Factors Regulating Bone Maturity and Strength in Poultry[1]

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ABSTRACT Adolescent meat-type poultry and cage layers exhibit a high incidence of bone problems that include bone weakness, deformity, breakage, and infection and osteoporosis-related mortalities. These problems include economic and welfare issues. To improve bone quality in poultry, it is essential to understand the physiological basis of bone maturity and strength in poultry. A complex array of factors that include structural, architectural, compositional, physiological, and nutritional factors interactively determine bone quality and strength. Bone is approximately 70% mineral, 20% organic, and 10% water. Collagen is the major organic matrix that confers tensile strength to the bone, whereas hydroxyapatite provides compressional strength. In recent years, the roles of different collagen crosslinks have been shown to be important in the increase of bone mechanical strength. Similarly, age-related glyco-oxidative modifications of collagen have been shown to increase the stiffness of collagen. These posttranslational modifications of matrix can affect bone quality as it would be affected by the changes in the mineralization process. Our studies show that the growth in the tibia continued until 25 wk of age, which correlated with the increase in the content of hydroxylysylpridinoline (HP) and lysylpyridinoline (LP), the collagen crosslinks. The tibia from 5-wk-old chicks were strong but brittle because of low collagen crosslinks and high mineral content. Bone maturity may relate to its crosslink content. Compared to crosslink content, bone density and ash content showed moderate increases during growth. The bones from younger turkeys were more susceptible to corticosteroid-induced stunting of growth, which also resulted in decreased bone strength. This review discusses how different factors can compromise bone strength by reducing growth, altering shape, affecting mineralization, and affecting collagen crosslinking.

(*Key words*: poultry, bone strength, bone maturity, collagen crosslinks)

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INTRODUCTION

There is a need for better understanding of bone strength in poultry because bone breakage and associated infections contribute to mortality, low productivity, and carcass condemnations. Market age poultry often suffer from lameness and bone deformities, which can cause bone breakage during catching and transportation and which create problems during processing (Gregory and Wilkins, 1992; Julian, 1998; Knowles and Wilkins, 1998). Bone weakness and other bone problems also constitute significant animal welfare issues because of lameness and mortalities stemming from leg weakness and osteoporosis in laying hens (Aziz-Abdul, 1998). Bone fragility and porosity also are correlated with the incidence of bone fragments in deboned meat products, and with discoloration of meat adjacent to bone due to leaching of blood, the product may be less appealing to consumers. Overall, the economic cost associated with bone problems in poultry can add up to several hundred million dollars a year. The purpose of this review is to outline factors that contribute to or compromise bone strength and quality in poultry.

PHYSICAL AND BIOCHEMICAL BASIS OF BONE STRENGTH

Bone is a dynamic tissue influenced by physiological, nutritional, and physical factors such as mechanical stress and physical activities. Like mammalian bones, the avian long bones are made up of compact cortical bone, which is the outer shell surrounding the cancellous or trabecular bone and marrow space. The trabecular bones are organized as a lattice and provide larger surface areas and show high turnover rates (Albright, 1987). The architectural organization of the lattice structure can also be important for the strength of a bone. Based on the structural organization, bone is either lamellar or woven. The former

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Abbreviation Key: AGE = advanced glycation end products; HP = hydroxylysylpyridinoline; LP = lysylpyridinoline; TD = tibial dyschondroplasia.

is a mature form of bone made up of cortical and cancellous bones. The lamellar bones are slow depositing bones with well organized, aggregated arrangements of collagen, whereas the woven bones deposit rapidly during intramembranous ossification or fracture healing and are distinguished by their loosely packed and randomly organized collagen fibers, lower mineral density, and high water content (Turek, 1984; Gorski, 1998). Flat bones such as skull bones, vertebral bones, and the metaphyseal end of long bones have large amounts of cancellous bone. As compared to cortical bone, cancellous bone is less calcified, plays a larger role in metabolic function, and undergoes continuous remodeling (Seifert and Watkins, 1997). In female birds coinciding with the maturation of ovarian follicles, a type of bone develops in the endosteal surface of long bones called "medullary bone" that is essentially woven bone with a high rate of remodeling providing Ca to meet the demand of eggshell formation (Dacke et al., 1993). Although the medullary bone lacks substantial intrinsic strength, it is likely to influence the mechanical strength of cortical bone (Knott et al., 1995; Flemming et al., 1998). Medullary bone is low in collagen but high in mineral, proteoglycans, and carbohydrates as compared to cortical or cancellous bone (Dacke et al., 1993; Rath et al., 1999).

Collagen is the major constituent of the organic matrix, contributing to the tensile strength of bone and providing oriented support to the mineral matrix (Riggs et al., 1993). The arrangements of collagen fibers in bone with respect to bone axes can also influence bone strength (Albright, 1987; Martin and Boardman, 1993). Approximately 80 to 90% of the organic matrix is comprised of collagen, which is a triple helical fibrous protein that forms the primary scaffolding of skeletal tissues, and provides oriented support for the mineralization process. Collagen, being the major organic constituent of bone, also is likely to affect its biomechanical strength. Impairment of collagen synthesis or assembly is bound to affect the biomechanical strength of tissues. Besides calcification, the collagen molecules undergo the process of fibrillogenesis and several other posttranslational modifications, such as hydroxylations and intermolecular crosslinking, that increase their tensile strength and help withstand physical stress (Eyre, 1996; Knott and Bailey, 1998). The most well-known, mature form of collagen crosslinks are pyridinium crosslinks, which are hydroxylysyl pyridinoline (HP) and lysyl pyridinoline (LP). Pyridinium crosslinks have been shown to be important in bone strength (Eyre, 1996). The latter form of crosslinks is specific to calcified tissues. Recently, Knott et al., (1995) have identified the presence of a "pyrrole" crosslink in avian bone collagen, which apparently contributes to the biomechanical strength of bones. Osteoporotic avian bones have a reduced level of pyrrolic crosslinks (Knott et al., 1995). Advanced glycation end products (AGE) that form by nonenzymatic crosslinking of glucose to proteins have been shown to be present in many connective tissues (Sell and Monnier, 1989) and are also present in bone. Pentosidine is the only identified glycation end product shown to be present in bone. However, the significance of pentosidine in bone is not understood, although it could likely influence the properties of collagen by imparting stiffness to the bone during the aging process (Bank et al., 1998; Knott and Bailey, 1998). Besides collagen, the other 10 to 15% of the organic matrix are proteoglycans, lipids, and noncollagenous proteins such as osteocalcin, osteonectin, and osteopontins. The noncollagenous proteins contribute to a variety of functions of bone such as matrix stabilization, calcification, and other metabolic regulatory activities (Termine and Gehron-Robey, 1996).

The mineral matrix is predominantly Ca and P in the form of hydroxyapatite, which constitutes ~60 to 70% of the bone weight and provides stiffness and compressional strength to the bone. The collagen molecules are arranged so that they overlap to produce repeating periodic segments of hole zones in collagen fibers where hydroxyapatite crystals deposit and calcification occurs (Turek, 1984). The calcification process is complex and is not well understood. However, hydroxyapatite crystals impart shear strength to the bone by binding to adjacent collagen fibers. Bone density is the mass of material per volume of bone, which includes both organic and mineral mass. Because the inorganic matrix is the major component of the extracellular matrix, bone mineral density is considered to reflect the status of bone health. Low bone density has been considered to be a risk factor for fracture. Bone also contains water, ~10 to 15% of total weight, which can be consequential to its viscoelastic properties. Underlying the structural complexities of bone are the osteoblasts, osteocytes, and osteoclasts. These cellular elements are responsible for matrix synthesis, mineralization, and resorption and are key determinants of bone chemistry, geometry, and strength.

BONE MATURITY

Bone maturity can be defined as the completion of basic structural development and mineralization and the attainment of optimal mechanical strength. The maturation process involves many different complex molecular and biochemical changes in bone that lead to the attainment of optimal physical and biomechanical properties. These changes can occur in collagen fiber diameter, crosslink content, and deposition of more lamellar type bones. Collagen crosslink content, for example, has been suggested as an indicator of bone maturity in mammals (Boskey et al., 1999) and can also be a useful marker of bone maturity in poultry. In rapidly growing poultry, bone development and maturity fail to keep pace with overall growth, thereby generating excess physical load and predisposing bone to deformity and fragility. However, the basis for such claims has not been verified. Loading experiments conducted in chicks and turkey poults by Patterson et al. (1986) did not show any increased incidence of leg deformity in these birds as compared to controls.

PARAMETERS OF BONE STRENGTH

Bone strength is the toughness or ability to endure stress; therefore it is related to ultimate load or stress at

which the bone will break. The load at break is the sum of all forces and moments applied to the bone (Nigg and Grimstone, 1994) and is the breaking strength of bone. It is actually the load or stress that is normalized to represent the force applied per square area at the time of break (Huff et al., 1980: Crenshaw et al., 1981; Turner and Burr, 1993; Einhorn, 1996). The term has sometimes been used interchangeably in reporting the quantity of loads used to break the bone, particularly when using the bones from small animals (Turner and Burr, 1993). However, the latter use of the term breaking strength can create confusion, particularly when comparing large versus small bones (Crenshaw et al., 1981). Therefore, the normalized value based on the area of impact is preferred when comparing bones of different sizes and from different ages of birds (Patterson et al., 1986). Even such calculations are not without error because of the structural irregularities of bone. For example, variations can stem from calculations with the assumption of bone as a solid cylinder as used by Huff et al. (1980) versus an ellipse with both internal and external diameters as used by Crenshaw et al. (1981). However, a comparison of both methods using both right and left tibia from the same birds under different experimental conditions showed comparable trends of changes in breaking strength (data not shown), suggesting that either assumption would suffice in the determination of bone strength. In three-point bending tests, the diameter of the bone at the point of impact is used for calculating the area. Under constant increments of loads, the bone undergoes elastic deformation until the point is reached where it is no longer resilient. The load required to reach this point is called yield strength or stress at yield (Turner and Burr, 1993). The stress at yield reflects the rigidity of bone as a whole, whereas the slope of the linear region of the stress vs. strain-curve is called Young's modulus or elastic modulus and reflects the intrinsic stiffness or rigidity and material properties of bone. High modulus may indicate bone to be more rigid and less ductile, whereas low modulus could mean the bone to be more ductile and less mineralized as may be expected of a rachitic bone (Turner and Burr, 1993). Strain denotes percentage of deformity; less mineralized bone has higher values. Compressional strength has also been used to measure bone strength, particularly in metaphyseal areas, but this method has not been used extensively in poultry. Because bone mineralization provides compressional strength to bone, the bone ash content or bone mineral densities have been used as the indices of bone strength.

Bone mineral density can be measured using noninvasive methods such as X-ray and ultrasound devices (Frost and Roland, 1991). However, the Archimedes' principle is conveniently used to measure the density of small pieces of bones following postmortem harvest. Bone density is considered to reflect bone mineral content. However, many recent studies have shown that the mineral density can be contingent upon the organic matrix chemistry of bones (Knott and Bailey, 1998). This relationship is due to modifications of matrix molecules, such as intermolecular crosslinks of collagen, interaction of proteoglycans and other noncollagenous proteins with collagen, and other glyco-oxidative changes in collagen and proteoglycans that can alter mineralization and biomechanical properties. In human studies, loss of collagen crosslinks from bone has been linked to osteoporosis (Oxlund et al., 1995; Eyre, 1996).

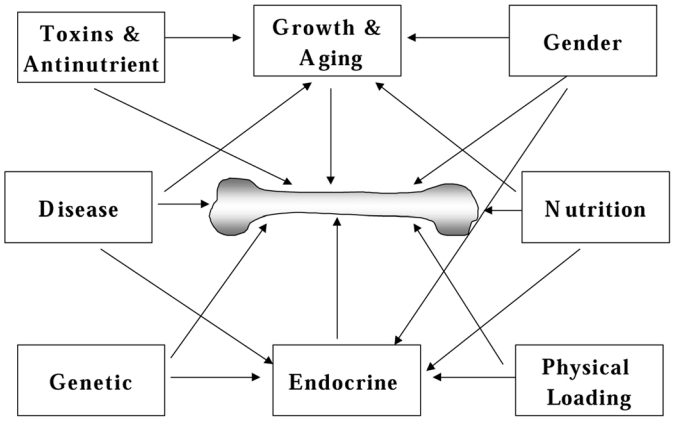

FIGURE 1. Factors affecting bone strength.

Overall, bone strength is related to its physical (shape, size, mass), architectural (collagen fiber orientation), and material (matrix molecules) properties. A deformed bone, such as a bent tibia, will have different strength as compared to a normal tibia despite similar material properties, such as its mineral and organic matrices. Similarly, changes in the matrix properties such as low calcification due to osteomalacia or over hydroxylation of collagen can impede collagen crosslinks and can alter bone strength.

FACTORS REGULATING BONE STRENGTH

Many factors can affect bone strength through their direct or indirect effects on bones. There is a vast literature on these aspects. However, most of the following discussions will be largely limited to factors relevant to poultry bone strength. Figure 1 illustrates different factors that can directly or interactively affect bone strength in poultry.

Growth, Gender, and Aging

One of the important determinants of bone strength is growth because bone mass increases with growth and bone strength is proportional to its mass (Frost, 1997; Seeman, 1999). However, age-related decreases in tensile strength in cortical and cancellous bone have been noted in humans (Wall et al., 1979; Danielson et al., 1993). There are very limited data on the age-related changes in bone parameters of poultry. McCoy et al. (1996) observed an age-dependent increase in bone strength of osteoporotic chickens.

To understand the relationship between growth, maturity, and aging, we compared the changes in the physical, compositional, and mechanical properties of tibial bones

TABLE 1. Age-related changes in physical, composition, and biomechanical properties of tibial bones from male broiler breeder chickens (n = 10 birds/group)[1]

Variables	5 wk	15 wk	25 wk	35 wk	45 wk	55 wk
Body weight (kg)	0.99[d]	2.57[c]	4.43[b]	4.69[b]	5.58[a]	5.57[a]
Tibia weight (kg)	9.58[c]	26.29[b]	40.29[a]	39.29[a]	42.55[a]	40.67[a]
Tibia length (mm)	87.2[d]	138.4[c]	148.0[b]	152.3[a,b]	155.8[a]	150.5[a,b]
Diaphyseal diameter (mm)	5.48[c]	8.10[b]	9.93[a]	9.02[a,b]	9.62[a,b]	9.52[a,b]
Ash (%)	64.99[a,b]	63.09[b,c]	62.19[c]	66.10[a]	65.89[a]	66.17[a]
Density (g/cc)	1.16[c]	1.25[abc]	1.18[bc]	1.43[a]	1.39[ab]	1.41[ab]
Breaking strength (kg/mm^2)	9.51[a]	4.70[b]	6.07[b]	10.89[a]	9.27[a]	9.78[a]
Strain (mm/mm)	0.05[b]	0.11[a]	0.16[a]	0.11[a]	0.14[a]	0.12[a]
Modulus (kg/mm^2)	361.6[a]	121.2[c]	105.7[c]	247.6[b]	197.9[b]	249.6[b]
HP (pmoles/mg bone)	30.1[d]	84.9[c]	134.6[ab]	135.0[ab]	145.0[a]	102.5[bc]
LP (pmoles/mg bone)	13.7[c]	97.2[b]	226.2[a]	237.3[a]	271.3[a]	210.5[a]
Collagen (mg/mg bone)	0.24	0.23	0.24	0.22	0.22	0.20

[a–d]Values with different superscripts in a row differ significantly ($P \leq 0.05$).

[1]HP = hydroxylysylpyridinoline, LP = lysylpyridinoline.

from 5- to 55-wk-old chickens. The results showed that although the relative collagen content of bone did not change between 5 and 55 wk of age, several allometric, biomechanical, and biochemical parameters showed significant changes. The weight, length, diameter, and pyridinium crosslink content reached maximum at 25 wk of age whereas the mineral content, density, and the breaking strength of bones did not reach maximum until 35 wk of age (Table 1). The changes in bone ash content were modest but significant between 25 and 35 wk of age. These results indicate that attainment of maturity or maximal physical and functional potentials of bone takes longer than the growth process itself. In an earlier study we have shown a high correlation between the changes in the crosslink content of bones and the bone strength in birds of 7 and 72 wk of age (Rath et al., 1999). However, in younger birds of 5 wk of age or less, bone strength was not related to the collagen crosslink content because 5-wk-old chickens showed higher breaking strength and low pyridinium crosslink content. Moreover, the bones were relatively brittle as indicated by high stress and modulus values and low strain values, which could be attributed to a relatively higher mineral matrix content relative to crosslink content. During the subsequent weeks of growth, the crosslink content increased several fold as compared to bone ash content, making the bones tougher and less brittle (Table 1).

In another study, we compared the bone strength and the ash content of chickens from Day 1 through 43. Both measures increased from Day 1 to a peak between 3 to 5 wk of age, followed by a subsequent decrease in strength, but not ash content, which remained unchanged (Figure 2). The results from these two studies indicate that the bone biomechanical properties can change according to the dynamics of physical and physiological changes in bone, causing a relatively stiff bone in early phases of growth to be replaced by a tougher bone. These changes may be related to the changes in collagen crosslinks. A comparison of bones from 25-, 75-, and 150-wk-old laying hens showed that 75-wk-old birds have stronger bones than the 25-wk-old birds, which did not change significantly at 150 wk of age (data not shown). Although these studies are limited to only 1 to 3 yr, which is far less than the longevity of the birds, these results do suggest that under controlled conditions of feeding, bone strength of poultry can be adequately maintained to last their productive life. On the other hand, the pentosidine levels increased with age (Table 2). Pentosidine is related to aging and may contribute to collagen stiffening associated with aging (Bank et al., 1998).

Gender is another factor that influences bone growth. In mammals, appendicular skeletal growth has been known to be gender specific (Seeman, 1999). Size differences, as well as hormonal differences, can account for the differences in the growth and bone strength between males and females. Male and female birds of the same age show different diaphyseal diameters, with the females showing a consistently lower value (Rath et al., 1999). However, a comparative study with 7- and 72-wk-old

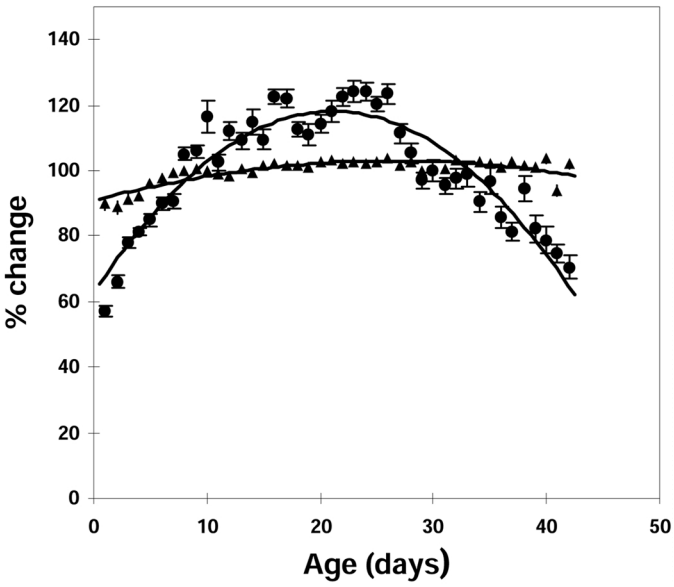

FIGURE 2. Changes in the ash content and breaking strength of tibia from male broiler chickens (n = 6 per point) from age Days 1 through 43. Each value represents percentage of change relative to Day 1 values calculated from six tibias derived from individual birds. (●) ash percentage: (▲) stress (kg/mm^2) at break or breaking strength.

TABLE 2. Age-related changes in the pentosidine[1] content of bone collagen in male (5- to 55-wk-old) and female (25, 75, and 150-wk-old) breeder chickens (n = 10)

Males						Females		
5 wk	15 wk	25 wk	35 wk	45 wk	55 wk	25 wk	75 wk	150 wk
0.17[d]	0.42[cd]	0.73[bc]	1.01[ab]	1.17[a]	1.21[a]	0.39[c]	0.71[b]	2.15[a]

[a–d]Means with the same superscript are not significantly different ($P \leq 0.05$).

[1]The pentosidine levels are expressed as millimoles per mole of collagen.

male and female chickens showed no significant differences in bone strength between young males and females. The bones from 72-wk-old hens were stronger and more rigid and had a lowered strain value, which is perhaps due to the presence of medullary bones.

Genetics

Genetics are another determinant of bone strength. In a number of studies of humans and other mammalian species, bone density has been shown to be a heritable trait (Boskey et al., 1999). Mutations in collagen can affect collagen synthesis, fibrillogenesis, or their posttranslational modifications, which could alter matrix chemistry and mineralization and predispose bone to fragility (Boskey et al., 1999). One well-known example is osteogenesis imperfecta in humans. Genetic defects causing bone weakness in poultry are not known, although tibial dyschondroplasia has been shown to be heritable (Leach and Nesheim, 1965; Wong-Valle et al., 1993) and could indirectly contribute to bone weakness in poultry during early growth periods. It is generally considered that modern poultry has been bred for superior meat production, possibly overlooking the consequences on bone quality. Nevertheless, the heavy-bred birds do not show any weaker bones than their lighter-weight egg-type counterparts (Newman and Leeson, 1997). Nestor and Emmerson (1990), with different genetic lines of turkeys, have shown that the birds with better walking ability had higher body weight as a consequence, perhaps, of better feed consumption. Their data indirectly suggest that birds with higher body weights may have stronger bones.

Physical Activity and Mechanical Stress

Bone adapts to changes in physical loading and activities by modeling and remodeling. Bone mass and strength increases with use (Lanyon, 1993). Cage layer osteoporosis in hens severely reduces bone strength because of high bone turnover related to eggshell formation and inadequate physical activity. Osteoporosis has been a major problem in the poultry industry because of the high rate of fracture and mortality (Newman and Leeson, 1997). However, the problem can be confounded by dietary insufficiency of nutrients such as Ca, P, and vitamins that maintain bone. Cortical bone loss is a risk factor for fracture. The effects of physical loading on cortical bone parameters has been shown by Lanyon (1993) using turkeys. Those studies have shown that physical loading is essential for the maintenance of cortical bone mass. Maintaining sufficient bone mass, architecture, and material properties may help improve cage layer osteoporosis. Several studies of alternative rearing systems for egg layers have included physical activities that would enhance bone strength (Whitehead, 1996; Newman and Leeson, 1997). These studies have shown positive effect and decreasing osteopenia in cage layers.

Nutrition and Vitamins

The role of nutritional factors is probably most relevant to poultry bone strength. This role can be subdivided into two categories: inorganic nutrients and organic nutrients. Calcium and P are primary inorganic nutrients because they form 95% of the mineral matrices, although there are several other inorganic elements present in the bone that may be important for bone health and strength. There are many studies of Ca and P that provide the basis for specific recommendations by the National Research Council (1994) for dietary Ca and P in broilers, breeders, and layers. Calcium homeostasis is an important driving force in the maintenance of bone strength. Low Ca stimulates secretion of PTH and vitamin D synthesis, which in turn activate release of bone minerals. Therefore, adequate Ca is necessary to decrease bone turnover. In poultry management, Ca deficiency does not seem to be a problem. However, there can be problems of malabsorption that can impair intestinal Ca absorption (Perry et al., 1991). Other contraindicative factors are high levels of phytate and cellulose fibers in the diet, which can interfere with Ca absorption. Hypocalcemia, or inadequate Ca levels in blood, is likely to decrease bone strength. Because vitamin D facilitates Ca absorption, supplementation of the diet with adequate Ca and vitamin D can be useful. In laying hens, high bone utilization can cause hypocalcemia; therefore, adequate dietary supplementation of Ca and vitamin D can improve bone strength. Similarly, an imbalance in P metabolism can affect skeletal integrity and strength. Because of the complex interaction among Ca, P, vitamin D, and other calcitropic hormones, it is necessary to judiciously balance the amount of Ca and P added in the poultry diet. Fluoride is another element that has been shown to be beneficial to bone by increasing bone density in chickens (Lundy et al., 1992; Rennie et al., 1997). However, many mammalian studies have shown that excess F consumption can also increase the risk of fracture (Bayley et al., 1990). A recent study of laying hens has shown dietary supplementation with bo-

ron improves bone strength (Wilson and Ruzler, 1998). A deficiency of Cu was shown to decrease collagen crosslink formation and to lower mineralization (Osphal et al., 1982). Aluminum, which also occurs in bone in trace levels, has been linked to osteomalacia and osteoporosis in mammals. In chickens, excess Al in feed produces generalized growth depression (Huff et al., 1996) and reduces bone strength (Johnson et al., 1992).

Vitamins are important feed additives in poultry diets. Because vitamin D is a calcitropic hormone involved in Ca absorption in the intestine, it is widely used as a feed supplement (Sanders and Edwards, 1991; Roland and Rao, 1992; Edwards, 1993; Newman and Leeson, 1999). Vitamin D has a major regulatory role in bone metabolism and bone strength (National Research Council, 1994). In human studies, Ca with vitamin D appeared to be superior to Ca alone in reducing the incidence of fracture by 50% (Dawson-Hughes et al., 1997). Because egg laying causes a heavy demand for Ca, it is essential to supplement vitamin D to maintain optimal bone health in laying hens. Occasional field rickets in certain flocks of birds can produce a major disaster due to poor mineralization and weak bones, which can be traced to vitamin D deficiency or vitamin D metabolism (Huff et al., unpublished data). Besides vitamin D, vitamins B_6, C, and K are integral to bone health because of their involvement in the synthesis of matrix constituents, such as collagen and osteocalcin, and formation of collagen crosslinks (Weber, 1999). Vitamin B_6 (pyridoxine) deficiency affects collagen crosslinks and bone mechanical properties (Masse et al., 1996). In addition, supplemental protein and carbohydrate as energy sources are also important for bone health. However, excess protein intake can produce negative Ca balance and stunt bone growth (Heany, 1998). Therefore, supplemental Ca with protein may be necessary to maintain optimal Ca balance and bone health. Similarly, diets consisting of highly saturated fats can have an adverse effect on bone mineralization, and low-fat diets can increase cancellous bone strength and bone mineral content (Wohl et al., 1998).

Infection and Immunity

Infection and stress can be risk factors for bone integrity leading to bone weakness. Although structural elements greatly contribute to the maturity and strength of bone, these elements can be affected under adverse conditions such as stress, infections, and inflammation. Bone infections such as osteomyelitis and osteonecrosis cause focal bone loss leading to bone weakness (Reece, 1992). Reo viruses cause "brittle bone syndrome" in chickens (van der Heide et al., 1981), although the exact mechanism is not understood. Avian leukosis virus has been implicated in avian osteopetrosis (Smith, 1982).

Inflammation induced-interleukin-6 (Il-6) and tumor necrosis factor (TNF) can enhance bone resorption (Manolagas, 1998). Although these aspects are not much studied in poultry, these factors can affect bone metabolism and strength. Changes in the structural integrity of the collagen framework, without affecting the bone Ca content, may change the mechanical properties and strength of bone. Immunosuppression causes osteopenia in mammals and affects cancellous and trabecular bone volumes (Cvetkovic et al., 1994: Erben et al., 1998).

Hormones and Cytokines

Hormones and cytokines have a profound effect on bone metabolism, growth, and remodeling, and, therefore, are consequential to their strength. Growth hormone facilitates cortical bone growth, whereas steroid hormones affect cancellous bone formation (Brook, 1995). Estrogen deficiency has long been recognized as a risk factor for osteoporosis and bone fragility. However, poultry caged layer osteoporosis is not related to sex hormone insufficiency. The medullary osteogenesis in poultry is under the control of the synergistic action of estrogen and androgens (Dacke et al., 1993). Our studies on the effects of various sex steroids on adolescent chickens showed that testosterone implants caused a significant increase in bone strength of young chickens (Rath et al., 1996). Corticosteroid hormones have strongly been implicated in mammalian osteoporosis (Reid, 1998). These steroids have multitudes of effects on cells such as slowing cell division and differentiation. In mature animals, corticosteroids can affect remodeling, perhaps by preventing the recruitment of osteoblasts and causing bone to weaken by preventing normal bone formation. We found the synthetic corticosteroid, dexamethasone, decreased bone strength of turkeys, although the severity of the effect was age dependent. Seven-week-old turkeys were more profoundly affected as compared to 22-wk-old birds (Table 3). The turkeys from both age groups were treated with 2 mg dexamethasone/kg BW three times on alternate days, 2 wk before sacrifice. The bone growth of 7-wk-old birds was severely stunted. The severity of the effects of dexamethasone was much reduced when the drug was administered at an age when bone development had nearly reached completion. Because there were no differences in any other biochemical or physical parameters of bones from these birds with severely stunted bone growth, we surmise that the observed effects of dexamethasone was due to the reduction in bone mass and remodeling process. These results reiterate the fact that bone mass is an important parameter for bone strength.

Toxins and Antinutrients

Some of the major factors that affect bone strength are antinutrients. These are various biotic and abiotic factors that affect bone growth, metabolism, and matrix constituents, thereby affecting bone strength. Lathyritic agent b-aminopropionitrile of certain sweet peas inhibits the enzyme lysyl oxidase, preventing collagen crosslinks and mineralization, which decreases bone strength (Seigel and Fu, 1976). Mycotoxin-contaminated diets can affect growth and cause bone fragility and decrease bone strength (Huff et al., 1980; Maurice et al., 1983). Many

TABLE 3. Age-dependent effects of dexamethasone (Dex) on turkey bone strength[1]

	Control			Dex		
Age (wk)	Load (kg)	Breaking strength (kg/mm^2)	Modulus (kg/mm^2)	Load (kg)	Breaking strength (kg/mm^2)	Modulus (kg/mm^2)
7 (n = 10)	33.2	10.2	444.9	18.4*	7.4*	360.3*
22 (n = 6)	60.5	7.9	841.2	48.9*	6.8*	716.8*

[1]Turkeys were given three injections of dexamethasone (Dex) on alternate days at a dose of 2 mg/kg BW 2 wk prior to euthanasia, where upon the tibial bones were harvested and used for biomechanical measurements.

*Significantly different ($P \leq 0.05$) from the control values.

antinutrients such as mycotoxins affect bone strength indirectly by affecting the metabolism of factors such as vitamin D that are essential for bone health (Duff et al., 1987).

IMPLICATIONS AND CONCLUSIONS

In poultry management, nutrition is central to the maintenance of skeletal health. Traditionally, Ca, P, and vitamin D supplements in feed have been used for the maintenance of skeletal quality and integrity because of their role in mineralization. Recently, more research on the use of phytase enzyme to improve availability of phytate-bound P have been undertaken (Edwards, 1993; Sohail and Roland, 1999). From the foregoing discussion, it should be evident that not only bone minerals, but also the bone organic matrix factors, are responsible for bone strength. The ash content and density between young and matured birds differ only by 5 to 10%, yet the strength of bone increases in correlation with collagen crosslink content (Rath et al., 1999), which raises the possibility that increasing collagen crosslink content of bones of adolescent birds will increase bone strength and mineralization. Whether it is feasible to preemptively increase crosslink content through nutritional interventions is not known. However, reduction of collagen crosslinks by inhibiting the enzyme lysyl oxidase with lathyritic agents or through experimentally induced Cu deficiency can reduce calcification and, consequently, bone strength (Osphal et al., 1982). However, there is no experimental evidence to suggest that increasing collagen crosslink formation will increase bone strength. Also, because growth and maturity are developmentally regulated, it is not clear if the process can be expedited. In adult birds such as laying hens, supplementing diets with vitamin D, Ca, P, phytase, vitamin C, and vitamin K, as well as protein energy enrichment diets, may be beneficial in maintaining bone mass and, thereby, bone strength. Because bone formation is an early event related to growth, nutritional interventions directed toward producing better bone mass may be helpful. Alternative design of cages to facilitate certain physical exercise has been considered to improve bone strength in laying hens (Rowland and Harms, 1970; Newman and Leeson, 1997).

In addressing bone strength in poultry, one cannot ignore the fact that skeletal deformity is one of the major risk factors for bone weakness and fragility. It is known that tibial dyschondroplasia (TD), which affects poultry during the early growth period, can produce bone deformities such as bending of the tibia due to metaphyseal weakness (Lynch et al., 1992). Although TD is eventually resolved and replaced by bone (Rath et al., 1994), this may not correct bone deformities. Tibial dyschondroplasia is also one of the main causes of bone breakage in market age poultry. From the foregoing discussion, it is clear that bone shape and architecture are some of the important factors related to bone strength. Therefore, reducing the problem of TD will lead to improvements in poultry bone strength.

In conclusion, it is appropriate to consider that, through strategic nutritional manipulations and management practices, it may be possible to increase or preserve bone mass and to maintain bone strength. Exploring the nutritional requirements that would improve collagen crosslinks may be useful in improving bone quality in meat-type poultry.

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