BONE AS AN ENDOCRINE ORGAN AND MINERAL NUTRITION OF DAIRY COW

D.M. MCNEILL1 and S.T. ANDERSON2

1School of Veterinary Science The University of Queensland
Gatton, Queensland 4343, Australia
d.mcneill@uq.edu.au
2School of Biomedical Sciences, The University of Queensland
St Lucia, Queensland 4072, Australia

ABSTRACT

In this review we highlight the recent discovery that bone is an endocrine organ and, inspired by this, propose new directions for research in dairy cow nutrition. Bone tissue releases a hormone called undercarboxylated osteocalcin that directly affects insulin secretion and glucose homeostasis, and is therefore linked to diabetes, at least in mice. Based on this finding, the hypothesis of “healthy bone for a healthy body” is proposed in relation to improved productivity in dairy cows. That is, feeding a cow to promote bone health will optimize lifetime productivity. This hypothesis hinges on the expectation that an improvement in a cow’s ability to maintain both calcium and glucose homeostasis will improve milk yield and reduce the risk of metabolic diseases such as milk fever (hypocalcemia), grass tetany (hypomagnesemia), hypophosphatemia or hyperphosphatemia, and ketosis. The likelihood that these disease states are causally related is also explored. It is concluded that a focus on macro-mineral nutrition, using bone metabolism markers as a guide, could foster the development of more successful feeding strategies for improved production in dairy cows.

Key words: Osteocalcin, Insulin, Calcium, Phosphorus, Glucose, Homeostasis

INTRODUCTION

Two of the most important chronic diseases known to man, osteoporosis and diabetes, have recently been linked through the discovery that bone tissue secretes a hormone, undercarboxylated osteocalcin (gluOC) that helps protect against type 2 diabetes. This discovery, in a mouse model, is generating great excitement amongst human health oriented researchers, and of course, drug companies. Could there be opportunities arising from this discovery that will also benefit food animal production?

Using the dairy cow as a model, we propose that the exploration of bone as an endocrine organ could develop strategies to improve the efficiency of production and reduce the risk of common metabolic diseases. The modern, high genetic-merit dairy cow presents as an excellent model by virtue of its extreme demand for the key nutrients, calcium and glucose, that are closely related to osteoporosis and diabetes. Whilst, dairy cows are commonly culled well before the later stages of life when osteoporosis and type 2 diabetes might occur in humans, cows still do suffer from similar diseases related to shorter-term perturbations in calcium and glucose homeostasis consequential to high metabolic demands.

Hypocalcemia (Milk fever) and ketosis are two examples of a failure in calcium and glucose homeostasis in the dairy cow. These metabolic diseases, in clinical and subclinical forms, depress milk production and probably fertility. Bone tissue plays an important role in the process of calcium homeostasis and possibly in the development of milk fever. If bone tissue also modifies glucose homeostasis, as found in the mouse, we propose that the state of a cow’s bone may also play a role in the development of ketosis. Hence, the aetiology of hypocalcemia could directly inform the aetiology of ketosis. In discussing this, the following research questions arise. What is “healthy” bone? How do we make bone healthier? Do cattle secrete gluOC? Do cattle with healthier bone secrete more gluOC? Is gluOC secretion related to calcium and glucose homeostasis and the risk of milk fever and ketosis? How do we manipulate gluOC
secretion in the cow and does this lower the risk of metabolic disease and increase productivity? Put simply, the following paper addresses the hypothesis of “healthy bone for a healthy body”. If we feed to advantage an animal’s bone tissue, does improved health and productivity likely follow?

**EVIDENCE FOR BONE AS AN ENDOCRINE ORGAN**

In 2007, Gerald Karsenty and team reported that bone produces a hormone called undercarboxylated osteocalcin (gluOC) that enhances glucose uptake and energy expenditure in a range of tissues throughout the body (Lee et al. 2007, Wolf 2008, Karsenty 2011). Glucose uptake is enhanced by gluOC acting to increase insulin production and secretion by the pancreas, as well as by causing a diverse range of tissues, such as the adipose, liver, and muscle, to become more sensitive to insulin.

Karsenty’s team (Lee et al. 2007) were investigating metabolic pathways in bone tissue by using a “gene knockout” technique in mice to help define the purpose of genes known to play key roles in bone remodelling. In a knockout model, genes are disabled at the embryonic stage using transgenic methodologies, and the consequences noted in subsequent transgenic generations, as they mature. One of the genes disabled was that coding for Osteocalcin (OC). Osteocalcin is the most prevalent type of non-collagenous protein in bone (Gundberg et al. 1984, Ducy et al. 1996). It is produced by the bone-forming cells called osteoblasts and was thought to function as the “glue” that sticks the hydroxypatite molecules together to allow the formation of mineralised bone. Surprisingly, they found that OC was not necessary for bone formation. In fact, OC knockout mice grow bone faster than mice with the OC gene intact (Ducy et al. 1996, Murched et al. 2004, Lee et al. 2007). More importantly, they realised that the OC knockout mice soon became obese and that prompted them to suggest that osteoblast activity is associated with energy metabolism, which they went on to confirm.

The OC knockout mice became diabetic - their tissues were less sensitive to insulin and their pancreas less able to secrete insulin (Lee et al. 2007). Karsenty’s team subsequently disabled another gene, expressed only in osteoblasts, called Esp. When Esp was disabled, the mice appeared protected from the development of diabetes and obesity. They went on piece together a new metabolic pathway that included the OC and Esp genes, or similar, working to regulate the production of undercarboxylated Osteocalcin (Lee et al. 2007, Ferron et al. 2010, Zee et al. 2012). The Esp gene codes for a protein, ESP, that is a member of the protein tyrosine phosphatase (PTPase) family. The mechanism of action of ESP is to impede the action of insulin receptors in the osteoblast which in turn reduces the production of OC and the activity of osteoclasts (the cells that drive resorption of bone). The osteoblasts control the activity of osteoclasts which in turn provide the acidic environment that decarboxylates OC to form gluOC. A minor complication here is that the Esp gene does not occur in the human genome and so may not occur in cattle (Ng 2011). However, Zee et al. (2012) recently confirmed that another member of the PTPase family, T-cell PTPase, encoded by the gene PTPN2, does exist in humans and acts in the same way as Esp to regulate bone resorption and insulin sensitivity. A search of the gene discovery database http://www.ncbi.nlm.nih.gov/gene, indicates at least two references to the existence of PTPN2 in cattle. Hence, communication between bone remodelling and energy metabolism, via gluOC, appears just as likely in cattle as in mice.

Ferron et al. (2012) have since demonstrated that intermittent injections of gluOC negate the development of type 2 diabetes and fatty liver in mice on high fat diets. They conclude that daily OC injections could prove to be a treatment for type 2 diabetes. Opportunities therefore exist for research into strategies to manipulate gluOC secretion in cattle to improve energy regulation and the development of metabolic diseases. Strategies to promote the production of gluOC may be those that facilitate the up-regulation of the activity of osteoblasts and osteoclasts whilst simultaneously down-regulating the activity of genes such as PTPN2. We consider attention to the improvement of bone health as a fundamental step towards this.
DEFINING BONE HEALTH

Inspired by the potential for gluOC to control energy metabolism in animals, we propose several perspectives to the consideration of what defines healthy bone, and not including extreme disease states such as osteochondrosis, bone cancer, and rickets. When bone health is discussed in the human literature the inferred definition is commonly that healthier bone has sufficient cortical bone density and possibly trabecular bone density and architecture to minimize the risk of fracture (Seeman 2008). Hence the focus in human studies is on osteoporosis, where bone density is generally determined by DEXA or CT scanning procedures. Unfortunately, live cattle are too large to fit into currently available DEXA or CT scanners. Bone biopsies can be taken for the measurement of bone density in cattle but the technique is invasive and the interpretive value of the most common biopsy, from the rib, is questionable (Hoey et al., 1982; Taylor et al., 2008). Comparative slaughter studies are another option for cattle. They are expensive and do not allow for repeated measures on the same animal, but should offer a more definitive result than bone mineral balance studies where for example P intake is corrected for P loss in feces and urine to estimate P balance, especially where the aim is to specifically target changes in maternal tissues through pregnancy and lactation.

From the perspective of animal production a further definition is that healthier bone has sufficient reservoir of bone mineral (calcium and phosphorus) to mobilize in times of need, as reviewed by McNeill et al. (2002). Using changes in the appearance of markers of bone metabolism in the plasma and urine, the most likely times to replenish bone is in mid-late lactation, and to mobilize, late pregnancy and early lactation (Liesegang et al., 1998; 2000; McNeill et al., 2002; Taylor et al., 2008; Bhanugopan et al., 2010). Commonly used markers of bone metabolism in cattle are, for bone accretion, OC and bone alkaline phosphatase (BAP), and, for bone resorption, pyrridinoline (PYD) and deoxypyrridinoline (tDPD).

Finally, in view of the existence of gluOC and its potential to minimize the risk of type 2 diabetes, we propose a third definition: healthier bone is that which is more communicative with other tissues and organs of the body. In this sense, healthier bone could be that which is more metabolically active as evidenced by an elevated rate of osteoblast and osteoclast activity coupled with a decline in the activity of the PTPN2 gene to improve the secretion of gluOC.

BONE HEALTH AND OSTEOCALCIN

Since gluOC is produced by the cells in bone responsible for bone growth then, by inference, healthier, actively growing, bone may produce more gluOC which may in turn makes the animal more sensitive to insulin. Consistent with this is the single study that we can find where gluOC has been measured in dairy cows. Kim et al. (2011) administered an exogenous injection of 1.25-hydroxy-Vitamin D3 (Calcitriol) to stimulate bone formation in non-pregnant non-lactating cows, and found that the injection elevated plasma calcium, total OC and gluOC within days. Whilst published data on levels of plasma gluOC in cows is currently lacking, there is good data available on levels of total OC (carboxylated + undercarboxylated forms). Total OC was a well accepted marker of bone formation, or more specifically, of osteoblast activity, well before the discovery of gluOC. In the cow, OC is elevated in the plasma of younger animals, especially foetuses, declines with age, especially prior to the first lactation, is lower in multiparous compared to primiparous cows, declines in late pregnancy and at the time of calving and rises in the days to weeks thereafter. Based on this pattern, which coincides with the pattern of change in the calcium and phosphorus requirements of the cow, OC has been proposed as marker of bone mineral balance (Davicco et al., 1990; Liesegang et al., 2000; Holtenius and Ekelund, 2005; Ekelund et al., 2006; Taylor et al., 2008, Bhanugopan et al., 2010; Kim et al., 2011; Sato et al., 2011).

Yet, despite the variety of publications on OC in cattle, we are yet to find any that have tested for a relationship between gluOC and insulin sensitivity. To test the hypothesis that faster growing bone produces more gluOC, we
are currently finalising an experiment in which young steers have been fed one of 5 different levels of dietary phosphorus for 5 months in order to generate a wide diversity bone growth rates and relate this to changes in insulin sensitivity status, hormones associated with the regulation of calcium homeostasis, plasma concentrations of phosphorus and magnesium, and markers of bone metabolism additional to gluOC and total OC.

**BONE HEALTH AND THE RISK OF METABOLIC DISEASES.**

Goff (2006) provides an excellent summary of advances in the use of nutritional strategies to reduce the risk of the most common metabolic diseases in dairy cows: milk fever (hypocalcaemia), fatty liver (liver lipodosis), and ketosis. Goff (2006) infers that milk fever and ketosis are linked by a sustained reduction in feed intake around the time of calving.

The currently understood cascade of events linking milk fever to ketosis is: calving/milk fever reduces feed intake; mobilisation of fatty acid from adipose tissue to the liver accelerates, especially in overfat cows; plasma glucose declines with a decline in gluconeogenesis by the liver; a further inability of the liver to cope metabolically, likely due to a lack of TCA cycle intermediates. This leads first to a build up of lipid in the cytosol of the liver cells and second to a diversion of fatty acid-derived and gastrointestinal tract-derived acetate from the TCA cycle to ketogenic pathways to form acetoacetate and beta-hydroxybutyrate (Goff 2006).

Consistent with Goff’s summary, Loor et al. (2007) showed the ease with which a hypoglycaemia, fatty liver, and ketosis can be precipitated by a sustained reduction in feed intake for about a week, soon after calving, and measured consequential change in the activities of an impressive range of genes linked to intermediary metabolism and signalling which predominately reacted in expected directions.

The determination of strategies to reduce the risk of both the acute and sub-clinical forms of milk fever and ketosis is now a very popular area of research and almost all focus on improvements to nutrition immediately pre-calving (McNeill et al. 2002). Lean et al. (2006) defined the nutritional risk factors affecting the development of milk fever, with a focus on nutrition in the 2 – 4 weeks pre-calving. The technique of meta-analysis was applied to data from 137 published trials to show that key pre-calving dietary predictors of milk fever are: increased levels of calcium up to approximately 1.5% DM; declining levels of magnesium; an increasing dietary cation to anion difference (DCAD, mEq: (sodium + potassium) – (chloride + sulphur)); potassium, sulphur, and uniquely amongst the majority of milk fever studies, increasing levels of phosphorus. Dietary magnesium is particularly important (Goff 2006). Amongst a range of hypothesised actions, sufficient magnesium links the sustained release of parathyroid hormone, the essential first signal that plasma calcium is declining, to the renal release of 1,25hydroxyvitaminD3 (calcitriol), which in turn stimulates the mobilisation of calcium from the bone and more efficient absorption of calcium from the small intestine. Hence hypocalcaemia, hypomagnesaemia, and hyperphosphatemia are linked.

We suggest that, just as an excessive decline in feed intake at calving links milk fever to ketosis, so too may bone health. Healthier bone may be able to release more bone mineral to buffer dietary inadequacies and so help to stabilise calcium homeostasis. Similarly, healthier bone might release more gluOC to help stabilise glucose homeostasis and slow the development of obesity. Obesity is a well accepted risk factor for ketosis (Goff 2006). Moreover, gluOC has been directly implicated in the hormonal feedback loop that links adipose tissue metabolism, via the hormone leptin, to the satiety centre in the brain (Lee et al., 2007; Wolf 2008). Enhanced secretion of gluOC could therefore help to maintain appetite. How then do we manage nutrition to enhance the capacity of an animal to secrete gluOC, if it indeed proves to be a link to improved glucose and appetite control in cattle? Will short term remedies be sufficient, such as those currently recommended in transition cow programs around the world (Lean et al., 2006). Or is a longer term view necessary, where nutritional strategies to rebuild bone lost in early lactation are applied in mid to late lactation/early to mid-pregnancy. Alternatively, the emphasis
may have to be on an improved skeletal development throughout the calf and heifer rearing stages.

Consequently, attention to bone health could help to reduce the risk of all the above metabolic diseases, so brings us to the hypothesis that healthy bone = healthy body.

CONCLUSION

Furthering our understanding of what constitutes healthy bone and how to monitor and modify it in the dairy cow could prove central to developing nutritional strategies to minimise the risk of the most common metabolic disease in the modern dairy cow: milk fever, ketosis, and fatty liver, and optimise milk production. Understanding the regulation and action of a new hormone produced by bone tissue, gluOC, provides an exciting first step.

REFERENCES


**DISCUSSION**

**Question:**

1. Is your experiment making balance Ca and P?

2. Do you consider the physiological status, especially in young animal more prone to P?

**Answers:**

1. No, only P was considered, but for practical Ca and P should be balanced. Balance Ca and P will be considered in next experiment.

2. Yes, especially for feeding rice bran and palm kernel cake, should consider Ca, Na and Cl.