## Technical Update



# UNDERSTANDING THE ROLE OF THE SKELETON IN EGG PRODUCTION

Soft bones and thin fragile bones are issues that affect producers of high-performing egg layers all around the world. Osteomalacia and osteoporosis most commonly arise from deficiency, imbalance or malabsorption of calcium, phosphorus, or vitamin  $D_3$ . The effect of skeletal issues on a flock typically is demonstrated by decreased production, crooked keels, fractures and poor shell quality.

All hens require a specific amount of nutrients to maintain production and skeletal structure. The skeletons and dietary requirements of hens are unique relative to the level of calcium consumed and the amount of bone that is constantly being built and resorbed. When soft bones or poor shell quality is found, usually at least one of the following factors is involved: pullet growing, nutrition, feed consumption, or disease.

#### GLOSSARY

Crooked: not straight; having curves

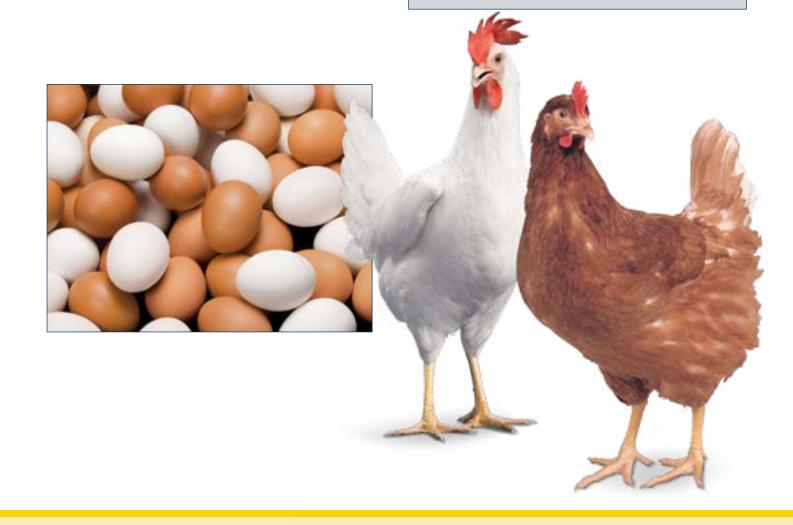
Labile: constantly undergoing change

**Osteomalacia**: decalcification or softening of bones due to bone mineralization issues caused by insufficient levels of available phosphorus and calcium or due to excessive resorption of calcium from the bone

**Osteoporosis:** a progressive bone disease resulting in decreased bone mass and density

**Resorb**: to dissolve/break down and assimilate

**Resorption**: destruction or loss of tissue or bone



#### **OVERVIEW OF BONE BIOLOGY**

The avian skeleton is a unique system that is specialized for flying, walking on two legs and laying eggs. Establishing and maintaining a strong skeleton is vital to ensure a productive laying hen. In order to understand the impact of diet on the laying hen, it is important to understand the biology of the skeleton. There are three different types of bone: cortical, trabecular and medullary.

- Cortical bone is the hard outer surface of the round bones, such as the femur or the humerus, and the flat bones, such as the skull or the pelvis.
- Trabecular or spongy bone is less dense than cortical bone and helps support the structure inside the cortical bones.
- Medullary bone is a specialized woven bone which serves as a calcium reserve for the demands of egg shell formation. Easily created and resorbed, medullary bone is ideally the first source mobilized when more calcium is required.  $\overline{\mathcal{A}}$

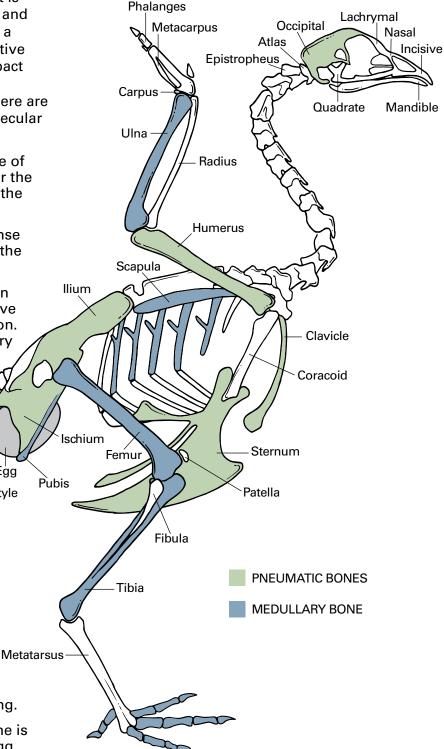
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While the outside appearance of avian bones is similar to those of mammals, there are several key differences.

- Fused vertebrae Several Pygostyle thoracic and lumbar vertebral spinal sections are fused together to form a more solid structure for flying.
- Keel The sternum or keel provides a large surface for attachment of the pectoral muscles which are important for energy storage and muscle yield.
- Pneumatic bones Hollow and air filled, these bones are part of the respiratory system and help with flying.
- Medullary bone This specialized bone is used as a source of calcium for the egg shell and only occurs in birds and some reptiles.

Bone growth and resorption is controlled and regulated by a few important cells and many different hormones. In healthy, well-fed birds, the cells and hormones work closely together to maintain bone structure and blood calcium levels needed for optimum production.





The important cells for bone growth and modelling are chondrocytes, osteoblasts, and osteoclasts.

Chondrocytes start the basic process for bone growth by secreting type II collagen and other important components for bone formation.

- Osteoblasts then produce the type I collagen and the increased levels of calcium and phosphate that result in the mineralization or ossification of the bone.
- Osteoclasts resorb bone for remodelling or for releasing calcium into the blood stream.

Bone growth and resorption is regulated by a number of different hormones which control when structural or medullary bone grows or resorbs, depending on the physiologic need.

- Growth hormone stimulates cellular growth and protein synthesis throughout the body.
- Thyroxine stimulates cell metabolism as well as osteoblast activity.
- Melatonin influences osteoblast activity. Melatonin levels are highest when birds are sleeping during the dark period and initiate a cascade of events affecting hormones necessary for egg production.
- Estrogen increases at sexual maturity and changes osteoblast activity from creating cortical and trabecular bone to creating medullary bone. After the first egg, the only way a hen can remodel structural bone is during periods of low estrogen, such as molt or breaks in lay during the normal production period.
- Calcitonin is released when there are high serum calcium levels and decreases osteoclast activity while increasing osteoblast activity which builds bone and lowers serum calcium levels.
- Parathyroid hormone (PTH) is released during periods of low serum calcium and binds to osteoblasts. This binding decreases osteoblast activity while releasing a compound that increases osteoclast activity, thus increasing serum calcium levels. Additional properties of PTH include increasing small intestine absorption of calcium and decreasing urinary excretion of calcium.
- Calcitonin and parathyroid hormone work together in feedback loops to ensure the proper levels of serum calcium are maintained.

## THE IMPORTANCE OF GOOD QUALITY PULLETS

A strong skeleton starts with good pullet quality. Best management practices should always be used when growing pullets. Please refer to the Hy-Line International Technical Update "Growing Management of Commercial Pullets" for information regarding pullet programs.

Chicks hatch with relatively underdeveloped internal organs and systems. The main systems that are developing in the initial weeks after hatch are the intestinal tract, the immune system and the integument (skin and feathers). The development of the intestine is crucial for nutrient absorption and will determine a hen's future production efficiency. Strong intestinal development will also strengthen the immune system and minimize the possibility of future enteric diseases.

Starting at about six weeks of age, pullets have a more mature intestinal tract and immune system and also are able to regulate body temperature which allows for more energy to be allocated for growth. The fastest rate of growth for the skeleton occurs between 6 and 12 weeks of age. During this time period, layer pullets gain an average of 90 to 110 grams (0.20–0.24 lb) of body weight per week. By 12 weeks of age, the skeleton is 95% developed and once the bone growth plates close around the time of sexual maturity, no more bone length can be added. Any delay in growth will affect the size of the mature bird and delay the onset of production.

At 13 weeks of age pullets have reached about 95% of their adult size, but only 75% of their mature weight. Over the next 6 weeks muscle, medullary bone and reproductive tract development will constitute much of the weight gain. Once birds have reached the appropriate level of development as determined by body weight, the flock will be ready for light stimulation to start egg production. Laying hens will continue to add muscle and bone mass and gain weight until around 32 weeks when the full mature body weight is achieved.

A pullet flock that experiences higher levels of stress during periods of rapid growth is more likely to have poor uniformity that can affect peak production. When a flock lacks uniformity, the lighter birds will not come into production for up to 10 weeks after birds that meet or exceed ideal body weights. Therefore while most of the flock may be laying at 96%, the 5% of the flock that is underweight may be laying sporadically (or not at all) and will reduce the peak production percentage. Stressors that may be avoided include moving, injecting with inactivated vaccine, wide ranges of environmental conditions inside the house, poor feed quality, crowding and any other abrupt changes in routine.

Crowding in the pullet house usually starts to affect birds at approximately 10–12 weeks when the skeleton is near full size. Crowded pullets will have issues with uniformity and body weight gain until transfer. Refer to the Performance Standards Manual for rearing space recommendations.

Measuring body weight gain and uniformity is an excellent method for tracking flock growth throughout pullet growing. Higher body weights result in larger skeletons and more muscle mass which leads to better production. Waiting to light stimulate until target weights are met is the most effective solution to ensure good persistency of lay and avoid a post-peak dip in production. Monitoring body weights every week should not stop at transfer. It is ideal to weigh every week up to 32 weeks of age and at least every 2 to 4 weeks until the end of lay. This practice will give an indication if nutrient intake is sufficient to support production, growth, and maintenance requirements of the bird.

#### SEXUAL MATURITY IN THE LAYING HEN

About two weeks prior to egg production, the hen will undergo sexual maturity. An increase of estrogen will stimulate development of the oviduct, reddening of the comb and wattles and a complete transition from building skeletal bone to building medullary bone.

To aid the growth of medullary bone before the first egg, it is recommended to introduce more calcium through the use of a Pre-Lay Diet. Constantly improving genetics provide producers with layers capable of very high peaks of lay and good persistency. To ensure that genetic potential is reached, building medullary bone and formulating diets with sufficient nutrient density to meet the daily requirements of the bird is very important. However, there can be a negative impact on feed consumption from the sudden increase in dietary calcium levels of 1% to above 4% at the start of lay. Field experience indicates that the use of Pre-Lay Diets helps as a transition between the Developer and the Peaking Diet. Correct feed formulation and matching diet density with consumption will minimize the impact of reduced calcification of bone over the laying cycle and extend the persistency of shell quality.

#### BONE QUALITY DURING PRODUCTION

The length and width of the poultry skeleton is complete when the hen has started to lay. However, the bone mineral density and content, as well as the ratio of cortical, trabecular and medullary bones can change dramatically. The laying hen skeleton is strongly influenced by level of egg production, diet formulation in relation to consumption and disease status. A well-grown laying hen will typically not face skeletal issues until after peak, even with mild to moderately deficient diets. With an underweight laying hen, nutrient deficiency will more quickly affect flock results. Mild to moderate nutrient deficiencies will usually cause skeletal and/or shell quality issues first with production issues following. Severe nutrient deficiency will still cause noticeable and rapid drops in production.

Many animals, including birds, experience thinning of the cortical and trabecular bone thickness with age. Laying hens also experience changes in overall bone strength. Although medullary bone is the most labile bone type, if a hen is calcium deficient, cortical and trabecular bone will also be mobilized as a calcium source. During the laying period, hens have been shown to have a net increase in bone mass as a result of medullary bone formation and the loss of structural bone. However, without a molt or cessation of production due to nutrient deficiencies, the constant high level of estrogen in a laying hen will prevent the repair of structural bone. Loss of cortical bone can result in crooked keel or bone fractures, both of which are detrimental to hen welfare and production.

As birds age, medullary bone content will increase while structural bone integrity typically decreases. Birds that lose too much cortical bone and gain medullary bone can have good shell quality, but are at a higher risk for keel curvature or bone fracture. The best way to minimize the loss of cortical bone is to ensure that the correct levels of calcium, phosphorus and vitamin  $D_3$  are fed throughout the lay cycle. Preventing loss of skeletal integrity starts with the pullet, but continues through the life of the bird. It is also important to understand the clinical signs of osteomalacia, osteoporosis and osteopenia and make the appropriate changes as soon as possible.

#### MONITORING SKELETAL INTEGRITY

The best way to minimize the impact of soft bones in a flock is to monitor regularly. Handling birds while weighing is the ideal way to collect this information. Select birds from the same cage, colony or section of the barn at least every 4 weeks to ensure consistency of data and provide early notice if crooked keels are observed.

- Keels are scored by feel and observation. The Hy-Line method is to score on a four point scale based on normal (#1), mild (#2), moderate (#3) or severe (#4) curvature.
- At the beginning of calcium, phosphorus or vitamin D<sub>3</sub> deficiencies, keels may be flexible, but not yet curved. This is an important clinical sign to note.
- Hens with recalcified, curved keels would indicate a nutritional deficiency earlier in the life of the flock.
- It is ideal to handle a minimum of 10 birds in at least 2 to 3 areas of a barn.

Overall, it is ideal to have greater than 90% of birds handled in the Score 1 or Score 2 category. More than 10% Score 3 or Score 4 birds, or increasing numbers every week, indicate the possibility of an issue.

#### #1 – Normal Keel





#### #2 – Mild Curvature





#### #3 – Moderate Curvature





#4 – Severe Curvature





#### NUTRITION

Feed consumption and nutrition always go hand in hand. All diet formulations must be based on feed consumption to ensure proper intake of the critical nutrients. As a result, all Hy-Line diets are recommended on the basis of total daily feed intake. Focusing on calcium and available phosphorus, as examples, the Hy-Line Brown and Hy-Line W-36 need to consume the following amounts each day in her ration:

If the hen is not effectively absorbing the calcium in the diet, she can be deficient (even with an accurate "calculated value" of the ration).

Additionally, the source of limestone should be checked. Low calcium content limestone (less than 37%) may contain other minerals that reduce the solubility (and therefore the calcium availability).

	Peakin	g Diet	Lay D	iet #2	Lay D	iet #3	Lay D	iet #4
	Hy-Line Brown	Hy-Line W-36	Hy-Line Brown	Hy-Line W-36	Hy-Line Brown	Hy-Line W-36	Hy-Line Brown	Hy-Line W-36
Calcium (g/day)	4.20	4.10	4.30	4.30	4.50	4.45	4.80	4.60
Available Phosphorus (mg/day)	460	485	420	470	380	450	360	400

Attaining the required levels for these nutrients is significantly dependant on daily feed consumption. A sample calculation for a bird recommended to have 4.00 grams of calcium per day and with an observed feed consumption of 95 grams is as follows:

4.00 g calcium		
needed x 100	=	4.21% c
95 g feed consumed	-	in the d

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Limestone particle size is also important for optimum shell quality. Pullets should have fine particle calcium, ideally less than 1.1 mm (1100 micron) average. It is best to use limestone flour for pullets as the smaller particles are more easily absorbed.

Layers should ideally receive a 50:50 ratio of large and fine particle limestone at the start of lay and transition to a 65:35 ratio (large: fine) by the end of lay. Changing the particle size ensures that more calcium will be available at night from the diet instead of from the bone. Further measures to increase night calcium availability include ensuring a last feeding 1-2 hours before lights are turned off and adjusting the feeding regime to 40% in the morning and 60% in the afternoon.

The large particle fed during lay should be around 2-4 mm (2000-4000 micron) average size with an ideal 3 mm (3000 micron) size. When calcium particle size is above 3.5 mm (3500 micron), the solubility rapidly decreases.

Depending on geological formation, different limestone structures exist which may also affect solubility and availability.

Phosphorus intake is similarly calculated, although the requirement definition is more complex due to different systems used to express available and digestible phosphorus. Care needs to be taken that requirements and the availability matrix are being expressed on the same system. There are ongoing projects in the US and the EU to review the phosphorus nutritional systems and create a more universal standard.

The use of phytase enzymes must be considered when formulating the diet. Phytase is an important tool in reducing both the cost and environmental burden of animal feeding; however care needs to be taken when applying matrix values. Matrix contributions need to be accurate for the phytase being used, the diet in which it is being used and the dosage being applied. The matrix contribution must not exceed the concentration of phytate phosphorus potentially available for release. This differs by phytase source, inclusion rate and the ingredient makeup of the diet.

Vitamin D<sub>3</sub> (cholecalciferol) is a critical nutrient that aids in calcium and phosphorus uptake in the small intestine, bone mineralization, inhibition of calcium excretion in the urine and immune system modulation. Vitamin D is typically included in the feed as vitamin  $D_3$ . It is absorbed in the small intestine and converted to 25-hydroxycholecalciferol in the liver.

This metabolite is then converted to the active form of 1, 25-hydroxycholecalciferol in the kidneys. Instead of vitamin  $D_3$ , 25-hydroxycholecalciferol can be used as a supplement for the hen. A more biologically active metabolite of vitamin  $D_3$ , 25-hydroxycholecalciferol provides a higher dose of vitamin D at a lower inclusion rate.

Other dietary factors are also important to consider in the efficiency of bone calcification and eggshell formation. This includes the acid base balance of the diet (or dietary electrolyte balance - DEB), vitamin K, zinc, copper, iron, manganese and magnesium. Some essential amino acids which are often limiting in laying diets (valine and arginine) may be important due to their role in calcium transportation and formation of the bone matrix.

#### FEED CONSUMPTION

Consumption volumes at the start of lay change quickly; for example, the Hy-Line Brown eats 80– 90 g/day at the start of lay and soon eats 110–115 g/day in about 4–6 weeks at peak production. Often, feed mills may only have one Peaking Diet that is formulated for 110–115 g/day intake for brown birds and 100–105 g/day for white birds. When intake at the start of lay is only 80–90 g/day, there will be deficiencies of 20–25% in calcium, phosphorus, essential amino acids and other nutrients. Creating a diet matrix can help ensure that the correct dietary formulation is used. Not all diets will be used, but it will simplify ordering feed for a farm. The bird is still growing until about 32 weeks and is adding muscle mass and bone density. If there are deficiencies in the diet, the impact may not be immediate, but can be felt late in lay when hen body reserves have been depleted.

#### DISEASE

Nutritional issues are usually the primary cause for decreased skeletal integrity and resulting shell quality problems. However, many subclinical respiratory and enteric diseases may have the same effect. A decrease in feed consumption or nutrient absorption can have a dramatic impact on shell and bone quality as described previously. Bacterial, viral or protozoal pathogens may cause temporary or permanent damage to the intestine and reduce the absorption of key nutrients. The duodenum in particular is the location in the intestine where calcium is actively absorbed in response to increased demand under hormonal influence involving vitamin D<sub>3</sub>. Diseases such as focal duodenal necrosis can damage the duodenum and may decrease the efficiency of absorption. Additionally, there are many diseases that cause shell quality issues by affecting the oviduct, including infectious bronchitis, Newcastle disease, Mycoplasma synoviae, egg drop syndrome (EDS) and avian influenza.

90 g/day  x      95 g/day       100 g/day  .x      105 g/day       110 g/day  .x      110 g/day  .x			Peaking Diet	Lay Diet #2	Lay Diet #3	Lay Diet #4
105 g/day	tion	90 g/day	x			
105 g/day	duns	95 g/day				
		100 g/day	x			
110 g/day x x x x		105 g/day				
	Daily	110 g/day	x	x	x	x

## ACTIONS WHEN SKELETAL ISSUES ARE IDENTIFIED

During routine handling, if soft bones or crooked keels are detected during lay, there are steps that can be taken to correct the issue.

**General** – should only be used until specific measures can be taken

- Add water-soluble vitamin D<sub>3</sub> or 25-hydroxycholecalciferol to the drinking water 1–2 times a week to increase calcium and phosphorus metabolism
- Add 2–4 mm large particle limestone or oyster shell to increase the level of calcium in the gizzard overnight and provide calcium for egg shell and strengthening bones.
- Review, and if necessary, increase the level of available phosphorus in the diet to help with bone strength and metabolism

#### Specific

- Verify or determine feed consumption of the flock
- Check the feed formula to ensure the correct amounts of calcium, phosphorus and vitamin D are specified
- Ensure that the daily levels of consumption meet the dietary needs of the hen
- Send feed samples for analysis to determine if calcium and total phosphorus levels are consistent with formulated values. When sampling, it is important to obtain a representative sample of the feed to minimize sampling error. A standard procedure for collecting accurate feed samples is to collect multiple sub-samples and mix them together. Use a portion of this mixed sample to send in for analysis.
- If a deficiency is found, work with the feed mill to prepare the correct formulation based on daily consumption
- Check the flock to see if there are any disease issues that are minimizing absorption or reducing feed consumption

Ensuring good skeletal development from rear into lay through best management and nutritional practices is essential to reach the genetic potential of the Hy-Line layer. Understanding the importance of the skeleton in laying hen production will help producers implement best practices. Formulating the diet to consumption and monitoring the skeletal quality will create the environment to attain the best and most economical production.



