Proceeding of the NO Laminitis! Conference 2013

Equine Cushing's and Insulin Resistance Group Inc. (ECIR Group Inc.)

Sep. 27 - 29, 2013
Jacksonville, OR, USA

OBESITY AND INFLAMMATION - THE ROLE OF ANTI-INFLAMMATORY MEDIATORS IN THE MANAGEMENT OF EQUINE INSULIN RESISTANCE.

Kathleen M Gustafson, PhD

Once considered only to be a reservoir for energy storage, adipose tissue is now recognized as the largest endocrine organ in the body. As such, adipose tissue plays an important role in whole body homeostasis. The ability to regulate metabolic processes is critical for survival. Thermoregulation, control of body pH, sodium and potassium, cardiac autonomic control, immune modulation and insulin regulation are all homeostatic mechanisms. This presentation deals primarily with insulin regulation. Insulin is involved in a wide range of intracellular pathways via enzyme cascades and coordinates the storage, mobilization and utilization of fuel in adipose tissue, liver and muscle.

Adipose tissue communicates with the rest of the body by synthesizing and releasing a host of secreted molecules, collectively designated as adipokines. These molecules have significant effects on total body glucose and lipid metabolism and insulin sensitivity. Excess adipose tissue results in vagal withdrawal which, in and of itself, results in increased cytokine release, higher heart rate and chronic, low-grade systemic inflammation. When adipocytes (fat cells) reach maximum storage capacity, cellular dysfunction occurs, resulting in release of cytokines, macrophage infiltration, necrosis and cell death. Activated macrophages also secrete cytokines that can impair adipocyte insulin sensitivity, further contributing to systemic insulin resistance (IR) and inflammation.

In humans and other animal models, lipotoxicity is associated with lipid accumulation in non-adipose tissue (liver, pancreas, skeletal muscle, heart) which can lead to cell dysfunction or cell death. There is no direct evidence that lipotoxicity occurs in the horse. However, the horse does demonstrate localized adipose tissue inflammation, particularly in the crest of the neck, which can be a key indicator of IR.

Some studies have shown that increased body condition score is associated with increased insulin, increased leptin and pro-inflammatory cytokine gene expression. (Suagee et al., J Vet Intern Med 2013, Vick et al., Am Soc Animal Sci, 2007) Another study has shown no relationship between IR, obesity and inflammation (Holbrook et al., Vet Immun and Immunopath, 2012) It may be important to differentiate between plasma cytokines (measured in most studies) and tissue cytokines. Burns et al., found the nuchal ligament adipose tissue (crest of the neck) to be the most reactive with respect to inflammatory signaling when compared to visceral fat. These findings also suggest that cytokine concentrations in adipose tissue are much higher than circulating levels, which may explain the inconsistent findings using plasma cytokine levels. (Burns et al., J Vet Intern Med, 2010.) If confirmed, this would indicate that the major effects of these secretory produces are local rather than systemic. There is no evidence to suggest that inflammatory cytokines induce laminitis, however, all homeostatic mechanisms are challenged by the systemic reaction to inflammation.

The role of insulin in homeostasis – IR is often held as insulin's inability to promote normal glucose uptake. However, the physiological role of insulin is more wide-ranging and includes the metabolism of all 3 macronutrients (carbohydrates, fats, and proteins) and a role in maintaining homeostasis. Insulin is a hormone that influences many metabolic pathways and organ systems, including amino acid transport, inhibition of lipolysis, fatty acid synthesis and transport, triglyceride and glycogen synthesis

among many others. Given the complexity of insulin action, attributing the negative effects of IR to "sugar" is vastly oversimplified.

The effect of exercise – Exercise has myriad physiologic benefits. In the acute phase, exercise stimulates the autonomic nervous system (ANS), increasing sympathetic drive to increase heart rate and cardiac output. To maintain autonomic balance, there is increased vagal nerve activity and release of acetylcholine, an important anti-inflammatory molecule. Exercise increases glucose uptake and increases insulin sensitivity. Muscle mass increases with exercise, which demands more energy and glucose. Further, exercise corrects endothelial dysfunction.

In humans, skeletal muscle mass (even "average" levels) relative to body weight is positivity associated with insulin sensitivity: more muscle, more sensitivity to insulin signaling. In horses, even light and moderate activity (but not turnout) improves insulin sensitivity. Exercise increases the expression and moves the glucose transporter GLUT-4 to the cell membrane, facilitating the uptake of glucose. Exercise reduces systemic insulin concentration and can improve adipose tissue insulin sensitivity. Not only is muscle demanding more glucose, so is adipose tissue. When able, exercise is the first line of defense in the management of IR. Exercise is the most natural, holistic and often overlooked therapy that provides greater benefits than diet alone. The combination of a balanced, moderate carbohydrate diet and exercise can produce extraordinary results.

Essential fatty acids (EFAs) – In the natural equine diet, fats and polyunsaturated fatty acids (PUFA) are found primarily in fresh grass. Fats and EFAs are also found in feedstuffs (sunflower, soybean, corn, flax). Once grass is cut and dried to hay, EFAs rapidly decay. EFAs play important roles in cell membrane function and fluidity, immune function and homeostasis. The most abundant EFAs in grass are alpha-linolenic acid (ALA) and linoleic acid (LA). ALA belongs to the omega-3 (n-3) family, while LA belongs to the omega-6 family (n-6). In grass, n-3 ALA is found in higher amounts than n-6 LA, in a ratio of about 4-6 times more ALA than LA. In many species, including the horse, these shorter chain EFAs are elongated through a series of enzymatic reactions to their longer chain forms. In addition, the EFAs synthesize eicosanoids through oxidation. These secondary signaling molecules play roles in a host body functions including inflammation and immunity, and act as cell messengers.

The balance of n-3 to n-6 fatty acids is important when one considers the effect of eicosanoid production. Eicosanoids can be classified as pro-inflammatory or anti-inflammatory. Since both ALA and LA use the same enzyme substrate to synthesize eicosanoids, the balance of pro- vs. anti-inflammatory will favor the EFA in greater abundance. Since the equine natural diet (grass) is 4 - 6 times greater in ALA than LA, we can presume that eicosanoid production is weighted towards the anti-inflammatory side. It is important to understand, however, that EFAs are *essential and n-6 EFAs should not be eliminated from the diet*. Rather, balance is critical; we use the natural equine diet as a guide. Because many IR horses are restricted from grass, supplementation with 4 - 6 oz/day ground flaxseed has been shown to be beneficial. Of all the seeds and grains, flaxseed has the EFA profile most similar to grass.

In humans, n-6 PUFAs in muscle membrane are positively related to IR; the higher n-6's, the higher IR. Conversely, higher n-3 PUFAs in muscle membrane are associated with greater insulin sensitivity (less IR). There are indications from the human literature that the longer chain n-3 EFAs may interact with exercise, as both exercise and long chain n-3 PUFAs have similar effects, for example, lowering heart rate through enhanced vagal function and reducing inflammation, presumably through interactions with acetylcholine. The mechanism behind n-3 PUFA anti-inflammatory effects are becoming clearer with the discovery of a cell receptor, GPR120. When this receptor is activated by n-3 PUFAs, it blocks the inflammatory cascade in pro-inflammatory macrophages.

In summary, equine IR with obesity presents many similarities to other species with respect to obesity and inflammation. Clearly, managing obesity is critical, but not all IR horses are obese – some present with normal body condition scores but show areas of patchy, localized fat. A fatty, inflamed nuchal crest is a fairly consistent finding (as well as a warning sign) and should be considered a site of adipose tissue inflammation and macrophage infiltration. With dietary management of simple sugars and starch,

with care not to exceed the caloric energy demands of the animal, with the proper balance of n-3:n-6 EFAs and above all, *exercise*, the inflammatory profile associated with equine IR can be controlled.

References

Suagee, JK, Corl BA, Crisman MV, Pleasant RS, Thatcher CD, Geor RJ. (2013) Relationships between body condition score and plasma inflammatory cytokines, insulin and lipids in a mixed population of light-breed horses. J Vet Intern Med. 27(1):157-63. PMID: 23216530.

Vick MM, Adams AA, Murphy BA, Sessions DR, Horohov DW, Cook RF, Shelton BJ, Fitzgerald BP. (2007) Relationships among inflammatory cytokines, obesity, and insulin sensitivity in the horse. Am Soc Animal Sci. 85(5): 1144-55. PMID: 17264235.

Holbrook TC, Tipton T, McFarlane D. (2012) Neutrophil and cytokine dysregulation in hyperinsulinemic obese horses. Vet Immun and Immunopath. 145(1-2):283-9. PMID: 22169327.

Burns, TA, Geor RJ, Mudge MC, McCutcheon LJ, Hinchcliff KW, Belknap JK. Proinflammatory cytokine and chemokine gene expression profiles in subcutaneous and visceral adipose tissue depots of insulin-resistant and insulin-sensitive light breed horses. (2010) J Vet Intern Med. 24(4):932-9. PMID: 20649750.