weekdays. Activity levels from February were lower than activity levels in August/September (marginal means: AEE: 494 vs. 694 kcal/day, $F_{1,45} = 6.32$, $P = 0.02$; PAL: 1.37 vs. 1.46, $F_{1,45} = 5.06$, $P = 0.03$; AC: 199,052 vs. 265,847, $F_{1,45} = 3.23$, $P = 0.08$). Activity levels from weekends were lower than weekdays, but this difference was not statistically significant (AEE: 525 vs. 662 kcal/day, $F_{1,45} = 2.97$, $P = 0.09$; PAL: 1.38 vs. 1.48, $F_{1,45} = 2.62$, $P = 0.11$; AC: 207,741 vs. 257,159, $F_{1,45} = 1.78$, $P = 0.19$).

We used a random effects model to examine the variance in PALs between participants, and found that 22% of the variance in PALs was attributable to participant identity (variance participant = 0.013, SD = 0.11, Residual = 0.145, SD = 0.21). Similarly, a participant's day one activity level was significantly correlated with his or her day two activity level ($r = 0.37$, $P = 0.01$). Although correlation coefficients between day 1 and day 2 activity levels are modest, Shuar in the study community engage in mixed subsistence and agricultural production, so day to day variability in activity

Table 5.1. Descriptive statistics for anthropometric and activity data^{a,b}

Measure	Males (n = 23)	Females (n = 26)	P	Combined (n = 49)
	Mean (SD)	Mean (SD)		Mean (SD)
Age (years)	32.7 (12.9)	27.8 (10.7)	ns	30.1 (11.9)
Height (cm)	155.6 (8.6)	146.0 (4.3)	***	149.5 (8.2)
Weight (kg)	58.2 (10.5)	50.0 (8.2)	***	53.7 (9.9)
BMI (kg/m ²) ^c	23.8 (2.8)	23.4 (2.8)	ns	23.6 (2.8)
TDEE (kcal/d)	2176 (396)	2033 (374)	ns	2100 (387)
BMR (kcal/d) ^d	1403 (170)	1444 (318)	ns	1425 (257)
AC ^e	290,064 (128,589)	224,900 (102,905)	*	255,000 (119)
AEE (kcal/d) ^f	762 (290)	573 (227)	*	661 (273)
PAL ^g	1.54 (0.18)	1.42 (0.19)	*	1.48 (0.19)

^a TDEE, total daily energy expenditure; BMR, basal metabolic rate; AEE, activity energy expenditure; PAL, physical activity level.

^b Differences between females and males are statistically significant at * $P < 0.05$; *** $P < 0.001$, ns = no significance.

^c Weight divided by height in meters squared (kg/m²).

^d Calculated using the Oxford equations (Henry, 2005).

^e Activity counts (AC) represent the frequency and intensity of acceleration events that occur during a user-defined epoch.

^f AEE from activity counts based on two-regression equation (Heil, 2006).

^g TDEE/BMR.

levels is expected, just as it would be under the traditional foraging economy.

We examined the age and sex patterning of activity levels using GAM, and fit thin plate splines to the age pattern (Table 5.2; Figure 5.1). GAM determines the optimum degrees of freedom for the spline fit through GCV minimization. In the GAM models the overall effect of sex was significant ($t = 2.29$, $P = 0.03$). For females the optimum model included no age effect. For males, the overall effect of age was also non-significant, but did include additional degrees of freedom ($F = 1.61$, estimated $df = 3.44$, $P = 0.19$). Examining a plot of the splines and individual data points (Figure 5.1) reveals that the AEE difference between males and females is driven largely by males in the 25 to 35 year age category and that the only obvious age effect is within this age group. Examining only individuals age 25-35 years shows that males and females differed

significantly in AEE ($t = 3.33$, $df = 17$, $P < 0.01$), PAL ($t = 3.05$, $df = 17$, $P < 0.01$), and AC ($t = 2.05$, $df = 17$, $P = 0.05$), whereas sex differences in activity measures were non-significant for other ages.

Table 5.2. GAM models for age, sex, and reproductive status effects on AEE.

	Without Reproductive Status				With Reproductive Status			
	B	SE	t	P	B	SE	t	P
Intercept	572.60	49.49	11.57	<.01	653.83	74.68	8.76	<.01
Sex (Male)	166.62	72.93	2.29	.03	0.73	96.62	0.01	.99
PL					-136.84	96.27	-1.42	.16
Sex (Male) x PL					473.82	145.41	3.26	<.01
	edf	Ref.df	F	P	edf	Ref.df	F	P
S(Age) Females	1.00	1.00	0.00	.99	1.00	1.00	0.05	.83
S(Age) Males	3.44	4.24	1.61	.19	1.00	1.00	0.21	.65
Model R² (adj)	.18				.23			

When the effect of pregnancy and lactation on activity levels was examined, a clearer picture of the sex differences in activity profiles emerged. We coded a variable indicating whether the individual (for females) or the individual's partner (for males) was pregnant or lactating (PL) and included this in our GAM (Table 5.2). Including this variable eliminated the age effect and revealed a significant interaction between having a PL female in the household and sex differences in AEE ($B = 473$ kcal/d, $t = 3.26$, $P < 0.01$). Removing the age terms from the model, we ran two-way ANOVAs on AEE, AC, and PAL with sex and the PL variable. The interaction between sex and family reproductive status was highly significant for each activity measure (PAL: $F_{1,45} = 18.8$, $P < 0.01$; AEE: $F_{1,45} = 10.9$, $P < 0.01$; ACs: $F_{1,45} = 6.0$, $P = 0.02$). Including variables for season of collection and whether the data was collected on a weekend did not

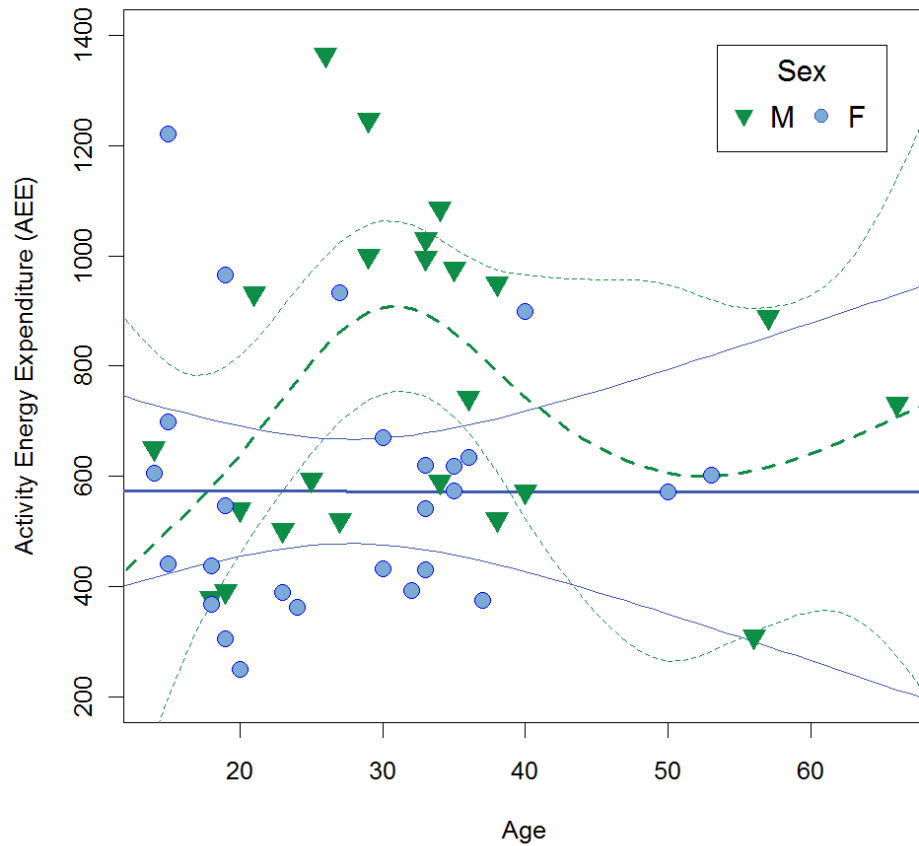


Figure 5.1. Activity energy expenditure (kcal/d) by sex and age. Lines are thin plate splines from the generalized additive model in Table 2 that does not control for reproductive status. Solid lines and circles are females, while dashed lines and triangles are males. Lighter colored lines are 95% confidence intervals for the spline value. Note that confidence intervals for males and females overlap except for ages from ~25 to ~35.

significantly alter these results (PAL: $F_{1,43} = 15.96$, $P < 0.01$; AEE: $F_{1,43} = 8.45$, $P < 0.01$; ACs: $F_{1,43} = 4.46$, $P = 0.04$). Moreover, neither season nor weekend remained significant in any model.

We next ran post-hoc pairwise comparisons between groups (Figure 5.2). In households without a pregnant or lactating female there were no significant sex

differences in activity levels. In families where the female was either pregnant or lactating, males had significantly higher activity levels than females (AEE: $t = 4.79$, $P < 0.01$; PAL: $t = 5.80$, $P < 0.01$; AC: $t = 3.28$, Bonferroni corrected $P < 0.01$). Compared to males without PL partners, males with PL partners had significantly higher AEEs

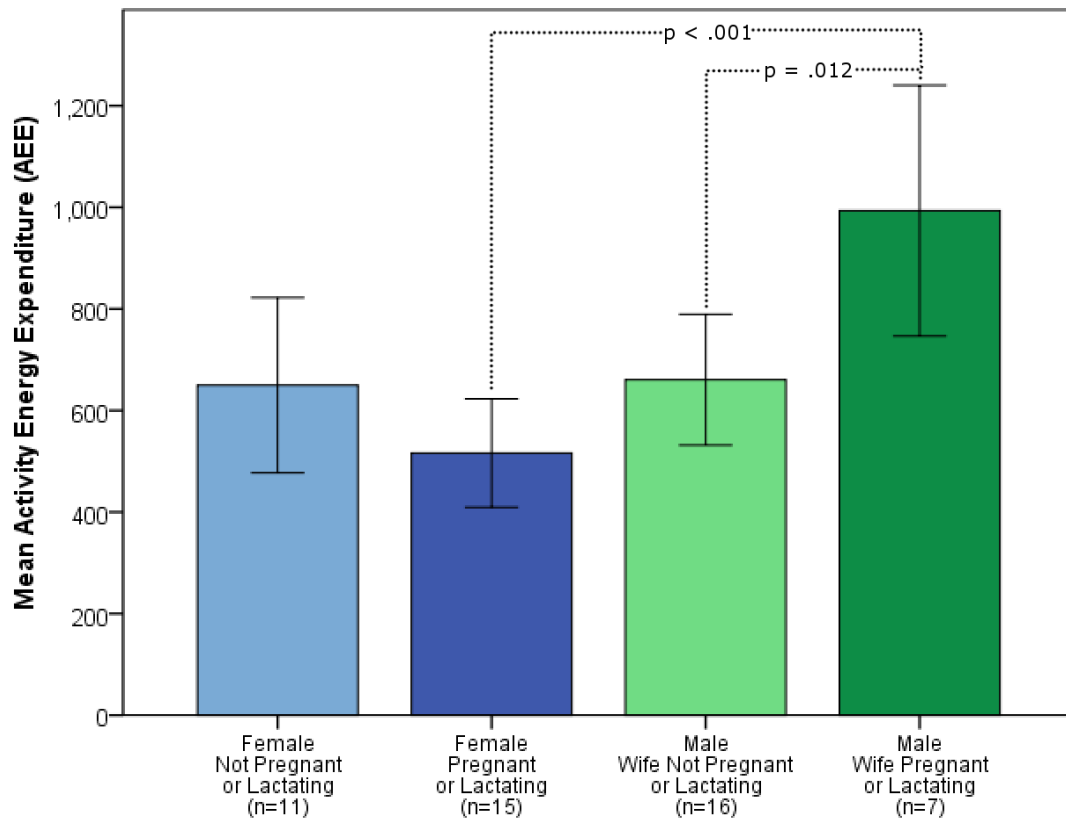


Figure 5.2. Mean activity energy expenditure (AEE) by sex and reproductive status. Error bars are 95% confidence intervals for the mean. Two-tailed t-tests were used for pairwise comparisons between males and females in each reproductive category and between reproductive status within each sex. A Bonferroni correction was applied to P -values to account for multiple testing. Non-significant P -values are not shown.

($t = 2.95$, Bonferroni corrected $P = 0.01$), and PALs ($t = 2.42$, Bonferroni corrected $P = 0.05$), but not higher ACs (Table 5.3). PL females had lower PALs than non-PL females

($t = 3.86$, $P < 0.01$) but not AEEs or ACs. Table 5.4 presents descriptive statistics for females based on reproductive status and illustrates that group differences are not the result of activity levels but rather the higher BMRs found in the PL sub-group.

Of the 49 study participants, 13 had partners who were also in the study. These included five monogamous couples and one male with two wives. Of these, four women were pregnant or lactating and three were not (Figure 5.3). Note that the male in couple 3 is polygynous: he lives primarily with female 3, but is also married to female 4, who lives in a house a short distance away. Both females were lactating at the time of the study. Couple 6 is a young NPNL pair and appears to follow the pattern of PL couples; this characteristic may be explained by their ownership of a small store where they sell a small amount of basic market goods including noodles, rice, eggs, soap, and oil. The NPNL wife often tends the store while caring for her two young children, which may account for why her activity level is relatively low compared to that of her husband. Although the sample size is small, a trend towards higher activity output by males with PL counterparts is evident.

Table 5.3. Descriptive statistics for males with pregnant or lactating partners (PL) compared to males without PL partners^{a,b}

	Males without PL partners (n=16)	Males with PL wives (n=7)	
Measure	Mean (SD)	Mean (SD)	P
Age (years)	33.4 (15.4)	31.0 (4.4)	ns
Height (cm)	154.2 (9.7)	158.8 (4.8)	ns
Weight (kg)	55.1 (10.7)	65.4 (5.0)	*
BMI (kg/m ²) ^c	22.9 (2.6)	26.0 (1.8)	*
TDEE (kcal/d)	2035 (347)	2498 (313)	*
BMR (kcal/d) ^d	1360 (174)	1505 (112)	*
AC ^e	263,493 (116,902)	350,798 (142,398)	ns
AEE (kcal/d) ^f	661 (242)	993 (267)	*
PAL ^g	1.48 (0.16)	1.66 (0.17)	*

Table 5.4. Descriptive statistics for females during various reproductive states^{a,c}

	Pregnant (n=5)	Lactating (n=10)	NPNL (n=11)	
Measure	Mean (SD)	Mean (SD)	Mean (SD)	P
Age (years)	24.4 (4.5)	31.6 (7.3)	25.9 (14.3)	ns
Height (cm)	148.2 (4)	146.7 (4.4)	144.3 (4.0)	ns
Weight (kg)	54.1 (7.3)	50.6 (8.4)	47.6 (8.2)	ns
BMI (kg/m ²) ^c	24.6 (2.8)	23.4 (2.7)	22.8 (2.9)	ns
BMR (kcal/d) ^d	1416 (193)	1769 (194)	1161 (99)	***
TDEE (kcal/d)	1927 (340)	2331 (234)	1811 (325)	***
AC ^e	191,847 (121,662)	193,014 (67,560)	268,912 (112,910)	ns
PAL ^g	1.36 (0.15)	1.3 (0.08)	1.55 (0.2)	*
AEE (kcal/d) ^f	511 (238)	519 (180)	650 (257)	ns

^a TDEE, total daily energy expenditure; BMR, basal metabolic rate; AEE, activity energy expenditure; PAL, physical activity level.

^b Differences between the groups indicated are significant at $p < 0.05$ in an independent samples t-test with equal variance assumed; ns = no significance.

^c Differences between females are statistically significant at $*P < 0.05$; $***P < 0.001$; ns = no significance.

^d Weight divided by height in meters squared (kg/m²).

^e Calculated using the Oxford equations (Henry, 2005).

^f Activity counts (AC) represent the frequency and intensity of acceleration events that occur during a user-defined epoch.

^g AEE from activity counts based on two-regression equation (Heil, 2006).

^h TDEE/BMR.

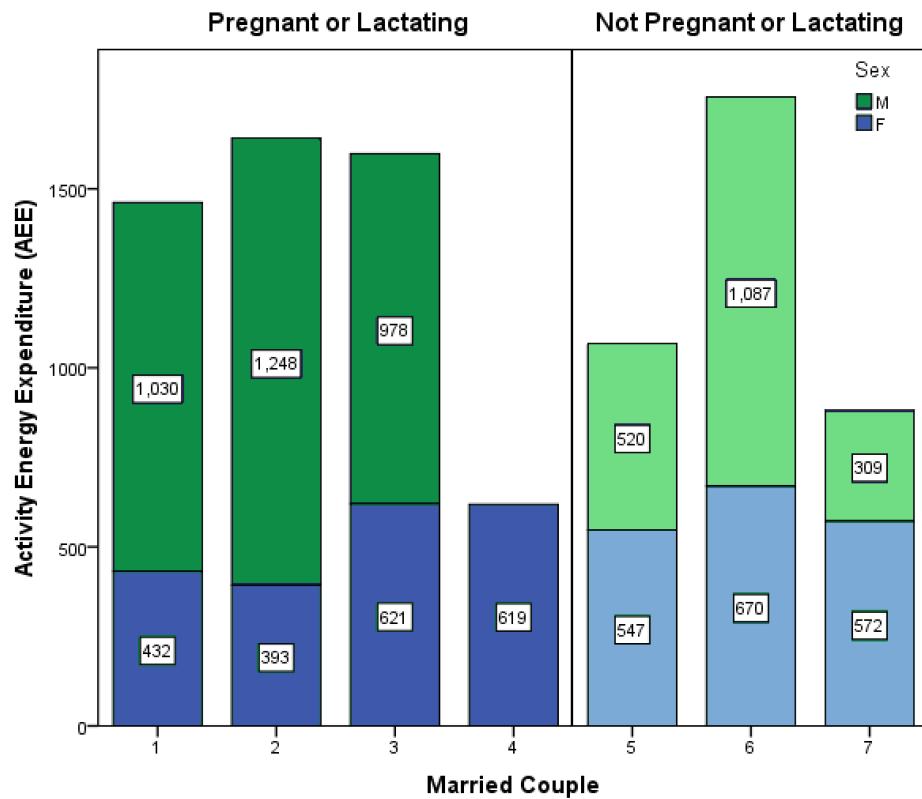


Figure 5.3. Activity energy expenditure (AEE) in kcal/d for married couples for which both partners participated in this study.

DISCUSSION

The present study used accelerometry to investigate physical activity patterns in Upano River Valley Shuar who are currently undergoing rapid economic development and lifestyle change. Despite the shift towards increasing market integration, Shuar from the region continue to be dependent on subsistence activities, particularly horticultural production. Most people in the study community actively engage in subsistence activities, spending most mornings and afternoons at their *chacras* (gardens) cultivating the land and harvesting cassava (yuca), plantains, papa china (a local tuber), maize, legumes, and other staple foods.

Physical activity levels for Shuar males are approximately 1.54, while Shuar females exhibit significantly lower PALs of 1.42. Based on FAO/WHO/UNU (2004) standards, average PALs among the Shuar are within the light (PAL ~1.4) to moderate (PAL ~1.75) range. Other measures of physical activity, including AC and AEE, were also significantly lower among females than males.

It is difficult to compare activity profiles between the Shuar and other subsistence populations since other energetics studies have used a variety of different techniques to assess physical activity (e.g., DLW, HR monitors, and time allocation). Few studies to date have used accelerometry to estimate energy expenditure in remote field settings. Further, because various types of accelerometers generate different output (e.g., uni-axial versus tri-axial accelerometers), comparing activity levels between studies using different types of accelerometers can also be problematic. Converting activity counts into common energetic parameters using published regression equations is one means of overcoming the obstacle of comparing between accelerometer types. However, comparisons between populations using different activity techniques will always introduce error so results must be interpreted with caution. Nevertheless, these problems of inter-study comparison are not limited to this study. Until researchers employ a standardized means of recording activity data, or systematize analytic procedures for cross-methodological comparisons, such approaches remain essential in order to address important issues in human biology, including those related to global health.

Shuar activity values are low when compared to other subsistence populations (Table 5.5). Using the DLW method, Snodgrass et al. (2006) found that among the indigenous Yakut of Siberia, individuals who participated in fewer subsistence activities

had significantly lower activity levels and consumed a greater percentage of market foods than individuals with more traditional lifestyles. The Shuar participant community is in the process of transitioning towards a market economy, so low activity levels might be similarly related to decreased participation in subsistence behaviors and increased market good consumption. However, among Gambian (Lawrence and Whitehead, 1988) and Andean Aymara (Kashiwazaki et al., 1995) rates of energy expenditure were attributed not to short bursts of intensive activity, but to extended periods of activities with moderate energy cost. In fact, in a comparison of activity levels across populations in developing countries, Dufour and Piperata (2008) show that subsistence activities may not necessarily involve high, though highly variable, energy expenditure, and therefore they caution researchers from making generalizations about the energy needs of subsistence-based lifestyles. Lower than expected activity values among the Shuar may therefore be explained by a focus on habitual tasks of long duration requiring relatively low energy output (e.g., gathering legumes, clearing weeds from *chacras*, or processing yuca for the production of *chicha* [a locally-made fermented beverage]). Alternately, the low values could be due to the fact that Shuar activity was measured only during dry season months. Although we documented variation in activity levels across the dry season, it seems unlikely that wet season activity levels would be higher, given that Shuar devote less time to hunting, fishing, and garden clearing activities during this rainier period. Future data collection is necessary to test this proposition.

Sex Differences in Activity Patterns

Energetics provides a powerful tool with which to study life history trade-offs under different ecological conditions (Snodgrass, 2011). Life history theory examines the age- and context-dependent resource allocations that organisms make between competing demands such as growth, reproduction, and somatic maintenance (Charnov and Schaffer, 1973; Gadgil and Bossert, 1970; Hill and Hurtado, 1996; Hill and Kaplan, 1999; Lessels, 1991; Stearns, 1976). For successful reproduction, women in subsistence-level societies must allocate sufficient resources to energy intensive pregnancy and lactation for extended periods of time, while simultaneously maintaining essential metabolic function and the physical activity levels necessary to acquire food, care for offspring, and engage in other critical daily activities (Bogin, 1999; Ellison, 1994; Hrdy, 1999). Biological and behavioral strategies regulating the balance of energy allocation to costly reproductive effort (e.g., pregnancy and lactation) and the energetic costs of food acquisition, somatic maintenance, and parental investment are therefore expected to have evolved. Strategies may involve increasing dietary intake or minimizing energy expenditure, such as through adjusting metabolic efficiency, drawing on fat stores, or reducing activity costs (Piperata, 2009; Piperata and Dufour, 2007; Ulijaszek, 1995). Adoption of any of these energetic strategies may be dependent upon the degree of social support available to the woman, including from husbands or older non-reproductive aged offspring who contribute substantially to the high costs of child-rearing. This investment from other community members would support heightened energy demands during critical reproductive periods by way of provisioning for young infants and mothers themselves (Hill and Hurtado, 2009; Hrdy, 2005; Marlowe, 2003; Meehan, 2009; Reiches et al., 2009). From this

Table 5.5. Activity profile comparison between the Shuar and published data sets^a

Population	Source	Method ^b	Sex	n	Age (years)	Weight (kg)	TDEE (kcal/d)	AEE (kcal/d)	BMR (kcal/d)	PAL	Activity Level ^c
Shuar	<i>This study</i>	ACC	M	23	32.7 (12.9)	58.2 (10.5)	2175.6 (395.5)	761.8 (289.5)	1403.3 (169.6)	1.54	L
			F	26	27.8 (10.7)	50.0 (8.2)	2033.2 (374.4)	572.7 (227.3)	1443.7 (317.8)	1.42	L
Coastal Ecuadoreans	<i>Leonard et al., 1995</i>	HRM	M	5	49 (19)	61.3 (9.2)	2414.7 (353.5)	885.7	1529	1.58	L-M
			F	5	34 (16)	55.7 (11.5)	1992.0 (138.5)	767	1225	1.63	L-M
Highland Ecuadoreans	<i>Leonard et al., 1995</i>	HRM	M	11	32 (12)	61.3 (9.2)	3807.2 (759.5)	2207.2	1600	2.39	H
			F	11	40 (13)	55.7 (11.5)	2457.7 (707.0)	1205.7	1252	1.97	H
Ache (Paraguay)	<i>Hill et al., 1984; Leonard and Robertson, 1992</i>	TIME	M	n/a	n/a	59.6	3327	1796	1531	2.17	H
			F	n/a	n/a	51.8	2626	1232	1394	1.88	H
Yakut (Siberia)	<i>Snodgrass et al., 2006</i>	DLW	M	14	33.3 (9.8)	72.2 (14.5)	3100.9 (1939.0)	1254.3 (168.7)	1848.7 (69.1)	1.68	L-M
			F	14	31.4 (9.6)	65.2 (19.5)	2297.7 (124.7)	765.5 (84.8)	1533.9 (65.0)	1.5	L
Burkina Faso	<i>Bleiberg et al., 1981</i>	TIME	M	11	45 (3.32)	56.5 (1.75)	2261 (67.4)	n/a	n/a	1.36	L
			F	14	30.6 (2.63)	49.9 (0.92)	2144 (49.1)	n/a	n/a	1.54	L

Table 5.5. (continued)

!Kung (Botswana)	<i>Lee, 1979; Leonard and Robertson, 1992</i>	TIME	M	n/a	n/a	46	2319	936	1383	1.68	L-M
			F	n/a	n/a	41	1712	613	1099	1.56	L
Huli (PNG)	<i>Yamauchi et al., 2001</i>	HRM	M	15	n/a	63.6 (7.3)	3138 (506.4)	1434	1704	1.84	M-H
			F	12	n/a	53.3 (7.6)	2639 (621)	1248	1391	1.88	H

^a Means (SD), ^b ACC, accelerometry; HRM, heart rate monitors; TIME, time allocation; DLW, doubly labeled water, ^c Activity level is determined using the FAO/WHO/UNU (2004) classification system of light (L), moderate (M), and heavy (H) activity levels.

perspective, female reproduction and energy dynamics may be better understood within the dynamics of a larger cooperative reproductive effort, which would take into account familial energetics or, at the very least, those of mating partners.

The present study was a preliminary investigation of the utility of accelerometry to study potential sex differences in energy expenditure and considered activity patterns based on reproductive status. Males had significantly higher PALs than females, but as over half of the women in the study were either pregnant or lactating, using PALs are problematic when comparing between sexes. When AC and AEE were used as the energy parameters, significant sex differences were maintained.

Male AEE was more variable than female AEE, with female activity values generally clustering under 600 kcal/d. Male activity peaked between the ages of 25-35 years. Almost all pregnant and lactating females fell into this age category; they also produced the lowest total energetic output in the study population. There were no significant differences in energetic output, as measured by AEE and AC, between females who were pregnant or lactating and those who were not. PALs did vary significantly between PL and NPNL females, a difference attributable to the higher BMR costs of energetically costly reproductive states. Interestingly, while BMR requirements are higher in PL women, their work output is fairly constant, comparable to that of NPNL female participants. A more critical observation is the significantly higher activity levels among males with PL partners compared to other males. Although the sample size is small, the data suggest that while the higher basal metabolic costs for PL females do not cause a decline in their energy expenditure, it appears to incite a compensatory increase in the activity output of male partners.

In some subsistence populations, women reduce energy expenditure during pregnancy and lactation, which appears to at least partially offset elevated energy needs (Panter-Brick, 1993; Piperata and Dufour, 2007). A decrease in female economic contribution can present costs to household production and, therefore, may prompt a compensatory increase in male provisioning as noted among the Ache and Hiwi foragers (Hurtado et al., 1992) and the Hadza hunter-gatherers (Marlowe, 2003). Our data suggest that the increased energetic needs of pregnant or lactating females initiates behavioral changes, not by a reduction in activity output by females themselves, but rather through an increase in the activity levels of other family members, more specifically, their male partners. The necessity to contribute to work output may demand women to maintain their baseline activity patterns while men are increasing their own economic contribution as a means of offsetting their partner's up-regulated metabolic costs.

Although the sample size in this preliminary study is small, the low PALs, AEEs and ACs among pregnant and lactating women coupled with significantly higher activity levels among men with pregnant or lactating partners suggest a cooperative effort to cope with high maternal energy demands. Despite the rapid shift towards increasing market integration among Shuar in the Upano Valley, the results of the present study are consistent with data from other subsistence-based populations in which male and children's subsistence activities appear to be critical for supplementing women's energetic needs during pregnancy and lactation (e.g., Ivey, 2000; Marlowe, 2003; Reiche et al., 2009). Although these data logically fit well with a cooperative breeding model of human reproduction (Hill and Hurtado, 2009; Hrdy, 2000), it should be emphasized that our results are based on a small sample, and further research is needed to confirm them.

Nevertheless, the results do indicate the potential utility of accelerometry for investigating such issues.

Field Methods of Measuring Physical Activity

Measurement of physical activity outside of a laboratory setting is notoriously difficult and no method is without problems (Snodgrass, 2011). A variety of methods are available for estimating physical activity levels and TDEE, the most accurate of which is the doubly labeled water (DLW) technique. DLW yields a measure of TDEE over the course of 1-2 weeks, based on the elimination rates of two labeled stable isotopes from the body. This technique is generally accepted as the most accurate measure of energy expenditure among free-living humans, and has been used extensively in clinical studies in industrial nations (Black et al., 1996; FNB/IOM, 2002; Speakman, 1997). DLW provides an excellent basis for analysis of average TDEE across longer timescales and is particularly useful for characterizing and comparing general activity levels between men and women, or between populations. Unfortunately, the high cost of DLW has limited its use in populations in the developing world (Coward, 1998). When data are available from non-Western groups, sample sizes are generally small (less than 50 individuals), which makes comparisons between populations problematic. Further, DLW does not provide information on daily or hourly fluctuations in energy use, or the energy costs associated with specific activities.

The time allocation technique is the most commonly used method for estimating activity levels in subsistence populations and can provide a record of daily or hourly changes in activity. This method estimates physical activity by either observing or using

interview techniques to estimate the amount of time an individual spends in different activities, and combining this with published information on energy costs of each activity (e.g., FAO/WHO/UNU, 2004; James and Schofield, 1990; Ulijaszek, 1995). Summing the results for one day thereby provides an estimate of TDEE. However, published values for the energy costs of different activities are typically based on only a few individuals and no data exist for many common activities researchers observe in field settings. In addition to being time and labor-intensive, the time allocation technique often substantially underestimates energetic parameters, since it does not record many involuntary activities (e.g., fidgeting). This method is most inaccurate at moderate to high activity levels, often underestimating TDEE by at least 15% (Kashiwazaki et al., 2009; Leonard et al., 1997; Spurr et al., 1996).

Heart rate (HR) monitoring can also be used to estimate physical activity, based on the known relationship of HR to energy expenditure. The HR monitor records HR at a designated interval (e.g., every minute) while participants wear the monitors during waking hours over the course of several days. At the end of the measurement period, HR data is downloaded from the instrument and energy expenditure is calculated. Despite proven accuracy of the measurement compared to DLW (e.g., Kashiwazaki, 1999), there are also limitations to this technique. For example, in order to obtain accurate results, one key issue involves the need for individual calibration of HR with energy expenditure. This process requires the relationship of HR to energy expenditure to be established for each individual both at rest and during graded sub-maximal exercise (Leonard, 2003).

Technological advances in accelerometry promise to help overcome some of the challenges of affordably quantifying physical activity under field conditions.

Accelerometers can objectively measure the movement of the body by detecting and recording acceleration in one or multiple planes (Chen and Bassett, 2005; Gerdhem et al., 2008; Heil, 2006). The devices are durable, non-intrusive, relatively inexpensive, and can provide detailed information on physical activity patterns, including the duration and intensity of activity. Several accelerometer devices, including the Actical, have been validated and show a high correlation with oxygen consumption and DLW (Heil, 2006; Hoos et al., 2003; Plasqui and Westerterp, 2007). However, it is important to note that most accelerometer validation studies to date have been conducted in laboratory-based samples of participants from industrialized nations.

The Actical is one of the more extensively validated commercially available accelerometer devices (Plasqui and Westerterp, 2007). Acticals are small, rugged data loggers, equipped with a highly sensitive multi-directional accelerometer. The ability to sense motion in more than a single plane is an advantage for measuring complex human movements (Heil, 2006). The Actical accelerometer generates a variable voltage based on amplitude and frequency of motion and produces an electrical current that varies in magnitude. An increase in the intensity of motion will result in an increase in voltage; this information is integrated over a user-selected epoch and recorded in onboard memory in the form of “Activity Counts.” An activity count (AC) is an arbitrary dimensionless unit that varies between different brands of accelerometer devices, so direct comparisons of raw activity counts from assorted devices are not inherently meaningful. However, ACs can be used to calculate AEE or TDEE using the device software or through accelerometer-specific published regression equations allowing for cross-methodological comparisons yet, as noted earlier, with limitations.

As with other means of activity recording there are restrictions to using accelerometry to determine activity levels and energy expenditure. These limitations vary with the type of device used and how it is utilized. For example, placement at the hip captures gross body movement but may be less sensitive to upper body movements or the energetic costs associated with load-carrying, which may result in underestimations of physical activity output (Swartz et al., 2000). Nevertheless, several studies have documented the validity of hip-placed activity monitors for estimating whole-body energy expenditure when compared to alternative sites such as the wrist and ankle (Freedson et al., 1998; Hendelman et al., 2000; Swartz et al., 2000). Another limitation is that accelerometers do not provide the context of activity, although this may be obtained by simultaneously incorporating other techniques (e.g., daily recall or direct observation). Further, as mentioned earlier, an issue with accelerometer devices that can be extended to all types of activity monitors is that comparison of activity across populations using different models of accelerometers will entail greater measurement error than will studies that use the same device, thus small but statistically significant differences must be interpreted cautiously. However, if one is interested in documenting relative activity differences within a population using identical devices and methods, these are minor limitations.

The current study represents a preliminary step towards greater use of accelerometry in non-clinical, non-Westernized contexts. The results presented here are consistent with predictions based on a life history theory approach to understanding energetics, thus illustrating the potential usefulness of accelerometer devices in addressing such questions. Clearly, at this preliminary stage, comparisons of activity

patterns with other populations must be interpreted with caution. However, given the importance of recognizing how shifts in energy expenditure can inform emerging global health issues, and the scant nature of these data from subsistence-level populations, researchers must overcome the urge to wait until a perfect dataset is available prior to reporting their findings.

CONCLUSIONS

The present study investigates energetic patterns in an Ecuadorian Shuar community undergoing rapid economic development and lifestyle change. It provides preliminary evidence that activity levels in this transitioning Shuar community are modest, especially when compared with other subsistence populations. This study also investigated life history trade-offs related to female reproductive status. With regard to the Shuar participant community, it suggests that female reproduction and energy dynamics may be better understood within the dynamics of a larger cooperative reproductive effort, including that of mating partners. Despite a shift towards market integration, Shuar pregnant and lactating females may adopt a cooperative strategy noted in other subsistence-based populations where male participation in subsistence activities is higher in order to compensate for their partners' elevated reproductive costs. Finally, this study demonstrates the promise of accelerometry use under field conditions and argues that despite some limitations, this technique offers useful information regarding population-level activity patterns.

BRIDGE TO CHAPTER VI

In Chapter V, I examine the behavioral strategies that may be adopted by Shuar participants in an Upano Valley community in order to meet the costs of female reproduction. Pregnant and lactating females seem to be adopting a strategy noted in other subsistence populations where male participation in subsistence activities increases to compensate for their partners' elevated reproductive costs. Chapter V also shows that among Shuar in this study, activity levels not as high as one might assume for a subsistence-based population and in fact, are relatively low compared to other non-Western populations. This chapter also explored the utility of accelerometry in human biology research. In the final concluding chapter, I further develop the connections between the studies in this dissertation and present general conclusions from these studies.

CHAPTER VI

CONCLUSIONS AND FUTURE DIRECTIONS

A basic tenet of life history theory is that organisms face trade-offs in the allocation of resources between competing demands. Natural selection has shaped mechanisms that generate age and context dependent strategies that regulate these investments across the life course. In an energy-restricted environment, fundamental trade-offs generally exist among the processes of somatic growth and maintenance, and reproduction. For women, in particular, reproductive effort entails exceptionally high costs to support the fetus during pregnancy and the infant during lactation, and this often leads to reduced energy and nutrients available for growth and somatic maintenance.

The overarching goal of the studies presented in this dissertation was to examine the biocultural adjustments that Shuar females make in response to these elevated energetic and nutrient costs of reproduction; for this reason, life history theory serves as the critical organizing paradigm of this dissertation. The first research objective was to examine trade-offs in energy use during critical female reproductive states, and the biosocial accommodations that are made in response to the high costs of these states. Specifically, this objective was achieved by investigating the relationship between reproductive variables and skeletal health among Shuar females, and by documenting the

behavioral adjustments made by women and their partners to offset the high energetic needs of reproduction. The second objective was to investigate sex- and age-related patterns of bone density among Shuar, as well as neighboring non-Shuar *Colonos*, and to establish a profile of activity patterns for the Shuar population.

In this concluding chapter, I synthesize the themes discussed in preceding chapters, and outline important avenues for future research.

SYNTHETIC OVERVIEW

In Chapter III, I presented a normative sex and age-related bone health profile for Indigenous Shuar and non-indigenous *Colono* populations. Minimal data are available on bone health from populations living in developing countries, and no normative datasets were previously available for either population. The results from this study demonstrate that both Shuar and non-Shuar *Colonos* experience age-related declines in bone density similar to those found among Western populations. These results contribute to cross-cultural data on skeletal health, and provide support for age-related influences on bone loss. Of course, more cross-cultural data from non-industrialized groups are necessary to elucidate the extent to which advanced age affects bone integrity within and across populations. However, while the gradual decline in bone health with age appears to be a universal aspect of the human condition, osteoporosis is not an inevitable end result. This condition may be curtailed by adjusting modifiable factors (e.g., lifestyle, activity, diet) that contribute to the peak amount of bone attainable and the rate of bone loss in adulthood.

Compared to other reference populations, Shuar have relatively high peak bone mass values, which afford them greater bone health values throughout life; similar patterns were found for Shuar women, although values decline in the postmenopausal years. Among Shuar men, relatively higher BMD is a stable characteristic even in advanced age; among Shuar women, BMD values are consistently higher than among *Colono* women, until peri-menopause. As in other populations, the onset of menopause appears to be associated with more rapid decrease in bone density, although it is important to remember that pre-menopausal Shuar women have higher bone mass than found in most other populations. Furthermore, the Shuar postmenopausal sample size is significantly smaller than the *Colono* group, so the extent to which the pattern of BMD values observed in postmenopausal Shuar women can be generalized to the larger population must be tested in future research by increasing the sample size of this cohort.

The relatively high bone density values documented among Shuar, particularly males, compared to other global reference populations is a particularly important result from Chapter III. This result makes a compelling case for reconsideration of what constitutes “normal” bone health in Western countries, and how bone density values should be interpreted for diagnostic purposes. As discussed in Chapter II, skeletal and general health profiles from Western industrialized populations reflect the collision of modern lifestyles with the ancestral *Homo* genome; specifically, poor-quality diets, reduced activity levels, and demographic shifts from high to low fertility, have major effects on many aspects of contemporary health. Shuar bone density data illustrate that the upper achievable limits of skeletal health may be inhibited by dietary, lifestyle, and reproductive factors in Western populations. A further empirical examination of the

dynamic influences of these factors on skeletal health among the Shuar is clearly necessary (see below: *Activity Patterns and Bone Density*; *Lifestyle Factors and Bone Density*).

Chapter IV systematically investigated hypotheses regarding female reproductive variables and their effects on bone density. In general, existing data that examine this relationship are inconsistent, although research has been largely based on Western, industrialized populations. There is a paucity of data from natural fertility subsistence populations, which is a critical oversight since reproductive patterns in many of these groups are vastly different from industrialized populations. Thus, the effects of high fertility and prolonged lactation on female skeletal health have been largely ignored in the literature, potentially obscuring the current understanding of the relationship between reproduction and BMD. Based on empirical evidence and understandings of the system of calcium turnover during various reproductive states, the hypotheses that were tested in Chapter IV included: 1) women with earlier menarcheal age will have higher BMD; 2) women with an older age at first parturition will have higher BMD; 3) women with more protracted periods of lactation will have lower BMD; and 4) women with longer IBIs will have greater BMD.

The most interesting result finds support for the first hypothesis, that is, that earlier menarche is associated with higher BMD later in life. This finding emphasizes the importance of early environmental and energetic conditions for regulating the timing of reproductive phases, which in turn, plays an integral role in establishing female bone status later in life. Two explanations accompany this finding. First, girls who experience early menarche are exposed to more cycles with fluctuating estrogens than girls with

delayed maturation. These estrogens will help to maintain bone mass during the course of their expanded reproductive life, and therefore the relationship between early menarche and higher postmenopausal BMD may be partly attributed to shifts in these hormones. Secondly, as noted in chapter II, human females exhibit determinate growth, with investment in longitudinal growth being reduced with the onset of menarche; this developmental stage marks the first major energetic investment into reproduction. Because positive energy balance (energy intake > energy expenditure) is critical for the onset of menarche, better nutritional status is not only related to greater stature, but to earlier menarche as well¹. The finding that early menarcheal age is predictive of postmenopausal bone density suggests that bone phenotype responds to these early environmental conditions, and is then canalized into a trajectory of higher BMD, despite transient perturbations during the lifetime. For future research, the importance of early life stages in establishing phenotypic quality throughout the life span emphasizes the importance of conducting skeletal health research among children and adolescents (see below: *Developmental and Early Childhood Origins of Bone Density*).

Confirming the second hypothesis, age at first parturition was positively associated with bone density among pre-menopausal lactating women, although not for postmenopausal women. Again, this may reflect a classic age-related trade-off between somatic and reproductive investment, with bone density traded off against reproduction. As discussed in Chapter IV, trade-offs between skeletal growth and early reproduction

¹ Some studies demonstrate that social and life history factors, such as extrinsic mortality risk, also appear to shape decisions of allocation. For example, because of a higher extrinsic mortality risk, an organism may anticipate a shorter life and therefore invests less into body size gain and accelerates maturation to ensure reproductive fitness (e.g., Ellison et al., 1993; Gluckman and Hanson, 2006; Jasienska et al., 2006).

have been documented in several studies (e.g., Hayslip et al., 1989; Kent et al., 1990; Kent et al., 1993; Sowers et al., 1993). Rooted in the principles of life history, bone density phenotype appears to be canalized by the timing of early life history events—a later age at first reproduction may allow more time for investment in accruing bone density, thereby affording these females higher bone density. In contrast, an early age at first pregnancy may channel energy and/or nutrients away from bone density investment.

The third and fourth hypotheses are inter-connected since lactation patterns shape IBI in natural fertility populations. Therefore, IBI and lactation are considered jointly here. Although cross-sectional rather than longitudinal, Chapter IV shows that the effect of lactation on bone density appears to be short-term and transient, with no significant long-term effects on bone density in postmenopausal life. In general, epidemiological and clinical research conducted with women from industrialized populations reach similar conclusions, showing that decreased bone density during lactation is recovered during weaning (e.g., Karlsson et al., 2005; Laskey and Prentice, 1997). This pattern of bone recovery demonstrates that the system of calcium turnover has evolved relatively efficient mechanisms to avoid potential depletion of maternal bone density over the course of the reproductive period. The evolution of such mechanisms is further supported by the finding that short IBIs did not have a deleterious impact on Shuar maternal skeletal health either in the pre- and postmenopausal life.

Chapter IV also shows that, in addition to lactation, parity has no significant effect on skeletal health among Shuar women, although this was not presented as a specific hypothesis. Independent of the influence of lactation, past studies have presented mixed results regarding the effects of offspring number on maternal BMD. While some

studies find a positive association between multiparity and bone health, perhaps due to the protective effects of increased intestinal calcium absorption, or to weight gain during pregnancy (e.g., Forsmo et al., 2001), other studies document a negative relationship; this negative association is attributed to the cumulative effects of high rates of bone turnover characteristic during pregnancy (e.g., Allali et al., 2007). In studies conducted with industrialized groups, there is no consistent relationship between the number of children and maternal bone mineral density at advanced ages during the lifespan; however, these inconsistent findings may be due to the rather low fertility of women in industrialized contexts. The minimal literature on BMD of females with high fertility and long breastfeeding periods are also inconclusive. Saudi females show significantly lower BMD values among multiparous women (Ghannam et al., 1999); however, this population also has a high prevalence of vitamin D deficiency, which may contribute to low bone mass. Conversely, studies on high fertility Omani (Bererhi et al., 1996) and Finish American women (Henderson et al., 2000) found no correlation between number of children and maternal bone density. Similarly, for equatorial Shuar women, who report as many as 15 offspring at completed fertility, offspring quantity has no effect on postmenopausal bone density. One possible explanation for this relationship among the Shuar may be that the capacity for multiple pregnancies reflects better overall phenotypic quality.

As discussed in Chapter II and IV, fecundity and fertility is regulated by energy availability (i.e., energy balance, energy storage, and energy flux) (Ellison, 2003). Women with good nutritional status generally experience earlier menarche and, therefore, have higher levels of reproductive hormones, specifically estrogens (Jasienska, 2009).

Higher estrogen levels are also associated with heightened probability of conception (Jasienska, 2009; Lipson and Ellison, 1996; Venners et al., 2006), which may theoretically lead to higher parity than nutritionally stressed women. In most natural fertility populations, women with relatively good nutritional status are thus likely to have higher parity, as well as higher bone density; women with lower parity will tend to have lower BMD. Therefore, any possible negative effect of high parity on BMD may not be detectable because it is those women with higher baseline bone density, potentially the result of more high-estrogen cycles, who also have overall positive energy balance and, thus, more children. The question that this prompts, then, is why multiple pregnancies and lactation cycles lead to depleted maternal fat and lean tissue stores, while similar effects are not demonstrated for skeletal reserves? After all, patterns of maternal depletion have been observed globally in nutritionally-stressed populations, in New Guinea (Garner, 1994; Tracer, 1991), India (Belevady, 1979), the Philippines (Adair, 1992), Peru (Yu and Shephard, 1998), Zaire (Pagezy, 1984), and Namibia (Kirchengast and Winkler, 1996), and even small but significant net loss of fat mass with each live birth has been documented in well-nourished women (Lassek and Gaulin, 2006; Rodrigues and Da Costa, 2001). A brief consideration of the role of fat deposition during reproduction may shed some light onto this issue.

The accumulation of fat in human evolution has been a major adaptive feature in our species, serving to buffer fluctuations in energy supply, and regulate reproduction and immune function (Kuzawa, 1998; Wells, 2005). Among women, sufficient fat deposition, and positive energy balance, play key role in reproductive fitness, partly through the regulation of menarche and conception. Furthermore, fat stores available at conception

and deposited during the first trimester, particularly in the gluteal femoral region, also fund the high costs of lactation and offspring brain growth. The additional energy requirements of lactation for exclusively breastfeeding women is estimated at 675 kcal/day, and one kilogram of maternal body fat yield may yield approximately 9,500 kcal of energy (Dewey, 1997; FAO/UNU/WHO, 2004). However, it appears that it is not simply fat-as-energy that is important during gestation and lactation, but perhaps more critical is the composition of fat. In addition to acting as reserves that provide energy necessary to maintain pregnancy and lactation, gluteal-femoral fat has high concentrations of certain fatty acids that are essential to fetal and infant brain growth (Lassek and Gaulin, 2006). Specifically, long-chain polyunsaturated fatty acids (LCPUFA), arachidonic acid (AA), and perhaps most importantly, docosahexaenoic acid (DHA) are key resources for neural development (Phinney et al., 1994). Therefore, the mobilization of larger amounts of fat during pregnancy and lactation may serve an adaptive function that meets the energetic and essential fatty acid requirements of the developing fetal and neonatal brain. In short then, because fat stores are critical for satisfying the needs of the growing human brain, the benefits of maternal mobilization of fat stores are so great that fat tissue cannot be entirely restored during repeated and closely spaced pregnancies, a problem exacerbated in an energy-stressed environment.

In an analogous way, maternal skeletal calcium stores are drawn upon to support offspring skeletal growth during pregnancy and lactation, so one might hypothesize that multiparity and breastfeeding multiple children would lead to a higher risk of osteopenia or osteoporosis later in life. Yet, it appears that the developmental requirements of calcium for fetal and neonatal skeleton growth may not be as challenging for the maternal

body to replenish than is body fat. Therefore, it may be the case that replenishing calcium stores is more easily achieved, perhaps because of greater availability of this nutrient from the environment (see Chapter II), than maternal fat reserves, thereby facilitating the recovery of bone loss between reproductive cycles.

Clearly, energy stores and availability are critical for female reproduction. Exactly how women access the energy stores necessary for reproduction is less obvious. To shed light on this phenomenon, Chapter V examines the energy use during reproduction in one rural Shuar community. In other cross-cultural studies, bio-behavioral strategies adopted by pregnant and lactating women have been shown to compensate for their elevated metabolic costs. Reductions in energy expenditure, increased caloric intake, increased investment by partners or kin, or a combination of these strategies are among some of the options available to women to offset the high costs of reproduction. These strategies are clearly dependent on energy availability in the local environment, as well as the availability of social support necessary to accommodate the woman's energetic state. Cross-sectional research presented in Chapter V shows that pregnant and lactating (P/L) Shuar women have physical activity levels similar to those of other women, suggesting that P/L women do not alter their subsistence activities. In other words, P/L women appear to continue to expend energy towards work output, but do not increase their energetic output in order to acquire more food to satisfy their elevated metabolic costs. However, males with pregnant and lactating wives do appear to increase their energetic output compared to other males. This suggests that Shuar males may be supporting the higher costs of their partner's pregnancy and lactation by increasing their contribution to subsistence activities. These results are consistent with data from other subsistence-based

populations in which subsistence activities of kin appear to be critical for supplementing women's energetic needs during pregnancy and lactation (e.g., Ivey, 2000; Marlowe, 2003; Reiche et al., 2009). Although the Chapter V results are based on a small sample, these data fit well with research on the Hadza that show that compared to other males, men with P/L partners increase their provisioning by providing hunted game and other high energy food resources, such as honey (Marlowe, 2003). Results from Chapter V also align with the cooperative breeding model of human reproduction that emphasizes the critical importance of individuals who invest into growing offspring apart from the mother, or allomothers (Hill and Hurtado, 2009; Hrdy, 2000).

Allomaternal support, according to the cooperative breeding hypothesis, was essential for the evolution of the human reproductive life history. This life history is partially characterized by high fertility coupled with short inter-birth intervals, resulting in multiple dependent offspring. Without support from allomothers, estimates from extant foraging populations suggest that individual forager women would likely not be able to support the energetic demands of their offspring (Kaplan et al., 2000). Allomothers would permit forager females to simultaneously support multiple exorbitantly costly offspring, and this represents a signature feature of the human reproductive pattern (Hrdy, 2000). For Shuar, increased work output by males may result in greater food provisioning to their P/L wives to offset the high energy costs of reproduction. Because allomothering behaviors by grandparents and older siblings have been observed among the Shuar (Blackwell et al., 2009), further exploration into their role in shaping maternal energetic strategies is warranted.

Results from Chapter V also emphasize the misconceptions that continue to abound in the literature, specifically with regard to activity patterns in subsistence-based populations. In a meta-analysis of traditional societies with varying subsistence strategies (e.g., pastoralists, agriculturalists, forager-horticulturalists, hunter-gatherers, swidden cultivators), little significant variation in activity levels were found across subsistence modes, with the exception of pastoralists who exhibit among the lowest PALs (Ulijaszek, 2001). Additional comparisons of subsistence-level populations with Western, sedentary populations show generally lower activity levels in the latter group (Bassett et al., 2004; Hayes et al., 2005; Leonard, 2008; Rode and Shephard 1994a, 1994b; Snodgrass, 2011). Based on this literature, activity data from Shuar forager-horticulturalists do not align with predictions about work output for subsistence -level groups. While the low to moderate PALs recorded for the Shuar may reflect methodological issues with accelerometry and underestimations of energy expenditure, the results may also make a compelling argument for reconsidering current assumptions about the activity levels in subsistence groups: subsistence activities may not necessarily involve high energy expenditure (Dufour and Piperata, 2008). Although Shuar participants exhibit lower than predicted activity levels, they demonstrate extended periods of activity throughout the day, suggesting a focus on habitual tasks of long duration that require relatively low energy output. Therefore, daily patterns of activity may be more informative for understanding energy dynamics than comparatively crude overall PAL values. More detailed studies of Shuar activity patterns, and those of other subsistence-based groups, are essential and planned for the near future. In order to contextualize the activities and link energetic costs to specific activity behaviors, these studies will include simultaneous

recording of data on activity levels using accelerometry and recording of the specific activities engaged in through focal-person follows.

FUTURE DIRECTIONS

In this section, I address potential avenues for future research among the Shuar, in order to better elucidate the dynamic interaction between various factors and skeletal health.

Activity and Bone Density

Physical activity has been associated with accretion and maintenance of bone quality and quantity (see Chapter II). This dissertation has demonstrated that the techniques of calcaneal ultrasonometry and accelerometry may be successfully employed in a remote field setting and thus, integrating activity data with skeletal information is entirely feasible. Unfortunately, because of timing and funding issues, technical problems that arose with using an ultrasonometer in Ecuadorian Amazonia, and the limited quantity of accelerometer devices available for this study, I was unable to link physical activity with bone density measures for this dissertation. As a result of these issues, few individuals participated in both the activity research and the skeletal diagnostic study, and even when these data were available, statistical analyses would be uninformative with such a small sample size. Given these pragmatic methodological issues, it is not surprising that there is no published research that has linked physical activity levels with bone density in a non-clinical, non-Western setting. Furthermore, even in the epidemiological and clinical literature where this relationship is considered among

industrialized populations, activity is measured using recalls or questionnaires, such as the International Physical Activity Questionnaire (IPAQ), which are inherently limiting due to their reliance on subjective measures of activity patterns. Additionally, questions used in surveys designed for Western industrialized populations, such as those from the IPAQ, are not easily applied to the subsistence lifestyle of traditionally-living groups. These include questions regarding the amount of time a participant spends engaged in vigorous/moderate/and light activity over the course of a day/week/month. The categories “vigorous,” “moderate,” and “light” are not easily interpretable and answers may vary based on how a participant interprets these words, and cross-cultural comparisons will be particularly difficult. Because of limitations in large-scale qualitative assessments of activity, there is a need, in both Western and non-Western contexts, for integrating bone density data with quantitative output of energy expenditure values from devices like accelerometers. As noted above, plans for gathering these data have been made for upcoming fieldwork with the Shuar.

Developmental Origins of Bone Density

The timing of early developmental events, as discussed in Chapter IV, may be considered proxies for the confluence of environmental conditions and genotypic quality that combine to establish a phenotype of bone density. In turn, bone density phenotype appears to be canalized by these early life events, carrying through into postmenopausal life, despite transient perturbations during the reproductive lifespan. These results, as well those from as other studies (e.g., Godfrey et al., 2001; Kung and Huang, 2007), suggest that the bone integrity may have an epigenetic component. Epigenetics refers to heritable

changes in gene expression that do not involve changes in the sequence of DNA (Kung and Huang, 2007). Several environmental factors interact with genes through epigenetic mechanisms, and these interactions act primarily during early life. With regard to bone density, it appears that early life environmental conditions coupled with individual genotypic quality set the trajectory for bone status that continues across the reproductive lifespan. Although genetic markers regulating bone quantity and quality have been identified (Drake et al., 2001; Van der Weyden et al., 2006), they may explain less than 10% of the variation in individual bone mass (Cooper et al., 2002).

Epigenetic evidence from physiological studies suggests that environmental context begins to shape bone density phenotype early in development, and this phenotype may be predicted by characteristics of maternal body size and lifestyle. Godfrey and colleagues, (2001) found that neonatal bone mass was positively associated with birth weight, birth length, and placental weight, and also highly correlated with maternal birth weight and tricep skinfold thickness at 28 weeks gestation. In contrast, maternal smoking and high maternal physical activity were negatively associated with neonatal bone mass. Additional support for intrauterine origins of adult bone density is garnered from research on endocrine programming (Dennison, et al., 1999; Fall et al., 1998; Phillips et al., 1998). These studies reveal that birth and infant weight may predict basal levels of growth hormone (GH) in adulthood, which help regulate longitudinal bone growth (see Chapter II). In turn, these hormones determined the rate of bone loss in adulthood. These findings are compatible with hypotheses that external stressors on the mother during gestation may alter the sensitivity of the epiphyseal plate to GH. This endocrine programming has the capacity to reduce peak bone mass, potentially reduce bone mineralization, and

predispose one to an accelerated rate of bone loss later in life (Cooper et al., 2002). In sum, these studies suggest that while there is a genetic component to establishing bone density, the phenotypic expression may be highly influenced by environmental conditions during early prenatal life, with resonating effects into adulthood, and potentially into the next generation. Controlling for the effects of myriad early environmental factors is a daunting, if not impossible task, particularly in a remote field setting. Nonetheless, it is important to acknowledge the phenotypic plasticity of the human body, and the extent to which development, growth, maturation, and senescence contribute to physiology and health across an individual's life.

The effect of early developmental life on bone integrity later in life highlights a compelling need for more bone health studies with adolescents. A feasible avenue of study in a remote field setting would be to collect data on adolescent bone density, and consider these values within the context of dietary intake, physical activity (both type and intensity), and socioeconomic status. A comprehensive study would also include interviews with parents regarding the child's nursing behaviors and his/her history of illness. Furthermore, if the child has older and/or younger siblings, this could impact birth spacing, and hence, the child's critical access to breast milk. The role of siblings in determining a child's growth pattern is rooted in principles of life history.

According to this theoretical approach, in a resource-constrained context, there should be trade-offs between offspring quantity and quality. Controlling for availability of resources, the more siblings a child has (i.e., the greater number of offspring a provider has), the less investment is available for each child. Within this framework, child growth and nutrition (i.e., quality) is predicted to be negatively associated with larger family size

(i.e., quantity). Hagen and colleagues (2006) show that these quantity-quality trade-offs are evident among larger Shuar families. Their study found that in households with a greater number of dependents relative to providers, child growth is inhibited. Examining skeletal health within the framework of parental investment, particularly quantity-quality trade-offs, would be one step towards further contextualizing the effects of early developmental life on bone health.

Lifestyle Factors and Bone Density

Developmental factors outlined above may be important to long-term skeletal health, yet some research suggests that characteristics of adult lifestyle are more significant agents of bone loss than fetal or childhood environment (e.g., Kung and Huang, 2007; Pearce et al., 2005). A longitudinal study of thousands of families in Newcastle, UK that tracked participants' health and lifestyles, found that intrauterine and childhood life predicted less than 10% of the variation in BMD among men and less than 1% among women in a 49-51 year old cohort (Pearce et al., 2005). While birth weight does seem to influence skeletal growth, factors in infancy and childhood, including family's socioeconomic status, nursing duration, and age at menarche (females), explained less variation than adult lifestyle factors (e.g., smoking, diet, and physical activity), after adjusting for adult height and weight. Pearce and colleagues (2005) conclude that the observed effect of fetal life on bone health in adulthood may be mediated through adult height and lifestyle. However, it is important to note that the vast array of lifestyle factors that may influence bone integrity, including smoking, alcohol, sedentism, and calorie-dense though nutrient-deficient diets, were not major forces in our

early evolutionary past when the system of calcium turnover evolved. Therefore, the deleterious health consequences of adopting lifestyles dramatically different from our evolutionary condition are not surprising.

Lifestyle variables for Shuar participants were collected in conjunction with bone density measures. The data that were collected included information regarding the ownership of goods that are indicative of increased market integration (MI) (e.g., refrigerators, televisions, and cellular phones), and items more aligned with a traditional and subsistence lifestyle (e.g., fishing nets, blowguns, and machetes). Based on the ownership of these goods, a combined score was created that provided a quantitative measure of degree of MI. This massive dataset has yet to be analyzed in its entirety, and thus, has not been presented as a data chapter in this dissertation. However, a preliminary analysis presented elsewhere (Madimenos et al., 2010) does show that among 46 Shuar for whom BMD and extensive lifestyle data were available, greater market integration was negatively correlated with bone values ($P < 0.05$). This suggests that there is a trend towards compromised bone health with increased market integration and lifestyle change among the Shuar. Future analysis will include lifestyle, dietary and reproductive factors to determine the role these variables play in shaping bone density in the short- and long-term. Similar data are also essential from other natural fertility subsistence populations.

Social Support and Bone Density

Because social context and availability of kin support play a critical role in shaping lactation behaviors, there may be an association between depth of social network and female bone integrity. Put simply, if social support in food sharing and work tasks

permits women to increase lactation duration for each child, this might be reflected in her bone status. Furthermore, because of the critical health benefits of breast milk for the growing neonate, investigating the skeletal health of the maternal/child dyad will be critical for understanding intergenerational trade-offs between maternal bone density and child health outcomes. However, the longitudinal design necessary for this type of study may be a limiting factor in a remote field setting. Research such as this would link core principles outlined in Chapter IV and V—that availability of social support during lactation is expected to determine nursing frequency and intensity, and this, in turn, may have implications for skeletal calcium turnover and bone loss in the immediate and long-term life. While lactation patterns are not significantly related to long-term bone mass in Chapter IV, the effects of breastfeeding on BMD has been found, to varying extents, in the epidemiological and clinical literature (see Chapter II and IV). As the bone density component of the Shuar Health and Life History Project is an ongoing endeavor, future research may further elucidate the associations between bone parameters and lactation patterns.

SHUAR: FERTILITY IN THE CONTEXT OF LIFESTYLE CHANGE

The Indigenous Shuar of Amazonian Ecuador is an ideal population with whom to examine the negotiations between energetic resources and reproductive strategies. As described in Chapter I, the Shuar population's historically notorious resistance to colonization has allowed them to maintain many aspects of their traditional indigenous lifeways, even in the face of economic and political change. While both migration by non-Shuar into Shuar territory as well as the increasingly aggressive MI in recent years

are threatening elements of their social, political, and economic ideology, Shuar in many Upano Valley and Trans-Cutucu communities continue to retain a strong sense of indigenous identity and pride. This resistance against the encroachment of colonizing forces on their land and, in many respects, on Shuar cultural identity is partially responsible for the continued participation in subsistence-related activities found in Upano Valley communities outside of regional centers such as Sucua and Macas.

However, as discussed in Chapter I, it is important to recognize that there are varying degrees of lifestyle change and MI at both inter- and intra-community levels. In addition to increasing access to market goods and foods, fertility patterns may change as a response to relatively higher costs of raising offspring, a pronounced issue for Shuar in large towns. Furthermore, fertility patterns may be modified as access to contraception becomes increasingly available, sought after and accepted. As this dissertation is broadly an investigation of Shuar fertility, a brief discussion is warranted regarding contraception and the term “natural fertility,” which has been applied throughout the text.

“Natural Fertility” and Contraception among the Shuar

The female participants included in this dissertation studies are considered “natural fertility,” a term that identifies a population that does not to limit, regulate, or control fertility. However, in the strict sense of the word, the Shuar in this study do not fully represent a natural fertility population. During the interview process, several females reported using medicinal herbs to prevent pregnancy, while others report applying the rhythmic method which involves abstaining from sexual intercourse around ovulation. A small number of Shuar females that were interviewed, who were not

included in the data analyses, reported a history of hormonal contraception use including oral, injections, and intrauterine devices (*T de cobré*). However, these latter methods of preventing pregnancy are not widely available in rural Shuar communities, and knowledge regarding options for pregnancy prevention is poor.

Over the course of several years of fieldwork, interviews with Shuar women have occasionally prompted a few to inquire about options for preventing pregnancy; a few men have also asked about family planning options (Sugiyama, personal communication 2011). Some women, who were pregnant, expressed that it was unwanted and asked about methods to prevent future pregnancies. These women were generally older and already had approximately 4-5 offspring. Many stated that the high costs of raising children and sending them to school under current conditions were primary factors motivating their search for contraceptive options. Only in one instance did a woman, who was early in her pregnancy, inquire about abortion. At the time, she was a single mother of eight children ranging in age from 2 to 17 and her husband had been sentenced to life in prison, leaving her to fend for the entire family. A local health professional who accompanied our team told her she had no choice but to have the baby. In the subsequent field season, the woman returned for a repeat examination but she had no infant nor did she report having a nursing child during the interview process. We did not inquire further.

Statistics for the country of Ecuador show that 36.3% of total births are unintended, including both unwanted and mistimed births, with the highest percentage of unintended pregnancies reported in the Amazon Basin region (43.3%) (CEPAR, 2004). It is important to note that abortion is illegal in Ecuador so this option is generally not

advocated by health workers. These statistics are not specific to the Shuar but they reflect a lack of government-sponsored resources for sexual education and contraceptive purposes. Although the Ecuadorian government has launched plans to combat unwanted pregnancy, including the National Plan for Adolescent Pregnancy Prevention², none have explicitly remedied the need to educate the groups that comprise the most vulnerable and neglected of Ecuadorian populations: rural and indigenous peoples and those living in isolated areas of the country (Goicolea et al., 2009).

The term “natural fertility” has been applied to the participants in this study, however it is important to recognize that options for contraception are available, if not easily accessible or acceptable. In general, the Shuar women from these studies demonstrate no history of contraception use, high rates of fertility, and extended lactation periods (relative to typical Western populations). For a study that investigates energy use during reproduction, these are ideal characteristics for a participant population.

CONCLUSIONS

Three key conclusions arise from this study. First, this dissertation emphasizes the importance in applying a biocultural framework to understand aspects of reproductive biology. Pregnancy and lactation, in particular, are states that involve adjustments to the maternal physiology, and also stimulate changes in the dynamics of the mother’s social network. These adjustments are further shaped by a variety of ecological and

² The National Plan for Adolescent Pregnancy Prevention is a government-sponsored program that started in 2007 in an effort to educate Ecuadorian adolescent girls to exercise their reproductive rights by creating access to a network of social services. Critiques of the plan report that adolescents are being told contradictory messages by health professionals who believe that while information on contraception should be provided, full access to contraception should be limited because adolescents are portrayed as being unable to take responsibility or understand sexual intercourse (Goicolea et al., 2009).

environmental conditions. A thorough investigation of the responses of female physiology during reproduction, therefore, may be incomplete without considering the contribution of various facets of a woman's social environment.

Second, this dissertation highlights the utility of applying a life-course perspective to understand health outcomes, and it recognizes that there may be cumulative effects of developmental stages on health across the lifespan. Because of the complex interplay between environmental factors, the timing of developmental stages, and bone density, broad, one-size-fits-all clinical recommendations for preventing future bone loss are not ideal. These general guidelines essentialize the responses of bone to external factors, and do not consider the cumulative effect of life stages on bone status, or the environmentally-specific stressors that may promote bone loss in an individual and on a populational level. Yet, clearly, the amount of resources and time required for developing individualized or population-specific recommendations for health issues are luxuries that most researchers and clinicians do not have. While providing an alternative to this approach is beyond the scope of this dissertation, the studies presented here stress the importance of approaching understandings of health processes within this framework.

Finally, this dissertation makes a compelling argument for the need for more health and aging studies from subsistence-based populations. Cross-cultural studies of aging, particularly from non-industrialized groups, are sparse, and many chronic conditions associated with aging have not been fully investigated. Such studies have the potential for elucidating the extent to which age-associated health changes are universal, or simply an artifact of the industrial/post-industrial condition. Ultimately, these data will prove to be an invaluable contribution to informing prevention and treatment efforts.

REFERENCES CITED

- Adair LS. 1992. Postpartum nutritional status of Filipino women. *Am J Hum Biol* 4:635-646.
- Adams JS, Hewison M. 2010. Update in vitamin D. *J Clin Endocr Metab* 95:471–478.
- Affinito P, Tommaselli GA, di Carlo C, Guida F, Nappi C. 1996. Changes in bone mineral density and calcium metabolism in breastfeeding women: a one year follow-up study. *J Clin Endocr Metab* 81:2314-2318.
- Agarwal SC, Glencross B. 2011. Examining nutritional aspects of bone loss and fragility across the life cycle in bioarchaeology. In: Moffat T, Prowse T (Eds). *Human diet and nutrition in biocultural perspective*. New York: Berghahn Books.
- Agarwal SC, Stuart-Macadam P. 2003. An evolutionary and biocultural approach to understanding the effects of reproductive factors on the female skeleton. In: Agarwal SC, Stout SD (Eds). *Bone loss and osteoporosis: an anthropological perspective*. New York: Plenum Press. pp. 105-119.
- Alenfeld FE, Engelke K, Schmidt D, Brezger M, Diessel E, Felsenberg D. 2002. Diagnostic agreement of two calcaneal ultrasound devices: the Sahara bone sonometer and the Achilles. *Br J Radiol* 75: 895-902.
- Allal N, Sear R, Prentice AM, Mace R. 2004. An evolutionary model of stature, age at first birth and reproductive success in Gambian women. *P R Soc London: Series B* 271:465–470.
- Allali F, Maaroufi H, El Aichaoui S, Khazani H, Saoud, B, Benyahya B, Abouqal R, Hajjaj-Hassouni N. 2007. Influence of parity on bone mineral density and peripheral fracture risk in Moroccan postmenopausal women. *Maturitas* 57:392-398.
- Anderson JB, Switzer BR, Stewart P, Symons M. 2004. Analysis of relations between nutrient factors and bone health. In: Holick M, Dawson-Hughes B (Eds). *Nutrition and bone health*. New Jersey: Humana Press . pp. 113-128.
- Anderson JJ, Pollitzer WS. 1994. Ethnic and genetic differences in susceptibility to osteoporotic fractures. *Adv Nutr Res* 9:129-149.

- Baker PT, Hanna JM, Baker TS. 1986. The changing Samoans: behavior and health in transition. Oxford: Oxford University Press.
- Barker DJ. 1995a. Fetal origins of coronary heart disease *BMJ* 311:171-174
- 1995b. The fetal origins of adult disease. *Proc R Soc London* 262:37-43.
- Barkmann R, Laugier P, Moser U, Dencks S, Padilla F, Haiat G, Heller M, Gluer CC. 2007. A method for the estimation of femoral bone mineral density from variables of ultrasound transmission through the human femur. *Bone* 40:37-44.
- Bartl R, Frisch B. 2004. Osteoporosis: Diagnosis, prevention, therapy. Berlin, Germany: Springer-Verlag.
- Bassett DR, Schneider PL, Huntington GE. 2004. Physical activity in an old order Amish community. *Med Sci Sports Exerc* 36:79–85.
- Belevady B. 1979. Quantity and composition of breast milk in malnourished mothers. In: Hambraeus L, Sjolín S (Eds). *The mother/child dyad—nutritional aspects*. Stockholm: Almqvist and Wiksell. pp 62–68.
- Benefice E, Monroy SL, Jimenez S, Lopez R. 2006. Nutritional status of Amerindian children from the Beni River as related to environmental, maternal and dietary factors. *Public Health Nutr* 9:327-335.
- Bentley G. 1985. Hunter-gather energetics and fertility: a reassessment of the !Kung San. *Hum Ecol* 13:79-109.
- Bererhi H, Kolhoff N, Constable A, Nielsen SP. 1996. Multiparity and bone mass. *Br J Obstet Gynaecol* 103:818-821.
- Bikle D, Majumdar S, Laib A et al. 2001. The skeletal structure of IGF-I deficient mice. *J Bone Miner Res* 16:1320–1330
- Black A, Lane M. 2002. Nonhuman primate models of skeletal and reproductive aging. *Gerontology* 48:72-80.
- Black AE, Coward WA, Cole TJ, Prentice AM. 1996. Human energy expenditure in affluent societies: analysis of 574 doubly-labeled water measurements. *Eur J Clin Nutr* 50:72-92.
- Blackwell AD, Pryor III G, Pozo J, Tiwia W, Sugiyama LS. 2009. Growth and market integration in Amazonia: a comparison of growth indicators between Shuar, Shiwar, and nonindigenous school children. *Am J Hum Biol* 21:161-171.

- Blanton CA, Kretsch MJ, Baer DJ, Staples RC. 2005. Measuring physical activity energy expenditure in normal-weight, premenopausal women. *FASEB Journal* 17:A290.7.
- Bleiberg FM, Brun TA, Goihman S, Lippman D. 1981. Food intake and energy expenditure of male and female farmers from Upper-Volta. *Br J Nutr* 45:505-515.
- Bogin B. 1999. Evolutionary perspective on human growth. *Annu Rev Anthropol* 28:109-153.
- Bogin B, Loucky, J. 1997. Plasticity, political economy, and physical growth status of Guatemala Maya children living in the United States. *Am J Phys Anthropol* 102: 17–32.
- Bonnick SL, Lewis LA. 2006. Bone densitometry for technologists. 2nd edition. Human Press, Inc: New Jersey.
- Borrini-Feyerabend G, Pimbert G and Pimbert M. 2005. Sharing power: Learning-by-doing in co-management of natural resources throughout the world. Tehran, IIED and IUCN/CEESP/CMWG, Cenesta.
- Bouillon R. 1991 Growth hormone and bone. *Horm Res* 36:49–55.
- Bove RB, Valeggia CR, Ellison PT. 2002. Girl helpers and time allocation of nursing women among the Toba of Argentina. *Hum Nat* 13:457-472.
- Brickley MB, Agarwal SC. 2003. Techniques for the investigation of age-related bone loss and osteoporosis in archaeological bone. In: Agarwal SC, Stout SD (Eds). *Bone loss and osteoporosis: an anthropological perspective*. New York: Plenum Press. pp. 157-172.
- Bridges P. 1989. Changes in activities with the shift to agriculture in the southeastern US. *Curr Anthropol* 30:385-394.
- Bronte-Tinkew J, DeJong G. 2004. Children's nutrition in Jamaica: do household structure and household economic resources matter? *Soc Sci Med* 58:499-514.
- Bunker VW. 1994. The role of nutrition in osteoporosis. *Brit J Biomed Sci* 51:228-240.
- Caselli I. 2011. Ecuador President Rafael Correa loses indigenous allies. BBC news. www.bbc.co.uk/news/mobile/world-latin-america-1294423. [Accessed on 19 April 2011].

- CEPAR. ENDEMAIN. 2004. Encuesta Nacional de Demografía y Salud Materno Infantil, 2004. [National Demographic and Maternal Health Survey, 2004] Quito. www.cepar.org.ec/. [Accessed on 3 October 2010].
- Cerroni AG, Tomlinson GA, Turnquist JE, Grynepas MD. 2003. Effect of parity on bone mineral density in female rhesus macaques from Cayo Santiago. *Am J Phys Anthropol* 121:252-269.
- Charnov EL, Schaffer WM. 1973. Life history consequences of natural selection: Cole's result revisited. *Am Nat* 107:791-793
- Chen KY, Bassett DR Jr. 2005. The technology of accelerometry-based activity monitors: current and future. *Med Sci Sports Exerc* 37:S490-500.
- Cho H, Stout SD. 2003. Bone remodeling and age-associated bone loss in the past: a histomorphometric analysis of the imperial Roman skeletal population of Isola Sacra. In SC Agarwal, SD Stout (Eds). *Bone loss and osteoporosis: an anthropological perspective*. New York: Plenum Press. pp. 207-228.
- Chowdhury S, Sarkar NR, Roy SK. 2002. Impact of lactational performance on bone mineral density in marginally-nourished Bangladeshi women. *J Health Popul Nutr* 20:26-30.
- Coall D, Chisholm J. 2010. Reproductive development and parental investment during pregnancy: moderating influence of mother's early environment. *Am J Hum Biol* 22:143-153.
- CODENPE. 2011. Nacionalidad Shuar. http://www.codenpe.gov.ec/index.php?option=com_content&view=article&id=281&Itemid=632&lang=es. [Accessed on May 5, 2011].
- Cohen MN, Armelagos GJ. 1984. *Paleopathology at the origins of agriculture*. Orlando, Florida: Academic Press.
- Committee of Revision. 1997. *Drug information for the health care professional*. Rockville, MD: United States Pharmacopeial Convention, Inc.
- Cooper C, Eriksson JG, Forsen T, Osmond C et al. 2001. Maternal height, childhood growth and risk of hip fracture in later life: a longitudinal study. *Osteoporos Int* 12:623-629.
- Cooper C, Fall C, Egger P, Hobbs R, Eastell R, Barker D. 1997. Growth in infancy and bone mass in later life. *Ann Rheum Dis* 56:17-21.

- Cooper C, Javaid MK, Taylor P, Walker-Bone K, Dennison E, Arden N. 2002. The fetal origins of osteoporotic fracture. *Calcif Tissue Int* 70:391-394.
- Cooper C, Walker-Bone K, Arden N, et al. 2000. Novel insights into the pathogenesis of osteoporosis: the role of intrauterine programming. *Rheumatology* 39:1312-1315.
- Charnov EL. 1993. *Life history invariants*. Oxford University Press.
- Charnov EL, Schaffer WM. 1973. Life history consequences of natural selection: Cole's result revisited. *Am Nat* 107:791-793.
- Chen KY, Bassett DR Jr. 2005. The technology of accelerometry-based activity monitors: current and future. *Med Sci Sports Exerc* 37:S490-500.
- Chin-Hong PV, McGarvey ST. 1996. Lifestyle incongruity and adult blood pressure in Western Samoa Source. *Psychosom Med* 58:130 -137.
- Cho H, Stout SD. 2003. Bone remodeling and age-associated bone loss in the past: a histomorphometric analysis of the imperial Roman skeletal population of Isola Sacra. In: Agarwal SC, Stout SD (Eds). *Bone loss and osteoporosis: an anthropological perspective*. New York: Plenum Press. pp. 207-228.
- Cordain L, Eaton SB, Sebastian A, Mann N, Lindeberg S, Watkins BA, O'Keefe JH, Brand-Miller J. 2005. Origins and evolution of the Western diet: health implications for the 21st century. *Am J Clin Nutr* 81:341-354.
- Coward WA. 1998. Contributions of the doubly labeled water method to studies of energy balance in the Third World. *Am J Clin Nutr* 68:962S-969S.
- Cowin SC. 2001. *Bone mechanics handbook*. Boca Raton: CRC press.
- Crouter SE, Clowers KG, Bassett Jr. DR. 2006. A novel method for using accelerometer data to predict energy expenditure. *J Appl Physiol* 100:1324-1331.
- Cumming R, Nevitt M. 1997. Calcium for prevention of osteoporotic fractures in postmenopausal women. *J Bone Min Resear* 12:1321-1329.
- Dargeant-Molina P, Favier F, Grandjean H. 1996. Fall-related factors and risk of hip fracture: the EPIDOS prospective study. *Lancet* 348:145-149.
- Dawson-Hughes B. 2004. Calcium and vitamin D for bone health in adults. In: Holick M, Dawson-Hughes B (Eds). *Nutrition and bone health*. New Jersey: Humana Press. pp. 197-210.

- Dennison E, Hindmarsh P, Fall C, Kellingray S, et al. 1999. Profiles of endogenous circulating cortisol and bone mineral density in healthy elderly men. *J Clin Endocrinol Metab* 84:3058-3063.
- Descola P. 1994. *In the society of nature: A native ecology in Amazonia*. Cambridge: Cambridge University Press.
1996. *The spears of twilight: life and death in the Amazon jungle*. London: Harper Collins Publishers.
- Dewey KG. 1997. Energy and protein requirements during lactation. *Annu Rev Nutr* 17:19-36.
- Drake TA, Schadt E, Hannani K, Kabo JM, et al. 2001. Genetic loci determining bone density in mice with diet-induced atherosclerosis. *Physiol Genomics* 5:205-215.
- Dressler WW. 1991. Social class, skin color, and arterial blood pressure in two societies. *Ethn Dis* 1: 60-77.
- Drinkwater BL, Chestnut CH. 1991. Bone density changes during pregnancy and lactation in active women: a longitudinal study. *Bone Miner* 14:153-160.
- Dufour DL, Piperata BA. 2008. Energy expenditure among farmers in developing countries: what do we know? *Am J Hum Biol* 20:249-258.
- Dufour DL, Reina JC, Spurr GB. 2002. Energy intake and expenditure of free-living, lactating Colombian women in an urban setting. *Eur J Clin Nutr* 56:1-9.
- Durnin JVGA. 1990. Low energy expenditures in free-living populations. *Eur J Clin Nutri* 44:S95-S102.
- Durnin JVGA, Wormesley J. 1974. Body fat assessed from total body density and its estimation from skinfold thickness on 481 men and women aged 16 to 72 years. *Brit J Nutr* 32:77-97.
- Early JD, Peters JF. 1990. *The population dynamics of the Mucajai Yanomama*. San Diego, California: Academic Press.
- Eastell R. 2005. Role of oestrogen in the regulation of bone turnover at the menarche. *J Endocrinol* 185:223-234.
- Eaton SB, Nelson DA. 1991. Calcium in evolutionary perspective. *Am J Clin Nutr* 54: 281S-287S.

- Eaton SB, Pike MC, Short RV, Lee NC, Trussell J, Hatcher RA, Wood JW et al. 1994. Women's reproductive cancers in evolutionary context. *Q Rev Biol* 69:353-367.
- El-Hajj Fuleihan G, Deeb M. 1999. Hypovitaminosis D in a sunny country. *N Engl J Med* 340:1840–1841.
- El-Sonbaty MR, Abdul-Ghaffar NU. 1996. Vitamin D deficiency in veiled Kuwaiti women. *Eur J Clin Nutr* 50:315–318.
- Ellison PT. 1982. Skeletal growth, fatness and menarcheal age: a comparison of two hypotheses. *Hum Biol* 54:269–281.
1994. Advances in human reproductive ecology. *Annu Rev Anthropol* 23:255-275.
2001. *On fertile ground. A natural history of human reproduction*. Cambridge: Harvard University Press.
2003. Energetics and reproductive effort. *Am J Hum Biol* 15: 342–351.
2008. Energetics, reproductive ecology, and human evolution. *Paleo-Anthropology*:172-200.
- Ellison PT, Jasienska G. 2007. Constraint, pathology, and adaptation: How can we tell them apart? *Am J Hum Biol* 19: 622–630.
- Ellison PT, Lipson SF, O'Rourke MT, Bentley GR, Harrigan AM, Panter-Brick C, Vitzthum VJ. 1993. Population variation in ovarian function. *Lancet* 342:433–434.
- Ellison PT, Peacock NR, Lager C. 1989. Ecology and ovarian function among Lese women of the Ituri Forest, Zaire. *Am J Phys Anthropol* 78:519–526.
- Ensom MH, Liu PY, Stephenson MD. 2002. Effect of pregnancy on bone mineral density in healthy women. *Obstet Gynecol Surv* 57:99-111.
- Eriksen EF, Kassem M, Brixen K. 1993. Growth hormone and insulin-like growth factors as anabolic therapies for osteoporosis. *Horm Res* 40:95–98
- Eriksen EF, Kassem M, Langdahl B. 1996 Growth hormone, insulin-like growth factors and bone remodelling. *Eur J Clin Invest* 26:525–534
- Evans JA, Tavakoli MB. 1990. Ultrasonic attenuation and velocity in bone. *Phys Med Biol* 35: 1387.

- Fall C, Hindmarsh P, Dennison E, Kellingray S, Barker D, Cooper C. 1998. Programming of growth hormone secretion and bone mineral density in elderly men: a hypothesis. *J Clin Endocrinol Metab* 83:135-139.
- FAO/UNU/WHO. 2004. Human energy requirements. Report of a joint FAO/WHO/UNU expert consultation. Rome: United Nations University.
- Feldblum PJ, Zhang J, Rich LE, Fortney JA, Talmage RV. 1992. Lactation history and bone mineral density among perimenopausal women. *Epidemiol* 3:327-531.
- Ferrari S, Rizzoli R, Slosman D, Bonjour JP. 1998. Familial resemblance for bone mineral mass is expressed before puberty. *J Clin Endocrinol Metab* 83:358–361.
- Fleming-Moran M, Santos R, Coimbra C. 1991. Blood pressure levels of the Suruí Indians of the Brazilian Amazon: Group- and sex-specific effects resulting from body composition, health status, and age. *Hum Biol* 63:835-861.
- Food and Agriculture Organization of the United Nations (FAO). 2007. Agricultural frontier and demographic transition in the upper Morona-Santiago watersheds, Ecuador.
- Food and Nutrition Board (FNB)/Institute of Medicine (IOM). 2002. Dietary reference intakes of energy, carbohydrates, fiber, fat, protein, and amino acids (Macronutrients). National Academies Press.
- Forsmo S, Schei B, Langhammer A, Forsén L. 2001. How do reproductive and lifestyle factors influence bone density in distal and ultradistal radius of early postmenopausal women? The Nord-Trøndelag Health Survey, Norway. *Osteoporos Int* 12:222-229.
- Forwood M. 2001. Physiology. In: Khan K, McKay H, Kannus P, Bailey D, Wark J, Bennell K (Eds). *Physical activity and bone health*. United Kingdom: Human Kinetics. pp. 11-21.
- Frost HL. 1960. Presence of microscopic cracks in vivo in bone. *Henry Ford Hospital Bulletin* 8:25-35.
1986. *Intermediary organization of the skeleton*. Vols. 1 and 2. Boca Raton, Florida: CRC Press.
- Frost HM. 2003. On changing views about age-related bone loss. In Agarwal SC, Stout SD. *Bone loss and osteoporosis: An anthropological perspective*. Springer, pp. 19-32.

- Freedson PS, Melanson E, Sirard J. 1998. Calibration of the computer science and applications, Inc. accelerometer. *Med Sci Sport Exer* 30:777-781.
- Friedlander JS, Howells WW, Rhoads JG (Eds). 1987. *The Solomon Islands Project: a long-term study of health, human biology, and culture change*. Oxford: Oxford University Press.
- Gadgil M, Bossert W. 1970. Life history consequences of natural selection. *Am Nat* 104:1-24.
- Gallagher JC. 2007. Effect of early menopause on bone mineral density and fractures. *Menopause* 14:567-571.
- Galloway, A. 1988. Long term effects of reproductive history on bone mineral content in women. Doctoral Dissertation. The University of Arizona.
- Galloway A. 1997. The cost of reproduction and the evolution of postmenopausal osteoporosis. In: Morbeck ME, Galloway A, Zihlman A (Eds). *The evolving female: a life history perspective*. Princeton, NJ: Princeton University Press. pp. 132-146.
- Gannage-Yared MH, Chemali R, Yaacoub N, Halaby G. 2000. Hypovitaminosis D in a sunny country: relation to lifestyle and bone markers. *J Bone Miner Res* 15:1856–1862.
- Garn SM. 1987. The secular trend in size and maturational timing and its implications for nutritional assessment. *J Nutr* 117:817–823.
- Garner P, Smith T, Baea M, Lai D, Heywood P. 1994. Maternal nutritional depletion in a rural area of Papua-New-Guinea. *Trop Geogr Med* 46:169–171.
- Gerdhem P, Dencker M, Ringsberg K, Akesson K. 2008. Accelerometer-measured daily physical activity among octogenarians: results and associations to other indices of physical performance and bone density. *Eur J Appl Physiol* 102:173-180.
- Gerdhem P, Obrant KJ. 2004. Bone mineral density in old age: the influence of age at menarche and menopause. *J Bone Miner Metabol* 22 372–375.
- Ghannam NN, Hammami, MM Bakheet SM, Khan BA. 1999. Bone mineral density of the spine and femur in healthy Saudi females: relation to vitamin-D status, pregnancy and lactation. *Cacif Tissue Int* 65:23-28.
- Gibson M, Mace R. 2005. Helpful grandmothers in rural Ethiopia: a study of the effect of kin on child survival and growth. *Evol Hum Behav* 26:469–482.

- Gigante DP, Horta BL, Lima RC, Barros FC, Victora CG. 2006. Early life factors are determinants of female height at the 19 years in a population-based birth cohort (Pelotas, Brazil). *J Nutr* 136:473–478.
- Gil A, Ochoa G, Omran R. 1981. In: Omran AR, Standley CC, Ochoa, G et al. (Eds). *Family formation patterns and health: further studies*. Geneva, World Health Organization. pp. 257-260.
- Gluckman P, Hanson M. 2004. *The fetal matrix: evolution, development and disease* Cambridge University Press.
- Godfrey K, Walker-Bone K, Robinson S, Taylor P, Shore S, et al. 2001. Neonatal bone mass: influence of parental birthweight, maternal smoking, body composition and activity during pregnancy. *J Bone Miner Res* 16:1694-1703.
- Godoy R. 2000. *Indians, rain forests, and markets: theory, methods, and analysis*. New York: Columbia University Press.
- Godoy R, Cardenas M. 2000. Markets and the health of indigenous people: A methodological contribution. *Hum Organ* 59:117-124.
- Goicolea I, Wulff M, Öhman A, San Sebastian M. 2009. Risk factors for pregnancy among adolescent girls in Ecuador's Amazon basin: a case- control study. *Rev Panam Salud Publica*. 26:221–228.
- Goldberg GR, Prentice AM, Coward WA, Davies HL, Murgatroyd PR, Sawyer MB, Ashford J, Black AE. 1991. Longitudinal assessment of the components of energy balance in well-nourished lactating women. *Am J Clin Nutr* 54:788–798.
- González-Reimers E, Velasco-Vázquez J, Arnay-De-La-Rosa M, Santolaria-Fernández, F, Gómez-Rodríguez M, Machado-Calvo M. 2002. Double-energy X-ray absorptiometry in the diagnosis of osteopenia in ancient skeletal remains. *Am J Phys Anthropol* 118: 134–145.
- Gray S. 1994. Correlates of dietary intake of lactating women in south Turkana. *Am J Hum Biol* 6:369–383.
- Grimes JP, Wimalawansa SJ. 2003. Breastfeeding and postmenopausal osteoporosis. *Curr Womens Health Rep* 3:193-198.
- Gross D, Eiten G, Flowers N, Leoi F, Ritter M, Werner D. 1979. Ecology and acculturation among native peoples of central Brazil. *Science* 206:1043-1050.

- Guillermo-Tuazon MA, Barba CVC, van Raaij JMA, Hautvast J. 1992. Energy intake, energy expenditure, and body composition of poor rural Philippine women throughout the first 6 months of lactation. *Am J Clin Nutr* 56:874–880.
- Gur A, Nas K, Cevik R, Aysegul JS, Ataoglu S, Karakoc M. 2003. Influence of number of pregnancies on bone mineral density in postmenopausal women of different age groups. *J Bone Miner Metab* 21:234-241.
- Gurven M, Kaplan H. 2008. Beyond the grandmother hypothesis: evolutionary models of longevity. In: Sokolovsky J (Ed). *Cultural context of aging: worldwide perspectives*. 3rd edition. Greenwood Press. pp. 53-60.
- Gurven MC, Kaplan H, Winking J, Rodriguez DE, Vasunilashorn S, et al. 2009. Inflammation and infection do not promote arterial aging and cardiovascular disease risk factors among lean horticulturalists. *PLoS ONE* 4(8):e6590.
- Guyton AC, Hall JE. 2011. Pregnancy and lactation. In: Guyton AC, Hall JE (Eds). *Textbook of medical physiology*. 12th edition. Philadelphia, PA: WB Saunders Company. pp. 1027-1041.
- Hagen EH, Barrett HC, Price ME. 2006. Do human parents face a quantity-quality tradeoff?: Evidence from a Shuar community. *Am J Phys Anthropol* 130: 405–418.
- Handa R, Kalla AA, Maalouf G. 2008. Osteoporosis in developing countries. *Best Pract Res Clin Rh* 22:693-708.
- Harner MJ. 1984. *The Jívaro, people of the sacred waterfalls*. Berkeley: University of California Press.
- Hawkes K, O'Connell J, Blurton Jones NG. 1997. Hadza women's time allocation, offspring provisioning, and the evolution of long postmenopausal life spans. *Curr Anthropol* 38:551 -577.
- Hawkes K, O'Connell JF, Blurton Jones NG, Alvarez H, Charnov EL. 1998. Grandmothering, menopause, and the evolution of human life histories. *Proc Nat Acad Sci* 95 1336-1339.
- Hayes M, Chustek M, Heshka S, Wang Z, Pietrobelli A, Heymsfield SB. 2005. Low physical activity levels of modern *Homo sapiens* among free-ranging mammals. *Int J Obes Relat Metab Disord* 29:151–156.
- Hayslip CC, Klein TA, Wray HL, Duncan WE. 1989. The effects of lactation on bone mineral content in healthy postpartum women. *Obstet Gynecol* 73:588-592.

- Heil DP. 2006. Predicting activity energy expenditure using the Actical activity monitor. *Res Q Exerc Sport* 77:64-80.
- Hendelman D, Miller K, Baggett C, Debold E, Freedson P. 2000. Validity of accelerometry for the assessment of moderate intensity physical activity in the field. *Med Sci Sport Exer* 32: S442-S449.
- Henderson PH 3rd, Sowers M, Kutzko KE, Jannausch ML. 2000. Bone mineral density in grand multiparous women with extended lactation. *Am J Obstet Gynecol* 182:1371-1377.
- Hendricks JW. 1993. *To drink of death: the narrative of a Shuar warrior in southeastern Ecuador*. Tuscon: University of Arizona Press.
- Henrich J. 1997. Market integration, agricultural change, and sustainability among the Machiguenga Indians of the Peruvian Amazon. *Hum Ecol* 25:319-351.
- Henry CJK. 2005. Basal metabolic rate studies in humans: measurement and development of new equations. *Public Health Nutr* 8:1133-1152.
- Hernandez-Avila M, Stampfer MJ, Ravnikaar VA, Willet WC, Schiff I, Francis M, Longscope C, Mckinlay SM, Longcope C. 1993. Caffeine and other predictors of bone density among pre-and perimenopausal women. *Epidemiol* 4:128-134.
- Hill KR, Hawkes K, Hurtado M, Kaplan H. 1984. Seasonal variance in the diet of Ache hunter-gatherers in Eastern Paraguay. *Hum Ecol* 12:101-135.
- Hill KR, Hurtado AM. 1996. The evolution of premature reproductive senescence and menopause in human females: an evaluation of the grandmother hypothesis. In: Betzig L (Ed). *Human nature: a critical reader*. New York: Oxford University Press. pp. 313-350.
2009. Cooperative breeding in South American hunter-gatherers. *Proc R Soc B* 276:3863-3870.
- Hill K, Kaplan H. 1999. Life history traits in humans: theory and empirical studies. *Annu Rev Anthropol* 28:397-438.
- Hillier TA, Rizzo JH, Pedula KL, Stone KL, Cauley JA, Bauer DC, Cummings SR. 2003. Nulliparity and fracture risk in older women: the study of osteoporotic fractures. *J Bone Miner Res* 18:893-899.
- Hinkley HJ, Drysdale IP, Walters NJ, Bird D. 2004. Normative data for ultrasound measurement of the calcaneus within different female ethnic groups. *Brit J Radiol* 77:740-744.

- Holick MF. 2008. Vitamin D: a D-lightful health perspective. *Nutr Rev* 66:S182–194.
- Holmberg-Marttila D, Sievanen H, Tuimala R. 1999. Changes in bone mineral density during pregnancy and postpartum: prospective data on five women. *Osteoporos Int* 10:41–46.
- Hoos MB, Plasqui G, Gerver WJM, Westerterp KR. 2003. Physical activity level measured by doubly labeled water and accelerometry in children. *Eur J Appl Physiol* 89:624–626.
- Houghton LA, Vieth R. 2006. The case against ergocalciferol (vitamin D₂) as a vitamin supplement. *Am J Clin Nutr* 84(4):694–697.
- Howe JC, Rumpler WV, Seale J. 1993. Energy expenditure by indirect calorimetry in premenopausal women: variation within one menstrual cycle. *J Nutr Biochem* 4:268–273.
- Hrdy SB. 1999. *Mother nature: a history of mothers, infants, and natural selection*. New York: Pantheon Books.
2000. The optimal number of fathers: evolution, demography and history in the shaping of female mate preferences. *Ann NY Acad Sci* 907:75–96.
2005. Comes the child before the man: how cooperative breeding and prolonged postweaning dependence shaped human potentials. In: Hewlett B, Lamb M (Eds). *Hunter-gatherer childhoods: evolutionary, developmental, and cultural perspectives*. New York, NY: Aldine. Pp. 65–91.
- Hreschchyshyn MM, Hopkins A, Zulstra S. 1988. Associations of parity, breastfeeding, and birth control pills with lumbar spine and femoral neck bone densities. *Am J Obstet Gynecol* 159: 318–322.
- Hurtado AM, Hill K, Kaplan H, Hurtado I. 1992. Trade-offs between female food acquisition and child care among Hiwi and Ache foragers. *Human Nat* 3:185–216.
- Huss-Ashmore R, Schall J, Hediger M (Eds). 1992. *Health and lifestyle change*. Philadelphia: University of Pennsylvania.
- Institute of Medicine. 1997. *Standing committee on the scientific evaluation of dietary reference intakes. Vitamin D. Dietary reference intakes: calcium, phosphorus, magnesium, vitamin D, and fluoride*. Washington, DC: National Academy Press. pp. 250–287.

- International Osteoporosis Foundation (IOF). 2010. Facts and statistics about osteoporosis and its impact. www.iofbonehealth.org/facts-and-statistics.html. [Accessed on 1 February 2011].
- Isaksson OG, Lindahl A, Nilsson A, Isgaard J. 1987. Mechanism of the stimulatory effect of growth hormone on longitudinal bone growth. *Endocr Rev* 8:426–438.
- Isgaard J, Moller C, Isaksson OG, Nilsson A, Mathews LS, Norstedt G. 1988. Regulation of insulin-like growth factor messenger ribonucleic acid in rat growth plate by growth hormone. *Endocrinol* 122:1515–1520.
- Ito M, Yamada M, Hayashi K, Ohki M, Uetani M, Nakamura T. 1995. Relation of early menarche to high bone mineral density. *Calcif Tissue Int* 57:11-14.
- Ivey PK. 2000. Cooperative reproduction in Ituri Forest hunter-gatherers: who cares for Efe infants? *Curr Anthropol* 41:856-866.
- Jaffe RB, Dell'Acqua S. 1985. The endocrine physiology of pregnancy and the peripartal period, Raven Press, New York.
- James GD, Brown DE. 1997. The biological stress response and lifestyle: catecholamines and blood pressure. *Ann Rev Anthropol* 26:313-335.
- James WPT, Schofield EC. 1990. Human energy requirements. New York: Food and Agriculture Organization of the United Nations/Oxford University Press.
- Jasienska G. 2001. Why energy expenditure causes reproductive suppression in women. An evolutionary and bioenergetic perspective. In: Ellison PT (Ed). *Reproductive ecology and human evolution*. New York: Aldine de Gruyter. p 59–85.
2003. Energy metabolism and the evolution of reproductive suppression in the human female. *Acta Biotheor* 51:1-18.
2004. Energetic factors and seasonal changes in ovarian function in women from rural Poland. *Am J Hum Biol* 16:563-580.
2009. Reproduction and lifespan: trade-offs, overall energy budgets, intergenerational costs, and costs neglected by research. *Am J Phys Anthropol* 21:524-532.
- Jasienska G, Ellison PT. 1998. Physical work causes suppression of ovarian function in women. *Proc Biol Sci* 265(1408):1847-1851.

- Jasienska G, Thune I, Ellison PT. 2006. Fatness at birth predicts adult susceptibility to ovarian suppression: an empirical test of the Predictive Adaptive Response hypothesis. *Proc Natl Acad Sci-Biol* 103: 12759–12762.
- Jelliffe DB, Maddocks I. 1964. Notes on ecologic malnutrition in the New Guinea highlands. *Clin Ped* 3:432-438.
- Johnell O, Nilsson B. 1984. Lifestyle and bone mineral mass in perimenopausal women. *Calcif Tissue Int* 36:354-356.
- Jones RE. 1989. Breastfeeding and postpartum amenorrhea in Indonesia. *J Biosoc Sci* 21:83-100.
- Jouanny P, Guillemin F, Kuntz C, Jeandel C, Pourel J. 1995. Environmental and genetic factors affecting bone mass. *Arthritis Rheum* 38:61–67.
- Kalkwarf HJ, Specker BL. 1995. Bone mineral loss during lactation and recovery after weaning. *Obstet Gynecol* 86:26-32.
- Kaplan H, Hill K, Lancaster J, Hurtado AM. 2000. A theory of human life history evolution: diet, intelligence, and longevity. *Evol Anthropol* 9:156–185.
- Karasik D, Arensburg B, Tillier A-M, Pavlovsky O. 1998. Skeletal age assessment of fossil hominids. *J Archaeol Sci* 25:689–696
- Karlsson MK, Ahlborg HG, Karlsson C. 2005. Female reproductive history and the skeleton—a review. *BJOG Inter J Obstet Gynaecol* 112: 851–856.
- Karsten R. 1935. The headhunters of western Amazonas: the life and culture of the Jibaro Indians of eastern Ecuador and Peru. Helsinki: Societas Scientiarum Fennica, *Commentationes Humanarum Littararum*, Vol. 7(1).
- Kashiwazaki H. 1999. Heart rate monitoring as a field method for estimating energy expenditure as evaluated by the doubly labeled water method. *J Nutr Vitaminol* 45:79-94.
- Kashiwazaki H, Dehima Y, Orias-Rivera J, Coward WA. 1995. Energy expenditure determined by the doubly labeled water method in Bolivian Aymara living in a high altitude agropastoral community. *Am J Clin Nutr* 62:901-910.
- Kashiwazaki H, Uenishi K, Kobayashi T, Rivera JO, Coward WA, Wright A. 2009. Year-round high physical activity levels in agropastoralists of Bolivian Andes: results from repeated measurements of DLW method in peak and slack seasons of agricultural activities. *Am J Hum Biol* 21:337-345.

- Kelley MA. 1980. Disease and environment: A comparative analysis of three early American Indian skeletal collections. Doctoral Dissertation. Case Western Reserve University.
- Kelly PJ, Morrison NA, Sambrook PN, Nguyen TV, Eisman JA. 1995. Genetic influences on bone turnover, bone density and fracture. *Eur J Endocrinol* 133:265-271.
- Kemper H, Twisk J, van Mechelen W, Post G, Roos J, Lips P. 2000. A fifteen-year longitudinal study in young adults on the relation of physical activity and fitness with the development of the bone mass: The Amsterdam growth and health longitudinal study. *Bone* 27:847-853.
- Kent GN, Price RI, Gutteridge DH, Smith M, Allen JR, Bhagat CI, et al. 1990. Human lactation: forearm trabecular bone loss, increased bone turnover, and renal conservation of calcium and inorganic phosphate with recovery of bone mass following weaning. *J Bone Miner Res* 5:361-369.
- Kent GN, Price RI, Gutteridge DH, Smith M, Allen JR, Rosman KJ, Smith M, et al. 1993. Effect of pregnancy and lactation on maternal bone mass and calcium metabolism. *Osteoporos Int* 3 (Suppl 1):44-47.
- Keramat A, Patwardhan B, Larijani B, et al. 2008. The assessment of osteoporosis risk factors in Iranian women compared with Indian women. *BMC Musculoskeletal Disord* 9:28.
- Khan K, McKay H, Kannus P, Bailey D, Wark J, Bennell K. 2001. Soft tissue determinants of bone mineral density. In: Khan K, McKay H, Kannus P, Bailey D, Wark J, Bennell K (Eds). *Physical activity and bone health*. United Kingdom: Human Kinetics pp. 71-86.
- Kirchengast S, Winkler EM. 1996. Differential fertility and body build in !Kung San and Kavango females from northern Namibia. *J Biosoc Sci* 28:193-210.
- Kline J., Stein Z, Susser M. 1989. *Conception to Birth: Epidemiology of prenatal development* New York, Oxford University Press.
- Kolthoff N, Eiken P, Kristensen B, Nielsen SP. 1998. Bone mineral changes during pregnancy and lactation: a longitudinal cohort study. *Clin Sci* 94:405-412.
- Konner M, Worthman C. 1980. Nursing frequency, gonadal function and birth spacing among !Kung hunter-gathers. *Science* 207:788.

- Kovacs CS. 2001. Calcium and bone metabolism in pregnancy and lactation. *J Clin Endocrinol Metab* 86:2344-2348.
- Kovacs CS, Kronenberg HM. 1997. Maternal-fetal calcium and bone metabolism during pregnancy, puerperium and lactation. *Endocr Rev* 18:832-872.
- Krall EA, Dawson-Hughes B. 1993. Heritability and life-style determinants of bone mineral density. *J Bone Miner Res* 8:1-6.
- Kramer KL. 2005. Children's help and the pace of reproduction: cooperative breeding in humans. *Evol Anthropol* 14:224-237.
- Kramer KL, Greaves RD. 2011. Juvenile subsistence effort, activity levels and growth patterns: middle childhood among Pumé foragers. *Hum Nat* (in press).
- Kramer MS. 1987. Determinants of low birth weight: methodological assessment and meta-analysis. *Bulletin of World Health Organization* 65:663-737.
- Kritz-Silverstein D, Barrett-Connor E. 1993. Early menopause, number of reproductive years, and bone mineral density in postmenopausal women. *Am J Public Health* 83: 983-988.
- Kröger H, Kotaniemi A, Kröger L, Alhava E. 1993. Development of bone mass and bone density of the spine and femoral neck—a prospective study of 65 children and adolescents. *Bone Miner* 23:171-82.
- Kung AWC, Huang QY. 2007. Genetic and environmental determinants of osteoporosis. *J Musculoskelet Neuronal Interact* 7:26-32.
- Kurz KM, Habicht JP, Rasmussen KM, Schwager SJ. 1993. Effects of maternal nutritional status and maternal energy supplementation on length of postpartum amenorrhea among Guatemalan women. *Am J Clin Nutr* 58:636-640.
- Kuzawa CW. 1998. Adipose tissue in human infancy and childhood: An evolutionary perspective. *Yrbk Phys Anthropol* 41:177-209.
- Lamke B, Brundin J, Moberg P. 1977. Changes of bone mineral content during pregnancy and lactation. *Acta Obstet Gynecol Scand* 56:217-219.
- Laskey MA, Prentice A. 1997. Effect of pregnancy on recovery of lactational bone loss. *Lancet* 349:1518-1519.
- Laskey MA, Prentice A, Hanratty LA, et al. 1998. Bone changes after 3 months of lactation: influence of calcium intake, breast-milk output, and vitamin D-receptor genotype. *Am J Clin Nutr*. 67:685-692.

- Lassek WD, Gaulin SJC. 2006. Changes in body fat distribution in relation to parity in American women. *Am J Phys Anthropol* 131:295-302.
- Lawrence M, Whitehead RG. 1988. Physical activity and total energy expenditure of child-bearing Gambian village women. *Eur J Clin Nutr* 42:145-160.
- Lazenby R. 1997. Bone loss, traditional diet, and cold adaptation in Arctic populations. *Am J Hum Biol* 9:329-341.
- Leatherman T. 1994. Health implications of changing agrarian economies in the southern Andes. *Hum Organ* 53:371-380.
- Lee M, Nahhas RW, Choh AC, Demerath EW, Duren DL, Chumlea WC, et al. 2010. Longitudinal changes in calcaneal quantitative ultrasound measures during childhood. *Osteoporos Int* doi:10.1007/s00198-010-1458-0.
- Lee RB. 1979. *The !Kung San: men, women and work in a foraging society*. Cambridge: Cambridge University Press.
- Lees CJ, Jerome CP, Register TC, Carlson CS. 1998. Changes in bone mass and bone biomarkers of cynomolgus monkeys during pregnancy and lactation. *J Clin Endocrinol Metab* 83:4298-4302.
- Leidy L. 1996. Lifespan approach. *Am J Hum Biol* 8: 699-702.
- Lenora J, Lekamwasam S, Karlsson MK. 2009. Effects of multiparity and prolonged breast-feeding on maternal bone mineral density. *BMC Women's Health* 9:19.
- Leonard WR. 2003. Measuring human energy expenditure: what have we learned from the flex-heart rate method? *Am J Hum Biol* 15:479-489.
2008. Lifestyle, diet, and disease: Comparative perspectives on the determinants of chronic health risks. In: Stearns SC, Koella JC (Eds). *Evolution in health and disease*. 2nd Edition. Oxford University Press.
- Leonard WR, DeWalt K, Uquillas J, DeWalt B. 1993. Ecological correlates of dietary consumption and nutritional status in highland and coastal Ecuador. *Ecology of Food and Nutrition* 31:67-85.
- Leonard WR, Galloway VA, Ivakine E. 1997. Underestimation of daily energy expenditure with the factorial method: implications for anthropological research. *Am J Phys Anthropol* 103:443-454.
- Leonard WR, Katzmarzyk PT, Stephen MA, Ross AGP. 1995. Comparison of the heart rate-monitoring and factorial methods: assessment of energy-expenditure in highland and coastal Ecuadorians. *Am J Clin Nutr* 61:1146-1152.

- Leonard WR, Robertson ML. 1992. Nutritional requirements and human evolution: A bioenergetics model. *Am J Hum Biol* 4:179-195.
- Levis S, Altman R. 1998. Bone densitometry: clinical considerations. *Arthritis Rheum* 41:577-587.
- Liebert MA, Snodgrass JJ, Blackwell AD, Madimenos FC, Sugiyama LS. 2010. The implications of varying degrees of market integration on blood pressure, glucose, cholesterol, and triglyceride levels in an indigenous lowland Ecuadorian population. *Am J Hum Biol* 22:260.
- Lindegärde F, Widen I, Gebb M, Ahren B. 2004. Traditional versus agricultural lifestyle among Shuar women of the Ecuadorian Amazon: Effects of leptin levels. *Metabolism* 53: 1355-1358.
- Lindsay R, Bush TL, Grady D, Speroff L, Lobo RA. 1996. Therapeutic controversy: Estrogen replacement in menopause. *J Clin Endocr Metab* 81: 3829-38.
- Lipson SF, Ellison PT. 1996. Comparison of salivary steroid profiles in naturally occurring conception and non-conception cycles. *Hum Reprod* 11:2090–2096.
- Lissner L, Bengtsson C, Hansson T. 1991. Bone mineral content in relation to lactation history and pre- and post-menopausal women. *Calcif Tissue Int* 48:319-325.
- Little MA, Leslie PW, Campbell KL. 1992. Energy reserves and parity of nomadic and settled Turkana women. *Am J Hum Biol* 4:729–738.
- Liu W, Xu CL, Zhu ZQ, et al. 2006. Characteristics of calcaneus quantitative ultrasound normative data in Chinese mainland men and women. *Osteoporos Int* 17:1216-1224.
- Lohman TG, Roche AF, Martorell R (Eds). 1988. Anthropometric standardization: reference manual. Champaign, IL: Human Kinetics Books.
- Looker A. 2003. Interaction of science, consumer practices and policy: calcium and bone health as a case study. *J Nutr* 133:1987S-1991S.
- Lopez JM, Gonzalez G, Reyes V, Campino C, Diaz S. 1996. Bone turnover and density in healthy women during breastfeeding and after weaning. *Osteoporos Int* 6:153-159.
- Lu F. 2007. Integration in the market among indigenous peoples: a cross-cultural perspective from the Ecuadorian Amazon. *Curr Anthropol* 48:593-602.

- Mace R. 1996. Biased parental investment and reproductive success in Gabbra pastoralists. *Behavioural Ecology and Sociobiology* 38:75–81.
- Mader E. 1999. Metamorfosis del poder: persona, mito, y visión en la sociedad Shuar y Achuar. Quito: Ediciones Abya-Yala.
- Mader E, Gippelhauser R. 2000. Power and kinship in Shuar and Achuar. In Schweitzer P (Ed). *Dividends of kinship, meaning and uses of social relatedness*. London: Routledge. pp. 61-91.
- Madimenos FC, Liebert MA, Snodgrass JJ, Blackwell AD, and Sugiyama LS. 2010. A profile of bone health in an Ecuadorian forager-horticulturalist population. *Am J Phys Anthropol* 141(S50):160.
- Marcus R. 1991. Skeletal ageing; Understanding the functional and structural basis of osteoporosis. *Trends Endocrin Met* March/April:53-58.
- Marlowe F. 2003. A critical period for provisioning by Hadza men: implications for pair bonding. *Evol Hum Behav* 24:217-229.
- Matkovic V, Jelic T, Wardlaw GM, Ilich JZ, Goel PK, Wright JK, et al. 1994. Timing of peak bone mass in Caucasian females and its implication for the prevention of osteoporosis. Inference from a cross-sectional model. *J Clin Invest* 93:799-808.
- Matsumoto I, Kosha S, Noguchi S, et al. 1995. Changes of bone mineral density in pregnant and postpartum women. *J Obstet Gynaecol* 21:419-425
- Mays S. 2000. Age-dependent cortical bone loss in women from 18th and early 19th century London. *Am J Phys Anthropol* 112:349–361.
- Mazess RB. 1978. Bone mineral in Vilcabamba, Ecuador. *Am J Roentgenol* 130:671-674.
- McDade T. 2001. Lifestyle incongruity, social integration, and immune function in Samoan adolescents. *Soc Sci Med* 53:1351-1362.
2002. Status incongruity in Samoan youth: A biocultural analysis of culture change, stress, and immune function. *Med Anthropol Q* 16:123-150.
- McSweeney K, Jokisch B. 2006. Report on results of the diagnóstico de la situación de salud y de los servicios de salud de las nacionalidades Shuar y Achuar. FICSH-FIPSE-FINAE. UNICEF-Ecuador.

- Meczekalski B, Podfigurna-Stopa A, Genazzani AR. 2010. Hypoestrogenism in young women and its influence on bone mass density. *Gynecol Endocrinol* 26:652-657.
- Meehan CL. 2009. Maternal time allocation in two cooperative childrearing societies. *Hum Nature* 20:375-393.
- Meijer GAL, Westerterp KR, Saris WHM, Ten HF. 1992. Sleeping metabolic rate in relation to body composition and the menstrual cycle. *Am J Clin Nutr* 55:637–640.
- Melton LJ III, Bryant SC, Wahner HW, O’Fallon WM, Malkasian GD, Judd HL and Riggs BL. 1993. Influence of breastfeeding and other reproductive factors on bone mass later in life. *Osteoporos Int* 3:76-83.
- Merz WA, Schenk RK. 1970. Quantitative structural analysis of human cancellous bone. *Acta anatomica (Basel)* 75:54.
- Miller JE, Rodriguez G, Pebley AR. 1994. Lactation, seasonality, and mother's postpartum weight change in Bangladesh: an analysis of maternal depletion. *Am J Hum Biol* 6:511-524.
- Minority Rights Group International. 2008. World Directory of Minorities and Indigenous Peoples - Ecuador: Shuar. www.unhcr.org/refworld/docid/49749d2cc.html [accessed 8 March 2011].
- Monteiro CA, Mondini L, Medeiros de Souza AL, Popkin BM. 1995. The nutrition transition in Brazil. *Eur J Clin Nutr* 49:105–113.
- Mosekilde, Li. 2000. Age-related changes in bone mass, structure, and strength—effects of loading. *Zeitschrift für Rheumatologie* 59 (Suppl. 1): I/1–I/9.
- Mosekilde Li, Mosekilde Le, Danielsen CC. 1987. Biomechanical competence of vertebral trabecular bone in relation to ash density and age in normal individuals. *Bone* 8:79-85.
- Munk-Jensen N, Nielsen SP, Obel EB, Eriksen PB. 1988. Reversal of postmenopausal vertebral bone loss by oestrogen and progestogen: a double blind placebo controlled study. *Br Med J* 296:1150.
- Murphy S, Khaw KT, May H, Compston E. 1994. Parity and bone mineral density in middle-aged women. *Osteoporos Int* 4:162-166.
- Naganathan V, Macgregor A, Snieder H, Nguyen T, Spector T, Sambrook P. 2002. Gender differences in the genetic factors responsible for variation in bone density and ultrasound. *J Bone Miner Res* 17:725-733.

- National Osteoporosis Foundation (NOF). 2010. Clinician's Guide to Prevention and Treatment of Osteoporosis. Washington, DC: National Osteoporosis Foundation.
- Nayak S, Olkin I, Liu H, Grabe M, Gould MK, Allen IE, Owens DK, Bravata DM. 2006. Meta-analysis: accuracy of quantitative ultrasound for identifying patients with osteoporosis. *Ann Intern Med* 144:832-841.
- Nguyen TV, Jones G, Sambrook PN, White CP, Kelly PJ, Eisman JA. 1995. Effects of estrogen exposure and reproductive factors on bone mineral density and osteoporotic fractures. *J Clin Endocrinol Metab* 80:2709-2714.
- Njeh, CF, Cheng XG, Elliot JM, Meunier PJ. 1999. Bone, bone diseases and bone quality. In: Njeh CF, Hans D, Fuerst T, Gluer CC, Genant HK (Eds). *Quantitative ultrasound: assessment of osteoporosis and bone status*. London: Martin Dunitz Ltd. pp. 1-20.
- Ohlsson C, Bengtsson BA, Isaksson OG, Andreassen TT, Slootweg MC. 1998. Growth hormone and bone. *Endocr Rev* 19:55-79.
- Ohta H, Sugimoto I, Masuda A, Komukai S, Suda Y, Makita K, Takamatsu K, Horiguchi F, Nozawa S. 1996. Decreased bone mineral density associated with early menopause progresses for at least ten years: cross-sectional comparisons between early and normal menopausal women. *Bone*. 18:227-231.
- Ordoñez F. 2011. Indigenous Will Stand with Protest to Freedom their Three Leaders. www.ecuadortimes.net/2011/02/03/indigenous-will-protest-to-freedom-their-three-leaders. [Accessed on 3 February 2011].
- Oreffo RO, Lashbrooke B, Rooch HI, et al. 2003. Maternal protein deficiency affects mesenchymal stem cell activity in the developing offspring. *Bone* 33:100-107.
- Ozdemir F, Demirbag D, Rodoplu M. 2005. Reproductive factors affecting the bone mineral density in postmenopausal women. *The Tohoku Journal of Experimental Medicine* 205: 277-285.
- Pacifici R. 2007. Estrogen deficiency, T cells and bone loss. *Cell Immunol* 252:68-80.
- Pagezy H. 1984. Seasonal hunger as experienced by the Oto and Twa women of Ntomba village in the equatorial forest (Lake Tumba, Zaire). *Ecol Food Nutr* 15:13-27.
- Panther-Brick C. 1993. Seasonality of energy expenditure during pregnancy and lactation for rural Nepali women. *Am J Clin Nutr* 57:620-628.

- Pavan L, Casiglia E, Carvalho-Braga LM, Winnicki M, Puato M, Pauletto P, Pessina AC. 1999. Effects of a traditional lifestyle on the cardiovascular risk profile: the Amondava population of the Brazilian Amazon. *J Hypertens* 17:749-756.
- Pearce KL. 2006. Breastfeeding and bone density change. Doctoral Dissertation. University of Massachusetts, Amherst.
- Pearce MS, Birrell FN, Francis RM, Rawlings DJ, Tuck SP, Parker L. 2005. Lifecourse study of bone health at age 49-51 years: the Newcastle thousand families cohort study. *J Epidemiol Commun H* 59:475-480.
- Peng TC, Kusy RP, Garner SC, Hirsch PF, De Blanco MC. 1987. Influence of lactation and pregnancy and lactation on mechanical properties and mineral content of the rat femur. *J Bone Miner Res* 2:249-257.
- Phillips DI, Barker DJ, Fall CH, Seckl JR, Whorwood CB, et al. 1998. Elevated plasma cortisol concentrations: a link between low birthweight and the insulin resistance syndrome? *J Clin Endocrinol Metab* 83:757-760.
- Phinney SD, Stern JS, Burke KE, Tang AB, Miller G, Holman RT. 1994. Human subcutaneous adipose tissue shows site-specific differences in fatty acid composition. *Am J Clin Nutr* 60:725-729.
- Pike I. 2001. The evolutionary and ecological context of human pregnancy. In: Ellison P (Ed). *Reproductive ecology and human evolution*. Walter de Gruyter: New York. pp. 39-58.
- Piperata BA. 2009. Variation in maternal strategies during lactation: the role of the biosocial context. *Am J Hum Biol* 21:817-827.
- Piperata BA, Dufour DL. 2007. Diet, energy expenditure, and body composition of lactating Ribeirinha women in the Brazilian Amazon. *Am J Hum Biol* 19:722-734.
- Plasqui G, Westerterp KR. 2007. Physical activity assessment with accelerometers: an evaluation against doubly labeled water. *Obes* 15:2371-2379.
- Pluskiewicz W. 1998. Bone status assessed by quantitative ultrasound in healthy postmenopausal Polish women: normative data. *Clin Rheumatol* 17:40-43.
- Popat VB, Calis KA, Vanderhoof VH, Cizza G, Reynolds JC, Sebring N, Troendle JF, Nelson LM. 2009. Bone mineral density in estrogen-deficient young women. *J Clin Endocrinol Metab* 94:2277-2283.

- Popivanov P, Boianov M. 2001. Osteoporosis in pregnancy and lactation. *Akush Ginekol (Sofia)* 41:40-43.
- Popkin BM. 2004. The nutrition transition: An overview of world patterns of change. *Nutr Rev* 62:140-143.
2006. Technology, transport, globalization and the nutrition transition food policy. *Food Policy* 31:554-569.
- Popkin BM, Guilkey DK, Akin JS, Adair LS, et al. 1993. Nutrition, lactation, and birth spacing in Filipino women. *Demography* 30:333-352.
- Pouillès J, Trémollières F, Bonneu M, Ribot C. 1994. Influence of early age at menopause on vertebral bone mass. *J Bone Miner Res* 9:311-315.
- Prentice A, Jarjou L, Cole T, Stirling D, Dibba B, Fairweather-Tate S. 1995. Calcium requirements of lactating Gambian mothers: effects of a calcium supplement on breast-milk calcium concentration, maternal bone mineral content, and urinary calcium excretion. *Am J Clin Nutr* 62:58-67.
- Prentice AM, Spaaij CJK, Goldberg GR, Poppitt SD, van Raaij JMA, Totton M, Swann D, Black AE. 1996. Energy requirements of pregnant and lactating women. *Eur J Clin Nutr* 50 Suppl 1:82-111.
- Proctor DN, Melton III LJ, Khosla S, Crowson CS, O'Connor MK, and Riggs BL. 2000. Relative influence of physical activity, muscle mass and strength on bone density. *Osteoporosis Int* 11:944-952.
- Reiches MW, Ellison PT, Lipson SF, Sharrock KC, Gardiner E, Duncan LG. 2009. Pooled energy budget and human life history. *Am J Hum Biol* 21:421-429.
- Rhee Y, Lee J, Jung JY, Lee JE, Park SY, Kim YM, Lee S, Choi HS, Kim SH, Lim SK. 2009. Modifications of T-scores by quantitative ultrasonography for the diagnosis of osteoporosis in Koreans. *J Korean Med Sci* 24:232-236.
- Roberts C, Manchester K. 2005. *The archaeology of disease*. New York: Cornell University Press.
- da Rocha FA, Ribiero AR. 2003. Low incidence of hip fractures in an equatorial area. *Osteoporosis Int* 14:496-499.

- Rode A, Shephard RJ 1994a. Growth and fitness of Canadian Inuit: secular trends, 1970–1990. *Am J Hum Biol* 6:525–541.
- 1994b. Physiological consequences of acculturation: a 20-year study of fitness in an Inuit community. *Eur J Appl Physiol* 69:516–524.
- Rodrigues MLC, Da Costa THM. 2001. Association of the maternal experience and changes in adiposity measured by BMI, waist:hip ratio and percentage of body fat in urban Brazilian women. *Br J Nutr* 85:107–114.
- Ross P, Huang C, Davis J, Imose K, Yates J, Vogel J, Wasnich R. 1995. Predicting vertebral deformity using bone densitometry at various skeletal sites and calcaneus ultrasound. *Bone* 16:325–332.
- Roy DK, O'Neill TW, Finn JD, Lunt M, Silman AJ, Felsenberg D, Ambrecht G, Banzer D, Benevolenskaya LI, et al. 2003. Determinants of incident vertebral fracture in men and women: results from the European Prospective Osteoporosis Study (EPOS). *Osteoporos Int* 14 19–26.
- Rubenstein S. 2001. Colonialism, the Shuar Federation, and the Ecuadorian state. *Environ Plan D* 19:263–293.
2002. Alejandro Tsakimp: a Shuar healer in the margins of history. University of Nebraska Press.
- Sampson H. 2002. Alcohol and other factors affecting osteoporosis risk in women. *Alcohol Res Health* 264:292–296.
- Santos R, Coimbra C. 1991. Socioeconomic transition and physical growth of Tupí-Mondê Amerindian children in the Aipuana Park, Brazilian Amazon. *Hum Biol* 63:795–819.
- Schnatz PF, Barker KG, Marakovits KA, O'Sullivan DM. 2010. Effects of age at first pregnancy and breast-feeding on the development of postmenopausal osteoporosis. *Menopause*. 17:1161–1166.
- Scholl TO, Hediger ML. 1993. A review of the epidemiology of nutrition and adolescent pregnancy: Maternal growth during pregnancy and its effect on the fetus. *J Am Coll Nutr* 12:101–107.
- Sear R, Allal N, Mace R, McGregor IA. 2004. Height, marriage and reproductive success in a Gambian population. *Res Econ An* 23:203–224.
- Sellen DW, Smay DB. 2001. Relationship between subsistence and age at weaning in “preindustrial” societies. *Hum Nat-Int Bios* 12:47–87.

- Seymour-Smith C. 1988. Shiuiar: identidad étnica y cambio en el río corrientes. Quito-Lima: Abya Yala-CAAP.
- Shephard RJ, Rode A. 1996. The health consequences of ‘modernization’: evidence from circumpolar peoples. Cambridge: Cambridge University Press.
- Silman AJ. 2003. Risk factors for Colles’ fracture in men and women: results from the European Prospective Osteoporosis Study. *Osteoporos Int* 14:213–218.
- Singh J, Prentice AM, Diaz E, et al. 1989. Energy expenditure of Gambian women during peak agricultural activity measured by the doubly-labeled water method. *Br J Nutr* 62:315-329.
- Sioka C, Fotopoulos A, Georgiou A, Xourgia X, Papadopoulos A, Kalef-Ezra JA. 2010. Age at menarche, age at menopause and duration of fertility as risk factors for osteoporosis. *Climacteric* 13:63-71.
- Sirén AH. 2007. Population growth and land use intensification in a subsistence-based indigenous community in the Amazon. *Hum Ecol* 35:669-680.
- Skarić-Jurić T, Zajc M, Narancić NS, Barbalić M, Salihović MP, Lauc LB. 2006. Calcaneous ultrasonographic assessment of bone mineral density in the Roma minority population of Croatia--preliminary report. *Coll Antropol.* 30:761-765.
- Sloboda DM, Hickey M, Hart R. 2010. Reproduction in females: the role of the early environment. *Hum Reprod Update* 1-19.
- Slootweg MC. 1993. Growth hormone and bone. *Horm Metab Res* 25:335–343
- Snodgrass JJ. 2011. Human energetics. In Stinson S, Bogin B, O’Rourke D, editors. *Human biology: and evolutionary and biocultural approach*. 2nd edition. New York: Wiley-Blackwell. In press.
- Snodgrass JJ, Leonard WR, Sorensen MV, Tarskaia LA, Alekseev VP, Krivoschapkin VG. 2006. The emergence of obesity among indigenous Siberians. *J Physiol Anthropol* 25: 75-84.
- Snodgrass JJ, Leonard WR, Tarskaia LA, Schoeller DA. 2006. Total energy expenditure in the Yakut (Sakha) of Siberia as measured by the doubly labeled water method. *Am J Clin Nutr* 84:798-806.
- Snodgrass JJ, Sorensen MV, Tarskaia LA, Leonard WR. 2007. Adaptive dimensions of health research among indigenous Siberians. *Am J Hum Biol* 19: 165-180.

- Speakman JR. 1997. Doubly labeled water: theory and practice. London: Chapman and Hall.
- Sperling S, Beyene Y. 1997. A pound of biology and a pinch of culture or a pinch of biology and a pound of culture? The necessity of integrating biology and culture in reproductive studies. In: Hager L (Ed). Women in human evolution. New York: Routledge. Pp. 137-152.
- Spurr GB, Dufour DL, Reina JC. 1996. Energy expenditure of urban Colombian women: a comparison of patterns and total daily expenditure by the heart rate and factorial methods. *Am J Clin Nutr* 63:870-878.
- Sorensen MV, Snodgrass JJ, Leonard WR, Tarskaia LA, Ivanov KI, Krivoschapkin VG, Spitsyn VA. 2005. Health consequences of postsocialist transition: dietary and lifestyle determinants of plasma lipids in Yakutia. *Am J Hum Biol* 17:576-592.
- Sowers MF. 1996. Pregnancy and lactation as risk factors for subsequent bone loss and osteoporosis. *J Bone Miner Res* 11:1052-1060.
2001. Premenopausal reproductive and hormonal characteristics and the risk for osteoporosis. In: Marcus R, Feldman D, Kelsey J (Eds). Osteoporosis. 2nd edition. San Diego: Academic Press. pp. 721-739.
- Sowers MF, Clark MK, Hollis B, Wallace RB, Jannausch M. 1992. Radial bone mineral density in pre- and perimenopausal women: a prospective study of rates and risk factors for loss. *J Bone Miner Res* 7:647-657.
- Sowers MF, Corton G, Shapiro B, Jannausch ML, Crutchfield M, Smith ML, Randolph JF, Hollis B. 1993. Changes in bone density with lactation. *J Amer Med Assoc* 269:3130-3135.
- Sowers MF, Eyre D, Hollis BW, Randolph JF, Shapiro B, Jannausch ML, Crutchfield M. 1995. Biochemical markers of bone turnover in lactating and nonlactating postpartum women. *J Clin Endocrinol Metab* 80:2210-2216.
- Sowers MF, Wallace RB, Lemke JH. 1985. Nutritional and non-nutritional correlates of bone mass among women at maximal bone mineralization. *Prev Med* 14:585-596.
- Stearns SC. 1976. Life history tactics: a review of the ideas. *Q Rev Biol* 51: 3-47.
1992. The evolution of life histories. Oxford: Oxford University Press.
- Stini WA. 1995. Osteoporosis in biocultural perspective. *Annu Rev Anthropol* 24:397-421.

- Stirling M. 1938 Historical and ethnographic materials of the Jivaro Indians. Smithsonian Institution Bureau of American Ethnology Bulletin 117(2).
- Stone MR, Rowlands AV, Eston RG. 2009. Relationships between accelerometer-assessed physical activity and health in children: impact of the activity-intensity classification method. *J Sports Sci and Med* 8:136-143.
- Strassman BI. 1997. Polygyny as a risk factor for child mortality among the Dogon. *Curr Anthropol* 38:688-695.
- Strauss J, Thomas D. 1988. Nutrient intakes and income. In: Chenery H, Srinivasan A (Eds). *Handbook of development economics, 1893-1908*. Amsterdam: North Holland.
- Streeten EA, Ryan KA, McBride DJ, Pollin TI, Shuldiner AR, Mitchell BD. 2005. The relationship between parity and bone mineral density in women characterized by a homogenous lifestyle and high parity. *J Clin Endocr Metab* 90:4536-4541.
- Streeter M, Stout SD. 2003. Histomorphometry of the subadult rib: age-associated changes in bone mass and the creation of peak bone mass. In: Agarwal SC, Stout SD (Eds). *Bone loss and osteoporosis: an anthropological perspective*. New York: Plenum Press. pp. 91-101.
- Stuart-Macadam P. 1995. Biocultural perspectives on breastfeeding. In: Stuart-Macadam P, Dettwyler KA (Eds). *Breastfeeding: biocultural perspectives*. New York: Walter de Gruyter, Inc. pp. 1-37.
- Sugiyama LS. 2004. Illness, injury, and disability among Shiwiar forager-horticulturalists: implications of health-risk buffering for the evolution of human life history. *Am J Phys Anthropol* 123:371-389.
- Sugiyama LS, Chacon R. 2000. Effects of illness and injury on foraging among the Yora and Shiwiar: pathology risk as adaptive problem. In: Cronk L, Chagnon NA, Irons W (Eds). *Human behavior and adaptation: an anthropological perspective*. New York: Aldine. pp. 371-395.
2005. Juvenile responses to household ecology among the Yora of Peruvian Amazonia. In: Hewlett B, Lamb M (Eds). *Hunter-gatherer childhoods: evolutionary, developmental, and cultural perspectives*. New York: Transaction. pp. 237-261.
- Swartz AM, Strath SJ, Bassett DR, O'Brien WL, King GA, Ainsworth BE. 2000. Estimation of energy expenditure using CSA accelerometers at hip and wrist sites. *Med Sci Sport Exer* 32:S450-S456.

- Takeda N, Miyake M, Kita S, Tomomitsu T, Fukunaga M. 1996. Sex and age patterns of quantitative ultrasound densitometry of the calcaneus in normal Japanese subjects. *Calcif Tissue Int* 59:84–88.
- Tataranni PA, Harper IT, Snitker S, et al. 2003. Body weight gain in free-living Pima Indians: effect of energy intake vs expenditure. *International Journal of Obesity* 27:1578–83.
- Taylor A. 1981. God-wealth: the Achuar and the missions. In: Whitten Jr NE (Ed). *Cultural transformations and ethnicity in modern Ecuador*. Urbana: University of Illinois Press.
- Theintz G, Buchs B, Rizzoli R, Slosman D, Clavien H, Sizonenko PC, et al. 1992. Longitudinal monitoring of bone mass accumulation in healthy adolescents: evidence for a marked reduction after 16 years of age at the levels of lumbar spine and femoral neck in female subjects. *J Clin Endocrinol Metab* 75:1060–1065.
- Thomas PA. 2007. Racial and ethnic differences in osteoporosis. *J Am Acad Orthop Surg* 15 Suppl 1:S26-30.
- Tracer DP. 1991. Fertility-related changes in maternal body composition among the Au of Papua New Guinea. *Am J Phys Anthropol* 85:393–405.
- Trivers RL. 1972. Parental investment and sexual selection. In: Campbell B (Ed). *Sexual selection and the descent of man*. Chicago, IL: Aldine. pp. 136–179.
1974. Parent-offspring conflict. *Am Zool* 14:249–264.
- Trovas G, Tsekoura M, Galanos A, Dionyssiotis Y, Dontas I, Lyritis G, Papioanou N. 2009. Quantitative ultrasound of the calcaneus in Greek women: Normative data are different from the manufacturer's normal range. *J Clin Densito* 12:353–359.
- Ulijaszek SJ. 1995. *Human energetics in biological anthropology*. Cambridge: Cambridge University Press.
2001. Work and climate in traditional subsistence economies. *J Physiol Anthropol* 20:105–110.
- Valeggia C, Ellison PT. 2001. Lactation, energetics, and postpartum amenorrhea. In: Ellison PT (Ed). *Reproductive ecology and human evolution*. Aldine de Gruyter, New York. pp 85–106.

- Van der Weyden L, Wei L, Luo J, et al. 2006. Functional knockout of the matrilin-3 gene causes premature chondrocyte maturation to hypertrophy and increases bone mineral density and osteoarthritis. *Am J Pathol* 169:515-527.
- Varennna M, Binelli L, Zucchi F, Ghiringhelli D, Gallazzi M, Sinigaglia L. 1999. Prevalence of osteoporosis by educational level in a cohort of postmenopausal women. *Osteoporos Int* 9:236-241.
- Venners SA, Liu X, PerryMJ, Korrick SA, Li ZP, Yang F, Yang JH, Lasley BL, Xu XP, Wang XB. 2006. Urinary estrogen and progesterone metabolite concentrations in menstrual cycles of fertile women with non-conception, early pregnancy loss or clinical pregnancy. *Hum Reprod* 21:2272-2280.
- Vico L, Prallet B, Chappard D, Alexandre C. 1991. Lumbar bone density and reproductive years. *J Bone Miner Res* S162.
- Vieth R. 2003. Effects of vitamin-D on bone and natural selection of skin color. In: Agarwal SC, Stout SD (Eds). *Bone loss and osteoporosis: an anthropological perspective*. New York: Plenum Press. pp. 139-154.
- Voland E. 1998. Evolutionary ecology of human reproduction. *Annu Rev Anthropol* 27:347-374.
- von Braun J, Kennedy E (Eds). 1994. *Agricultural commercialization, economic development, and nutrition*. Baltimore: Johns Hopkins University Press.
- Waldron L, Nowotarski M, Freimek M, Henry JP, Post N, Witten C. 1982. Cross-cultural variation in blood pressure. *Soc Sci Med* 16:419-430.
- Wardlaw GM, Pike AM. 1986. The effect of lactation on peak adult shaft and ultra-distal forearm bone mass in women. *Am J Clin Nutr* 44:283-286.
- Weaver DS. 1998. Osteoporosis in the bioarchaeology of women. In: Grauer L, Stuart-Macadam P (Eds). *Sex and gender in paleopathological perspective*. Cambridge: Cambridge University Press. pp. 27-46.
- Weinstein RS, Hutson MS. 1987. Decreased trabecular width and increased trabecular spacing contribute to bone loss with aging. *Bone* 8:137-142.
- Wells JCK. 2005. Evolution of the human profile of fatness. *J Hum Ecol* 13:17-22.
- Westerterp KR. 2009. Assessment of physical activity: a critical appraisal. *Eur J Appl Physiol* 105:823-828.

- Williams GC, Nesse RM. 1991. The dawn of Darwinian medicine. *Q Rev Biol* 66:1–22.
- Wood JW. 1994. Dynamics of human reproduction: biology, biometry, demography. Hawthorn, New York: Aldine de Gruyt.
- Woolf AD, Pfleger B. 2005. Burden of osteoporosis and fractures in developing countries. *Current Osteoporosis Reports* 3:85-91.
- World Health Organization [WHO]. 2003. Prevention and Management of Osteoporosis. Geneva: World Health Organization. Report of a WHO scientific group. WHO Technical Report Series 921.
- World Health Organization/Food and Agriculture Organization [WHO/FAO]. 2003. Diet, nutrition and the prevention of chronic diseases: Overcoming impediments to prevention and control. WHO Technical Report Series 916.
- Wosje K, Specker BL. 2000. Role of calcium in bone health during childhood. *Nutr Revs* 58:253-268.
- Wuster C. 1993 Growth hormone and bone metabolism. *Acta Endocrinol* 128:14–18.
- Yamauchi T, Umezaki M, Ohtsuka R. 2001. Physical activity and subsistence pattern of the Huli, a Papua New Guinea highland population. *Am J Phys Anthropol* 114:258-268.
- Yao M, McCrory MA, Ma G, Li Y, Dolnikowski GG, Roberts SB. 2002. Energy requirements of urban Chinese adults with manual or sedentary occupations, determined using the doubly labeled water method. *Eur J Clin Nutr* 56:575-584.
- Yu DW, Shephard GH. 1998. Is beauty in the eye of the beholder? *Nature* 396:321-322.
- Zanker C, Hind K. 2007. The effect of energy balance on endocrine function and bone health. *Med Sport Sci* 51:81-101.
- Zapatos-Rios G, Urgilés C, Suárez E. 2009. Mammal hunting by the Shuar of the Ecuadorian Amazon: is it sustainable? *Oryx* 43:375-385.