Applied equine nutrition and training

Equine NUtrition and TRAining COnference (ENUTRACO) 2009

edited by:

Arno Lindner

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Editorial

It is a great pleasure to provide this book to you! It honours the engagement of the authors of the articles and the sponsors of ENUTRACO 2009 Fundación para la Promoción del Deporte Ecuestre (Foundation to Promote Equine Sports), Lohmann Animal Health and Lonza and will supply you with a unique blend of actual, scientific but easy to understand and by experts commented information on nutrition and training aspects of horses. I am very grateful!

Wish you pleasant reading!

Arno Lindner

Articles

Gastric ulcers: diet and treatment

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Gastric ulcers are common in performance horses (Bertone, 2000; Johnson *et al*., 1994; McClure *et al*., 1999; Murray, 1989; Murray *et al*., 1989, 1990, 1996, 2001; Rabuffo *et al*., 2002; Sandin *et al*., 2000; Vatistas *et al*., 1999a; Wilson, 1986). Equine Gastric Ulcer Syndrome (EGUS) is the term used to describe ulcers in the terminal esophagus, nonglandular stomach, glandular stomach, and proximal duodenum. Diagnosis of this condition requires a thorough history, interpreting subtle and vague clinical signs, endoscopic examination, and response to treatment. All ages and breeds of horses are susceptible to EGUS and current therapeutic strategies focus on blocking gastric acid secretion and raising stomach pH. Pharmaceutical agents typically target the histamine type-2 receptor (histamine receptor antagonists) or the proton pump (proton pump inhibitors [PPI]) which in turn inhibits parietal cell acid secretion. These pharmaceutical agents, especially the latter, are very effective in increasing stomach pH but are also quite expensive. Thus, a more comprehensive approach to treatment and prevention of EGUS includes determining and correcting of the underlying cause, environmental management, dietary manipulation and pharmacologic intervention. These proceedings focus on a comprehensive approach to diet and treatment in horses with EGUS.

Anatomy and physiology of the equine stomach

The proximal third of the equine stomach is lined with non-glandular stratified squamous epithelium, an extension of the esophagus. The majority (80%) of ulcers are found in this region. The distal two-thirds of the stomach is covered by glandular mucosa, which is responsible for secretion of mucus, hydrochloric acid (HCl), and Pepsinogen (Murray, 1992). Ulcer development in these regions is thought to be an imbalance between protective and aggressive factors (Table 1). The nonglandular squamous mucosa is predisposed to acid injury because it lacks the protective and buffering capacity provided by the

Table 1. Physiologic factors affecting ulcer development.

Aggresive factors	**Protective factors**	
	Non-glandular mucosa	**Glandular mucosa**
Hydrochloric acid secretion	Epithelial restitution	Epithelial restitution
Organic acid production	Intracellular bicarbonate	Bicarbonate-mucus layer
Pepsin conversion from	Mucosal blood flow	Mucosal blood flow pepsinogen
Duodenal reflux of bile acid		Prostaglandin E production

bicarbonate-rich mucus found in the glandular region (Andrews *et al.*, 1999a; Ross *et al.*, 1981). The glandular region, on the other hand, has extensive protective mechanisms, including a bicarbonate-rich mucus layer, an extensive capillary network, and rapid restitution of epithelium when injured. Ulcers in this region are less common and heal rapidly.

Etiology for EGUS in adult horses

Horses are continuous gastric HCl secretors, and acid exposure is thought to be the primary cause of EGUS (Cambell-Thompson and Merritt, 1990). Several acids (HCl, volatile fatty acids (VFAs), and bile acids) have been shown to cause damage to the non-glandular region of the equine stomach (Berschneider *et al.*, 1999; Nadeau *et al.*, 2000, 2003). In a recent report, HCl alone and in combination with VFAs caused inhibition of cellular sodium transport, cell swelling, and eventual ulceration, when exposed to the non-glandular squamous mucosa at pH $\leq$ 4.0. Ulcerogenic effects of the VFAs in combination with HCl were dose dependent and the intensity of damage varied between the VFAs studied (Nadeau *et al.*, 2003). Bile acids were shown to increase the non-glandular mucosal cell permeability to hydrogen ions, which eventually lead to ulceration (Berschneider *et al.*, 1999). However the effect of bile acids in EGUS is questionable because they usually come from less acidic duodenal reflux and are non-ulcerogenic at a pH > 4 (Argenzio, 1999; Berschneider *et al.*, 1999). Pepsinogen, which is cleaved to pepsin at a pH < 4, has a role in the development of EGUS. This proteolytic digestive enzyme may act synergistically with HCl to result in acid damage. While HCl and stomach pH have been incriminated as the main culprits causing EGUS, it is likely that a combination of HCl, organic acids, and pepsin act synergistically to cause EGUS.

Risk factors in adult horses

While acid injury has been implicated in the cause of EGUS, several risk factors for its development have been identified (Table 2) (Andrews *et al.*, 1999a; Murray *et al.*, 1996; Rabuffo *et al.*, 2002).

Exercise intensity

Horses involved in training and racing are at high risk to develop EGUS (Vatistas *et al.*, 1999b). Current prevalence figures show that 60 to 90% of performance horses have EGUS. Recently it has been shown that horses running on a high-speed treadmill have increased abdominal pressure and decreased stomach volume. The authors speculated that contraction of the stomach allowed acid from the glandular mucosa to reflux up into the non-glandular mucosa leading to acid injury (Lorenzo-Figueras and Merritt, 2002). Daily exercise may increase the exposure of the non-glandular mucosa to acid explaining the increased prevalence of gastric ulcers in horses in training. Furthermore, an increase in serum gastrin concentration has been shown to occur in

Table 2. Clinical signs and risk factors of EGUS.

Clinical signs adults	**Foals**	**Risk factors**
Acute colic	Diarrhea	Stress
Recurring colic	Abdominal pain	Transportation
Excessive recumbency	Restlessness	High-grain diet
Poor body condition	Rolling	Stall confinement
Partial anorexia	Lying in dorsal recumbency	Intermittent feeding
Poor appetite	Excessive salivation	Intense exercise
Poor performance/training	Bruxism	Racing
Attitude changes	Intermittent nursing	Illness
Stretching often to urinate	Poor appetite	NSAID use
Inadequate energy		Management changes
Chronic diarrhea		

exercising horses (Furr *et al.*, 1994). This increase in serum gastrin may increase glandular HCl secretion that may lead to acid damage.

Intermittent vs. continuous feeding

Horses grazing at pasture have a decreased prevalence of EGUS. During grazing, there is a continuous flow of saliva and ingesta that buffers stomach acid, keeping stomach pH ≥ 4 for a large portion of the day. On the other hand, when feed is withheld from horses, before racing or in managed feeding stables, gastric pH drops rapidly and the non-glandular mucosa is exposed to an acid environment. Intermittent feeding has been shown to cause and increase the severity of gastric ulcers in horses and this technique has been developed as a model to consistently produce EGUS (Feige *et al.*, 2002; Murray, 1994; Murray and Schusser, 1993). The non-glandular mucosa is the most susceptible to ulceration in horses subjected to intermittent feeding due to its lack of mucosal protective factors.

Diet

Diet has been implicated as a risk factor for EGUS. Serum gastrin concentrations are high in horses fed high concentrate diets. Also, high concentrate diets are high in digestible carbohydrates. Digestible carbohydrates are fermented by resident bacteria, resulting in the production of VFAs, which in the presence of low stomach pH (≤ 4), cause acid damage to the non-glandular squamous mucosa (Nadeau *et al.*, 2003). Furthermore, a recent study in horses fed a high protein and calcium diet (alfalfa hay/grain) showed higher stomach pH than horses fed a low protein and calcium (brome grass hay) diet. The high protein and calcium diet had fewer and less severe gastric ulcers. Thus feeding alfalfa hay may have some protective effect on the non-glandular mucosa in horses.

Transport stress

Transporting horses has been implicated as risk factor for EGUS. Transportation of horses has been associated with dehydration, increased threat of respiratory illness (pleuritis, pleuropneumonia), and immune suppression (Watson, 2002). When horses are being transported, water and feed consumption is usually decreased which may cause an increased incidence of EGUS. Transportation has been shown to increase the severity of gastric ulcers in horses (C.R. Reinemeyer personal communication). However, a recent endoscopic study in western performance Quarter horses subject to frequent travel and intensive training had a lower prevalence (40%) of gastric ulcers than horses in race training, calling into question the effect of these factors on the development of EGUS in western performance horses. The authors' speculated that demeanor played a role in the lower prevalence of ulcers in this breed of horse. Quarter horses are typically calm compared to Thoroughbred racehorses, which may explain the higher prevalence of EGUS (93%) in Thoroughbreds.

Stall confinement

Stall confinement has been implicated as a risk factor for EGUS. Horses that are housed in pastures have a decreased prevalence of gastric ulcers, compared to horses that are housed in stall. The reason for this may be multifactorial, as horses that are stalled may be fed intermittently and housed without exposure to other horses (Feige *et al.*, 2002).

Non-steroidal anti-inflammatory drugs

Non-steroidal anti-inflammatory drugs (NSAIDs), phenylbutazone and flunixin meglumine, have been shown to induce gastric ulcers in horses (MacAlliser and Sangiah, 1993). However, the use of NSAIDs in race horses has not been shown to be a risk factor for EGUS in multiple epidemiologic studies (Johnson *et al.*, 1994; McClure *et al.*, 1999; Murray *et al.*, 1996; Rabuffo *et al.*, 2002; Vatistas *et al.*, 1999a). In one study NSAIDs did cause ulcers in the glandular mucosa and increased the severity of ulcers in the non-glandular squamous mucosa (MacAllister *et al.*, 1992). NSAIDs are thought to cause more severe ulcers in the glandular stomach mucosa because of their effect on prostaglandin inhibition. Prostaglandin inhibition by NSAIDs results in decreased mucosal blood flow, decreased mucus production, and increased HCl secretion. While prostaglandins are also important in the regulation of acid production and sodium transport, it may be their effect on mucosal blood flow that is the most important (Murray, 1991a). Adequate blood flow is necessary to remove the hydrogen ions that diffuse through the mucus layer covering the glandular mucosa.

Gastric mucosal ischemia may lead to a hypoxia induced cellular acidosis, release of oxygen free radicals, phospholipase, and proteases, which may damage the cell membrane leading to necrosis.

Helicobacter *spp.*

While *Helicobacter* spp. is an important cause of ulcers in other species, it has recently been cultured from the manure of healthy horses and the new species has been named *Helicobacter equorum* (Moyaert *et al.*, 2007). Also, Helicobacter-specific DNA was isolated from glandular and non-glandular mucosa of seven horses in another study (Scott *et al.*, 2001). The importance of this discovery is unknown and the role of *Helicobacter* spp. in EGUS remains speculative, in light of other reports in which no organisms were seen in necropsy specimens from the stomach of horses with and without EGUS (Johnson *et al.*, 1994). More studies are necessary to define the role of *Helicobacter equorum* in EGUS, since most ulcer occur in the nonglandular stomach. Helicobacter-related gastric ulcers in people commonly occur in the glandular mucosa. However, horses with chronic recurring gastric ulcers may benefit from antibiotic, probiotic and antacid treatment much like people, because bacteria (*e.g. E. coli*) colonize gastric ulcers in horses and may inhibit healing as in other species.

Clinical signs of EGUS

Clinical signs associated with EGUS are numerous, and often vague. Ulcers are more common in horses showing clinical signs (Table 2) (Andrews *et al.*, 1999a; Murray *et al.*, 1989). In Thoroughbred horses in race training, gastric ulcers were associated with poor performance, poor hair coat, picky eating, and colic (Vatistas *et al.*, 1999a). Of horses with a client complaint of conditions associated with gastric ulcers, or showing subtle signs of poor health, gastric ulcers were identified in 88-92% compared to 37-52% identified in horses not showing clinical signs (Bertone, 2000; Murray *et al.*, 1989; Vatistas *et al.*, 1999a). In addition to an increased prevalence of ulcers in clinically affected horses, the severity of ulceration may be correlated with the severity of the symptoms (Murray *et al.*, 1989; Vatistas *et al.*, 1994, 1999a).

Diagnosis

While an appreciation of the basic anatomy and physiology of acid production is important, it is equally important to be able to identify horses which would benefit from anti-ulcer therapy (Figure 1). Gastroscopy is the only definitive diagnosis of gastric ulcers currently available. The procedure for gastroscopy has been described in detail elsewhere, but requires at least a 2 meter endoscope to visualize the non-glandular mucosa and Margo plicatus and a 2.5 m to 3 m to visualize the pylorus and proximal duodenum in most adult horses (Andrews *et al*., 2002; Murray *et al*., 2001). Once visualized, ulcers should be scored for severity (Andrews *et al*., 1999a). Use of a scoring system allows the clinician to monitoring healing and evaluate efficacy of treatment.

Currently there are no hematologic or biochemical markers to diagnose EGUS. However, O'Connor and Cohen (2002) recently evaluated the potential of a sucrose absorption test to diagnose gastric ulcers. Sucrose is a large carbohydrate broken down in the brush border of the intestine to glucose and fructose. If absorbed by the body, it must be absorbed across damaged gastric mucosa. It is not metabolized by the body, and is excreted in the urine. Concentrations of sucrose rose in the urine of horses with experimentally induced gastric ulcers. Initial evaluation of this test suggested that it holds promise to diagnosis EGUS, without the expense and expertise of gastroscopy, but more research is needed to further refine this technique.

Because of the lack of any additional laboratory diagnosis, in situations where ulcers are strongly suspected, but gastroscopy is not available, it may be worthwhile to start empirical treatment and observe for resolution of clinical signs. If the horse does not respond to treatment, referral to a facility with a gastroscope is indicated.

Treatment of EGUS

The goals of antiulcer therapy are to relieve pain, eliminate clinical signs, promote healing, prevent secondary complications, and prevent recurrence. The mainstay of EGUS treatment is increasing the stomach pH and suppression of stomach HCl acid secretion. Because of the high recurrence rate, effective acid control should be followed

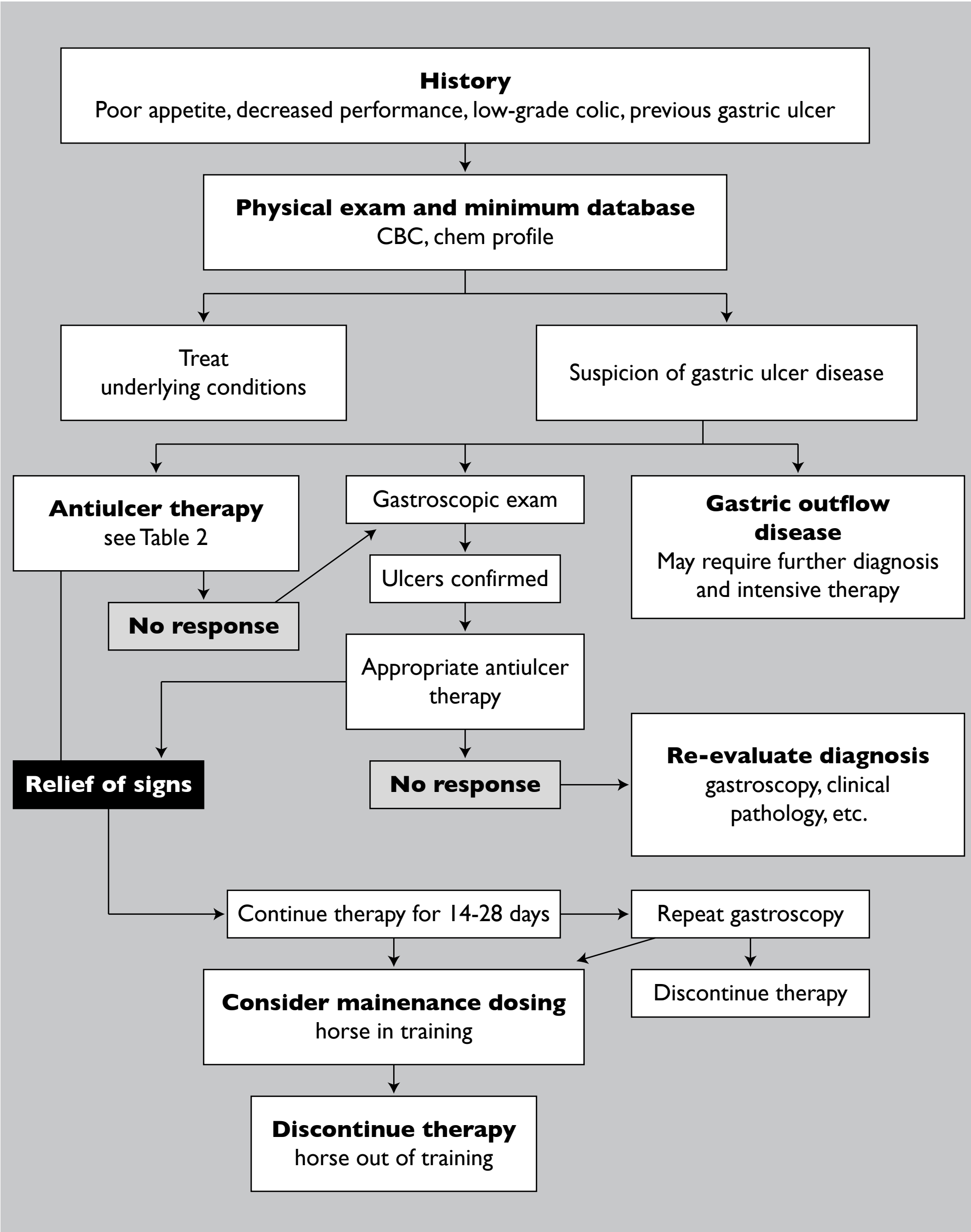

Figure 1. Diagnosis and treatment of EGUS.
Reproduced with permission from Andrews F, Bernard W, Byars D, Cohen N, Divers T, MacAllister C, et al. *Recommendations for the diagnosis and treatment of equine gastric ulcer syndrome (EGUS). Equine Vet Educ 1999; 1 (2): 122-134.*

by altered management strategies and/or long-term treatment to prevent ulcer recurrence.

Pharmacologic therapy

Once EGUS is diagnosed, therapy should be initiated to achieve the above outlined goals. Some ulcers heal spontaneously, but the majority need pharmacologic therapy to achieve healing, especially while horses remain in athletic training (Murray *et al*., 1995; Rabuffo *et al*., 2002). There are many approaches to treating EGUS but the mainstay is acid suppressive therapy, which sets up a permissive environment in the stomach to allow ulcer healing. Many pharmacologic agents are available to treat gastric ulcers in man but few have been shown to be effective in treatment and prevention of EGUS. Of these products, only Gastrogard® is approved by the FDA for treatment and prevention of recurrence of EGUS. However, other therapies have been used to treat EGUS with mixed success and their advantages, disadvantages, and evidence are presented below. Doses for these pharmacologic agents are listed in Table 3.

Table 3. Drug therapy for treatment of EGUS

Drug	Dosage	Route of administration	Dosing interval
Omeprazole	1.0 mg/kg	Intravenously	q 24 h
Omeprazole	4 mg/kg	Orally	q 24 h
Omeprazole (prevention)	1 mg/kg	Orally	q 24 h
Ranitidine	1.5 mg/kg	Intravenously	q 6 h
Ranitidine	6.6 mg/kg	Orally	q 8 h
Famotidine	0.3 mg/kg	Intravenously	q 12 h
Famotidine	2.8 mg/kg	Orally	q 12 h
Misoprostol	5 µg/kg	Orally	q 8 h
Sucralfate	20-40 mg/kg	Orally	q 8 h
AlOH/MgOH antacids	30g AlOH/15 g MgOH	Orally	q 2 h
Bethanechol	0.025-0.30 mg/kg	Subcutaneous	q 3-4 h
Bethanechol	0.3-0.45 mg/kg	Orally	q 6-8 h
Erythromycin lactobionate	0.1-1.0 mg/kg	Intravenously	Undetermined
Cimetidine	not effective for treatment of EGUS		

Acid suppression therapy

Antacids

Antacids neutralize stomach HCl, however aluminum containing antacids may have mucosal protective effects by stimulating prostaglandin production. Their usefulness in controlling stomach pH in the horse is questionable. A liquid mixture containing aluminum hydroxide (30 g) and magnesium hydroxide (15 g) (Maalox TC®, 250 ml, PO) increased stomach pH in horses for 2 hours after administration (Clark *et al.*, 1996). Another study found that magnesium hydroxide (7.2 g) and aluminum hydroxide (8.1 g) had a moderate and short-lived effect on pH (Murray and Grodinsky, 1992). It would appear from this data that every two hour dosing of large volumes of an antacid is needed to treat EGUS. Their prolonged use may interfere with the absorption of electrolytes (e.g. calcium, magnesium, and phosphorus). Thus, antacids such as aluminum hydroxide/magnesium hydroxide may relieve clinical signs of EGUS, but are probably not effective in healing gastric ulcers.

Recently, an equine nutraceutical antacid was developed containing aluminum phosphate, calcium carbonate, dihydroxy-aluminum sodium carbonate, (Neigh-Lox®, Kentucky Equine Research) and is sold as a pelleted feed additive to relieve heartburn in horses (Pagan, 1997).

To the authors' knowledge, there are no published scientific studies proving efficacy of this compound in the treatment or prevention of EGUS.

Histamine Type-2 receptor antagonists

Histamine stimulates acid secretion from the parietal cells (Kitchen *et al.*, 1998). Histamine type-2 receptor antagonists decrease acid secretion by competitively binding to the histamine receptor, thus blocking histamine attachment and stimulation of gastric acid secretion. Additionally, these agents may inhibit acid secretions stimulated by gastrin and acetylcholine (Brunton, 1996). Cimetidine and ranitidine have been used extensively to treat EGUS.

Cimetidine

Cimetidine has been used since the early 1980s to treat and prevent ulcers in horses and foals (Sangiah *et al.*, 1988). Cimetidine (20-25 mg/kg, PO, 6-8 hrs; 6.6 mg/kg, IM/IV, q6h) is currently recommended for treatment of EGUS, although doses vary among clinicians (Andrews *et al.*, 1999a). Cimetidine is less potent than ranitidine, with a variable oral absorption (Sangiah *et al.*, 1988; Smyth *et al.*, 1990). It may inhibit the hepatic microsomal enzyme system and prolong half-life of drugs with high first pass hepatic metabolism (Plumb, 2002). When compared to omeprazole (4 mg/kg and 2 mg/kg PO SID), cimetidine (20 mg/kg PO TID) is not as effective healing EGUS of horses in race training (Nieto *et al.*, 2001). Although cimetidine is effective in treatment of gastric ulcers in man, there is little scientific evidence in the veterinary literature showing that cimetidine has efficacy in the treatment of EGUS.

Ranitidine

Ranitidine hydrochloride (6.6 mg/kg) is 4 times more potent than cimetidine (Plumb, 2002). When given orally (6.6 mg/kg, PO, q8h), it suppresses acid output and maintains a median stomach pH of 4.6 (Murray and Schusser, 1993). At a dose of 6.6 mg/kg given orally every 8 hours, ranitidine is able to successfully limit ulcer development in a feed deprivation model (Murray and Eichorn, 1996). Lower doses (4.4 mg/kg, PO, q8h) given orally are ineffective for treatment of EGUS (Holland *et al.*, 1997; Johnson *et al.*, 2001). The recent availability of the generic ranitidine has made this drug popular and effective in treating EGUS (MacAllister and Sangiah, 1993). Ranitidine (6.6 mg/kg, orally, q8h) has efficacy and is recommended for treatment of EGUS, but owner compliance is difficult.

While ranitidine and cimetidine have been the most studied, other H2 antagonists have been evaluated experimentally and may allow for less frequent dosing and more effective acid suppression (MacAllister *et al.*, 1992; Orsini and Spencer, 2001; Orsini *et al.*, 1991). Bioavailability and pharmacodynamic studies with famotidine (2.8 mg/kg, PO, q12h; 0.3 mg/kg, iv, q12h) in horses suggest that it can be used for treatment of EGUS, but may be cost prohibitive (McClure *et al.*, 1999).

Proton Pump Inhibitors (PPIs)

The use of proton pump inhibitors offers many advantages over H2 antagonists. Once daily administration and ability to block gastric acid secretion regardless of stimulus is the advantage of these drugs for treatment of EGUS.

Omeprazole

Omeprazole, a substituted benzimidazole, is the only approved drug for the treatment of EGUS. Omeprazole oral paste (4 mg/kg, PO, q24h) inhibits gastric acid secretion for 24 hours in horses (Daurio *et al.*, 1999). In an acid environment omeprazole is activated to a sulphenamide derivative and binds reversibly to the $H+/K+$ ATPase in parietal cells and inhibits transport of hydrogen ions into the stomach. Because of its effect on the cell, omeprazole is often called a proton-pump inhibitor. The effect on gastric acid secretion is dose and time dependent. Omeprazole in a gel suspension suppressed basal acid secretion by 83.7% 5 hours after a single dose of 1.5 mg/kg. After 5 days of once daily dosing at 1.5 mg/kg, acid secretion was suppressed 93% (Andrews *et al.*, 1995). Omeprazole is metabolized in the liver and excreted in urine and bile, and significant liver disease may affect the metabolism of the drug. Long-term administration in dogs, did not cause any clinical, hematological, or biochemical alterations, but did cause a reversible gastric mucosal hypertrophy (Sundell and Nillsson, 1986). Long-term administration of high doses causes hyperplasia of ECL cells and gastric carcinoid tumors in rats (Tielemans *et al.*, 1989). No significant side effects have been reported in horses treated for up to 90 days.

Omeprazole has been shown to be an effective treatment for EGUS at a dose of 4 mg/kg orally once daily (Andrews *et al.*, 1995, 1996b; MacAllister *et al.*, 1999; Murray *et al.*, 1997). A recent study of 565 horses in race training found that 96% of the 147 horses being administered H2 antagonists had gastric ulcers, with 61% considered to be severely affected (Nieto *et al.*, 2002). Of the horses not receiving H2 antagonists, 88% had gastric ulcers, with 58% considered severe. All of the horses in the study were put on a 28-day course of omeprazole. There was a statistical improvement in performance, weight gain, attitude, appetite, and appearance after treatment. Endoscopically 65% of the horses with gastric ulcers that were treated were healed and 94% were improved. The primary reason for the failure of treatment with H2 antagonists was owner compliance and incorrect dosing. A second study found that 99% of spontaneous ulcers in adult horses and foals over 4 weeks of age were improved with 86.7% healed with omeprazole treatment (MacAllister *et al.*, 1999). Effectiveness of omeprazole was also shown to increase the rate of healing in horses with ulcers removed from race training (Murray *et al.*, 1997).

Coating or binding agents

Sucralfate and bismuth subsalicylate are two compounds that bind to stomach ulcers and promote healing. Sucralfate is a hydroxyl aluminum salt of sucrose octasulfate and binds to the negatively charged particles in the ulcer bed, buffering HCl by increasing bicarbonate secretion, stimulating prostaglandins production, and adhering to the ulcer bed (Borne and MacAllister, 1993). In the stomach, sucralfate is converted to a sticky amorphous mass, thought to prevent diffusion of hydrogen into the ulcer. In a clinical trial in horses, sucralfate (22 mg/kg, PO, q8h) did not improve subclinical ulcer healing in 6 and 7 month old foals. In a rat model sucralfate successfully prevented gastric ulceration in a dose dependent manner after an ischemia reperfusion injury (Wada *et al.*, 1997). Recently, GastrafateTM (20-30 ml, PO, q12h; MFP, Ltd, Sterling CT), an emulsion (10% Sucralfate, Calcium Carbonate, DASC, Magnesium Hydroxide, Carboxylate, Apple Flavor, Methyl & Propyl Parabens, and Water) has been advertised as a treatment for EGUS. This quantity of Gastrafate would provide 3 g of sucralfate to a patient, which is significantly less than the 9-18 g currently recommended for treatment of EGUS. Currently, there is no published scientific data on the efficacy of this product in treating EGUS. Therefore, sucralfate alone may not be beneficial in treatment of EGUS, but can be used in conjunction with acid suppressive therapy and may be more suited for treatment of Right Dorsal Colitis (colonic ulcers) at a dose of 22 mg/kg, PO, q6-8h.

Bismuth containing compounds may have a coating effect similar to sucralfate. Additionally it will inhibit the activation of pepsin and increase mucosal secretion (MacAllister, 1995). A compound containing 26.25 g of bismuth failed to raise the pH in 5 horses (Clark *et al.*, 1996). Bismuth subsalicylate may be converted to sodium subsalicylate in the gastrointestinal tract, which may cause gastric irritation. Also, salicylates, similar to aspirin, decrease prostaglandin secretion and may further compromise an already damaged mucosa (Martindale, 1982). Thus, compounds containing bismuth are not recommended for treatment of EGUS. However, bismuth is used as part of the therapy in humans with *Helicobacter pylori* induced gastric ulcers and may be used in horses with chronic recurring gastric ulcers in which Helicobacter is suspected.

Synthetic hormones

Misoprostol, a synthetic PGE1 analogue, is effective in the treatment of gastric and duodenal ulcers in man. Acid suppression, increased mucosal blood flow, increased bicarbonate secretion, and increased mucosal restitution are mechanisms of misoprostol. In one study, misoprostol (5 μg/kg, PO) increased stomach pH and inhibited gastric acid secretion for 8 hours. Misoprostol is contraindicated in pregnant and nursing horses due to its effect on increasing uterine contraptions. Although no signs of side effects have been reported in horses, side effects reported in other species include: diarrhea, cramping, flatulence, and uterine contraction. Side effects are dose dependent (Plumb, 2002).

A somatostatin analogue, octreotide acetate, has also been evaluated in horses (Sojka *et al.*, 1992). Octreotide (0.5 to 5.0 μg/kg) raised the gastric pH > 4 for approximately 5 hours, with no adverse effects noted. While octreotide appears to be very safe in human patients, it requires multiple daily dosing and is cost-prohibitive in horses. The benefit of using a somatostatin analogue is the prevention of hypergastrinemia associated with long-term use of acid suppressive drugs. The hypergastrinemia has a positive tropic effect on gastric cells and may result in proliferation (Tielemans *et al.*, 1989). Because somatostatin inhibits gastrin secretion, this hypertrophy is avoided, however no case of gastric hypertrophy has been reported in horses following long term use of acid suppressive drug therapy.

Prokinetic agents

Prokinetics agents may be valuable as an adjunct therapy in the treatment of EGUS and when there is adynamic ileus and gastroduodenal reflux. Bethanechol (0.25 mg/kg, IV) and erythromycin lactobionate

(0.1 and 1.0 mg/kg, IV) increased solid phase gastric emptying time in horses (Ringger *et al.*, 1996). No adverse effects were seen in healthy patients, however other forms of erythromycin can cause fatal colitis in adult horses at antimicrobial doses. Both prokinetics increase gastric emptying versus saline, but bethanechol appeared to be superior. It increased gastric emptying rate of solid food versus erythromycin and increased gastric emptying rate of liquid versus saline. Bethanechol is a synthetic muscarinic cholinergic agent that is not degraded by acetylcholinesterases. The only side effect of the bethanechol administration was increased salivation. Other authors have recommended a dose of 0.025-0.030 mg/kg subcutaneously every 3-4 hours followed by oral maintenance therapy of 0.3-0.45 mg/kg 3-4 time daily (Murray, 1991b). It is also possible that gastroduodenal reflux may worsen after treatment in patients with a proximal small intestinal obstruction (Ringger *et al.*, 1996).

*Antibiotics (*Helicobacter *spp.)*

Treatment of *Helicobacter pylori* infection in humans greatly accelerates ulcer healing, and eradication reduces the risk of reoccurrence. In people combination therapy is the treatment of choice and includes acid suppressive therapy (PPIs or H2 antagonists) and antibiotics. Several examples include: (1) omeprazole or ranitidine, clarithromycin, and amoxicillin or metronidazole, (2) omeprazole or ranitidine, bismuth, tetracycline, and metronidazole, (3) omeprazole or ranitidine, bismuth, tetracycline, and furazolidone. A 10-14 days treatment period is successful in 65% to 80% of clinical cases. This is less than the 95-99% seen in clinical trials, which is likely due to poor patient compliance. These same combination therapies have been proposed for treating dogs and cats with Helicobacter infections. Extrapolating from other species, an oral combination therapy in horses with chronic active, non-healing ulcers (caused by a *Helicobacter* species), would be omeprazole (4 mg/kg q24h), metronidazole (15 mg/kg q6h) and/or trimethoprim/sulfa (25 mg/kg q12h), and bismuth subsalicylate (3.8 mg/kg q6h). An initial 14-day treatment period could be prescribed, which should be followed by gastroscopy. Omeprazole therapy should be continued for the full 28 day if needed.

Other treatments

Other treatments including, furosemide, fenbendazole, G.U.T, and Nutrient Buffer® have been suggested for treatment of EGUS, but efficacy has not been reported in scientific literature. Furosemide showed a strong correlation to reduced severity of ulcers in Thoroughbred horses in race training (Pagan, 1997). However, this same correlation has not been found in any other studies. It was theorized that furosemide might modify gastric mucosal blood flow.

Fenbendazole (6 g, PO, q24h for 5 days), because of its chemical similarity to omeprazole, has been suggested to be an effective treatment of EGUS. However, in one study gastric ulcer scores were significantly improved after 2 weeks of fenbendazole treatment, but after 4 weeks of treatment gastric ulcer scores were no different than controls (MacAllister, 1995). Thus, fenbendazole is not effective in treatment of EGUS.

Other nutraceuticals such as G.U.T. and Nutrient Buffer® claim to be useful in horses with EGUS but no data has been published to show that these compounds have any effect in horses.

Duration of treatment

It is difficult to predict how long a gastric ulcer will take to heal, so treatment duration must be tailored for the individual horse. In a feed deprivation model of ulcer induction, ulcers were healed or almost healed in the horses after 9 days of pasture turnout (Murray, 1994). Omeprazole treatment in Thoroughbred horses in training resulted in healing in 57%, 67%, and 77% of horses treated for 14 days, 21 days, and 28 days, respectively (Andrews *et al.*, 1999b). Horses with spontaneous occurring ulcers in a field trial treated with omeprazole showed 86% healing after 28 days of treatment (MacAllister *et al.*, 1999). We recommend endoscopic examination after 14 days of omeprazole therapy to determine if the ulcers are healed. If the gastric ulcers are healed then the horse can be put on omeprazole (2 mg/kg, PO, q24h) to prevent recurrence of ulcers while the horse is in training. If the ulcers are still present then the full 28-day course of omeprazole should be followed and the horse further evaluated after that time. When endoscopy is not available, horses should be treated for at least 28 days. It should be noted that clinical signs might resolve before complete healing has taken place. Signs of poor appetite, colic or diarrhea will usually resolve soon after initiating treatment, and the horse is expected to make improvements in body condition and attitude within two to three weeks. H2 antagonist therapy should be continued for at least 14-21 days, but healing has taken over 40 days in some studies.

In general it may take longer to treat large ulcers, more severe ulcers, and ulcers in the non-glandular mucosa (MacAllister, 1995). In cases where clinical signs have resolved and the risk factors for ulcer development are low, spontaneous healing of ulcers may occur without further treatment. However, spontaneous healing will not occur in horses that continue intensive training, and ulcers may re-occur in those successfully treated if therapy is discontinued. If clinical signs attributed to EGUS have not resolved after 48 hours of treatment, the diagnosis or therapy should be re-considered.

Dietary management

In conjunction with pharmacological therapy, environmental and dietary management may be helpful to facilitate ulcer healing. In man, ingestion of a high protein meal is a stimulus for the release of gastrin and subsequently gastric acid (Merki *et al.*, 1991). In the horse, the ingestion of a grain meal resulted in a higher gastrin stimulus than grass hay. Horses fed hay versus withholding feed had similar acid output, but higher gastric pH. It was theorized that the salivary bicarbonate and the buffering effect of the hay were responsible for the higher pH (Smyth *et al.*, 1988). Providing constant access to alfalfa or good quality hay will also help to raise the pH (Nadeau *et al.*, 2000). In addition to providing constant access to feed to buffer gastric acidity, modifying the diet may help prevent ulcers. Although the VFA concentrations were higher, an alfalfa/grain diet had a higher pH and less ulcers than a plain bromegrass hay diet (Berschneider *et al.*, 1999; Nadeau *et al.*, 2003). In that study no gastric hormones were measured, and it was hypothesized that the calcium could have a direct effect on gastric secretions or the protein was acting as a buffer for the pH. In a rat model a diet of 2% calcium inhibited basal gastric secretion, but not secretions in response to histamine stimulation (Fisher *et al.*, 1990).

Pasture turnout is the best dietary method of controlling gastric ulcers. One study found that simply putting a horse in a stall with *ad lib* grass hay created ulcers, while horses maintained on pasture rarely have gastric ulcers. Management of the diet fed to stalled horses can be modified to decrease the risk of ulcer. Current dietary recommendations include providing continuous feeding of good quality grass and/or alfalfa hay. Sweet feed should be kept to a minimum and grains like barley or oats can be substituted to decrease its fermentation to VFAs.

Prevention

Treatment strategy may vary on an individual basis. While acid suppression is the cornerstone of therapy, preventative therapy must be continued unless risk factors are eliminated. Because of difference in training/management requirements, several courses of therapy may be pursued.

Pharmacotherapy

Omeprazole and the H2 antagonists can be used at their standard doses in hopes of preventing ulcers. There are no published studies evaluating the preventative effects of H2 antagonists, but omeprazole at 2 mg/kg has been shown to prevent ulcer recurrence after treatment in horses in race training (Andrews *et al*., 1999b).

Management and feeding strategies

Different combinations of preventative and therapeutic treatments may be employed in differing situations (Table 4).

Race training

There is limited opportunity to modify the management of horses that are in race training. Due to the high prevalence of EGUS in this population, preventative therapy is strongly encouraged. Horses treated successfully will have reoccurrence of ulcers if appropriate therapy is not instituted. The best strategy for managing horses in race training includes feeding free choice alfalfa, providing Omeprazole (4 mg/kg, PO, q24h, for 28 days) for treatment and Omeprazole (2 mg/kg, PO, q24h) for prevention of recurrence until the horse is removed from race training. A second option would be to feed alfalfa

Table 4. Treatment/preventative strategies.

Strategy	Exercise regimen	Grass hay vs Alfalfa hay	Husbandry	Intermittent vs free choice feeding	Pharmacotherapy
Strategy 1	Race training/ intensive training	Alfalfa	Stall confinement	Free Choice	Gastroguard 4mg/kg 28 days then 2/mg/kg maintenance
Strategy 2	Race training/ intensive training	Alfalfa	Stall confinement	Free Choice	Ranitidine 6.6 mg/kg PO TID for treatment and maintenance
Strategy 3	Race training/ intensive training	Alfalfa	Stall confinement	Free Choice	Antacid 30/15 g Al/Mg before exercise and in evening
Strategy 4	Moderate training, Show/performance horses	Alfalfa	Stall confinement with limited pasture turnout	Free choice when in stall	Same as 1 & 2, or treat with omeprazole or ranitidine 3 days before and throughout show
Strategy 5	Brood mare, trail horse, pleasure horse	Either	Maintained on pasture	On pasture	Not necessary except when diagnosed with ulcers. Treat for 14 days and reevaluate ulcers

free choice and treat with ranitidine (6.6 mg/kg, PO, q8h) until the ulcers are healed (via gastroscopy) and then maintain on Omeprazole, at 2.2 mg/kg orally once daily. A final option to consider would be to treat the horses with 30 g AlOH and 15 g MgOH (approximately 250 mls Maalox TC, PO) after the horse has finished its evening meal and within two hours of an intensive work. The horses could be kept on Omeprazole, (2 mg/kg, PO, q24h) during this time (Table 4).

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Practical and controversial news on the relation between nutrition and sports performance: 2007 to now – part 1

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The topic 'horse nutrition' is scientifically framed by the view on the impact of dietary carbohydrates on the endocrine adjustment of the metabolism (Bailey *et al.*, 2007; Vervuert *et al.*, 2009b). Indeed, the laminitis-insulin resistance complex is still a hot spot. But traditional aspects like electrolyte supply (Waller *et al.*, 2009) or growth (Donabedian *et al.*, 2008; Morel *et al.*, 2007; Vervuert *et al.*, 2007b) and performance show their actuality as well. Finally the increasing utilisation of herbs and spices (Hsia *et al.*, 2007; Williams and Lamprecht, 2008) and the interaction of nutrition and behaviour are challenging areas of feeding practice and research (Brokner *et al.*, 2008; Hothersall and Nicol, 2009; Kruger and Flauger, 2008; Scaramuzzi and Martin, 2008).

In some cases we received very detailed information at molecular level (Arkill and Winlove, 2008; Faleiros *et al.*, 2009; Hodavance *et al.*, 2007; Lepeshkevich and Dzhagarov, 2009; Ramery *et al.*, 2008; Zeng *et al.*, 2008) by recent methodological developments.

Dietary carbohydrate profile

Rapid fermentable carbohydrates have been suspected to be strong triggers for laminitis. The differentiated feed analysis and moreover the models based on dietary oligosaccharides expanded enormously the insight in the pathophysiology of laminitis (Bailey *et al.*, 2002, 2003; Kalck *et al.*, 2009; Longland and Byrd, 2006; Van Eps and Pollitt, 2006). Acidic malfermentation in the hindgut is the key element in these models and focusses the view on the carbohydrate-intestinal microbiota-axis. It has been shown that the diet determines the microbial community in any part of the gastro intestinal tract (De Fombelle *et al.*, 2003). But regardless of the fact that a cecal acidosis affects equine foot like rumen acidosis in dairy cows affects claw health (Danscher *et al.*, 2008; Lean *et al.*, 2008; O'Grady *et al.*, 2008; Plaizier *et al.*, 2008; Westwood *et al.*, 2003) a major part of the laminitis drama obviously is written by the endocrine control of energy metabolism with insulin as the most prominent part but having an endocrine active adipose tissue with its cytokines in the back (Hoffman *et al.*, 2003; Kronfeld *et al.*, 2006; Treiber *et al.*, 2005, 2006a,b). Insulin itself has a laminitis inducing potential and the insulin resistance of tissues which intensively metabolise glucose predispose to laminitis; inflammatory responses from external as well as internal challenges are able to lower insulin sensitivity within short time (Vick *et al.*, 2008; Toth *et al.*, 2008). Obesity as a disease of civilisation in humans, pets and equids is the most risky condition to induce insulin resistance (Adams *et al.*, 2009; Carter *et al.*, 2009a,b; Firshman and Valberg, 2007; Vick *et al.*, 2007). More detailed knowledge is required to organise an optimal procedure of caloric restriction (Van Weyenberg *et al.*, 2008) and exercise to induce adipose tissue catabolism and insulin sensitivity enhancement. The knowledge of glycemic and insulinemic responses on dietary carbohydrates is essential to govern control of energy metabolism (Vervuert *et al.*, 2008a,b, 2009b). In addition, the quality of the response on starch intake by horses claims for a more strict limitation of starch intake at 0.1 g starch per kg body weight per meal, quite lower as derived from the starch impact on hindgut fermentation. Recognising the need for high energy intake of high performance horses, the carbohydrate profile of the diet needs more precise differentiation and guidelines. The manipulation of the response in foals of the diet in dependence on the nutrition of the mare during gestation shows that a programming can be activated by a 'cafeteria-style' of horse diet which may compromise the health status of a population (George *et al.*, 2009; Ousey *et al.*, 2008).

Energy and nutrient supply to the performance horse

The NRC updated the requirement data (NRC, 2007), the German and French committees are busy to finalise the revisions of their requirement and recommendation booklets. It is evident that there is still a discrepancy between the knowledge on the biochemical network of energetics on one side and the evaluation of exercise intensity as well as the energy requirement on the other side (Martin-Rosset, 2008). Work on movement of mass and trunk deformation may help to improve the description of exercise from physical standpoint (Nauwelaerts and Clayton, 2009; Nauwelaerts *et al.*, 2009) and heart rate may act as an indicator for the effort (Coenen, 2008); an intelligent system combines current theoretical models to an exercise assessment system (Ellis, 2008). But simple key facts are not sufficiently confirmed:

- the conversion of metabolisable energy into mechanical work;
- the energy equivalent for lactate accumulation during exercise;
- exercise associated energy consumption after exercise;
- change in basal metabolic rate by training.

Regardless of the limitations to model energy requirements for exercise there is a deficit in the evaluation of the feeding value of single feeds, compound feed as well as total rations. Although we have models to estimate digestible, metabolisable or net energy (Martin-Rosset, 2005; Martin-Rosset *et al.*, 1994, 2006; Zeyner and Kienzle, 2002) based on proximate analysis we have differences in glucose supply to the system, that means differences in digestibility due to the processing of feed (Vervuert *et al.*, 2007a, 2008). Moreover, taking into account the ration dependent variation in hindgut fermentation and the differences in the profit of volatile fatty acids in terms of gluconeogenesis, it is beneficial to substitute the digestible energy system for evaluating the feed by metabolisable energy. This is also the most convenient level to calculate requirements because oxygen consumption as the basic information about energy expenditure represents after suitable conversion metabolisable energy.

Regarding the performance horse energy is the dominating issue but it is a misunderstanding to exclude protein. The elevated protein turnover e.g. due to increased mitochondrial mass or needs in amino acids for lactate management indicates a change in protein requirement. It is questionable to use growth data (protein content in daily gain) to reflect the protein metabolism of performance horses. The utilisation of amino acids during exercise is confirmed for equine (Matsui *et al.*, 2006). This as well as the benefits of combined carbohydrate and protein supplementation for performance in humans (Betts *et al.*, 2007; Etheridge *et al.*, 2008; Koopman *et al.*, 2004) require an intensified discussion on amino acid intake in performance horses. Exciting work on the mammalian target of rapamycin (mTOR) show the deep impact of exercise plus amino acids on muscle cell behaviour (Dreyer et al., 2008). The fact that in practice protein intake overrides common recommendation may induce the conclusion that this is simply a scientific playground. But in fact it is essential to have a precise view on amino acid turnover at first (Ball *et al.*, 2007) and to exchange the traditional statement exercise needs no protein. In addition, the function of mTOR shows a significant role of specific amino acids. Therefore it seems beneficial to develop a promising system to evaluate protein in feed by estimating the precaecally digestible protein and amino acids respectively. The ideal protein concept introduced for pigs is a promising model for the horse too.

The electrolyte balance of performance horses is ruled by the amount of cutaneous losses which can be >60 g sodium and >120 g chloride in the case of >20 litres of sweat. Doubtless improper electrolyte and water supply limits performance (Harris, 2009). The electrolyte supplementation is an area of conflict as in practice supplementation of such amounts is not routinely implemented. The improved rehydration as well as the elevated glycogen resynthesis (Waller *et al.*, 2009) declare the need for a compensation of cutaneous losses and for a specific procedure to distribute electrolyte intake in relation to time of exercise.

The electrolyte supply needs to be organised as a part of the total mineral intake considering the link to acid base balance. It is common to have a calcium intake in performance horses clearly above requirements (Vervuert and Coenen, 2008). High calcium may elevate renal excretion of other minerals and high calcium diets may elevate gastrin secretion in equines like in other species having an impact on gastric functions. On the other hand there are some indices for an exercise induced increase in calcium requirement and the actual recommendation includes a calcium supply above maintenance level for exercising horses (NRC, 2007) although this recommendation is insufficiently confirmed by experimental data. However, the long lasting modification in mineral intake may leave foot prints. The mineral status of the hair, particularly the calcium concentration was found to correspond significantly with cardiac function (Suzuki *et al.*, 2007). Again this shows the demand to revise mineral recommendations for performance horses. The traditional factorial approach to derive requirements seems to be incomplete.

Referring to the performance horse, the work on trace elements shows some unexpected outcome and leaves some questions unanswered. The degree of cartilage irregularities did not correspond to copper status of mare or foal (Gee *et al.*, 2007). But in practice an enforced intake of copper, iron and selenium is highly welcomed. In contrast, the base to recommend general elevated trace element supply to performance horses is weak and neglects possible side effect. The liver is in charge of the copper and iron management but plays a role in the oxydative stress too. The high performance horse is well know to be heavily impacted by oxydative stress (Kirschvink *et al.*, 2008). Considering a temporary loss of the efficiency of the intestinal barrier during and after exercise as shown in humans (a loss in tissue specific defence potential has been shown in horse for its lung) a temporary elevated uptake of copper and particularly iron could occur and present an additional oxidative principle. A further weak position is presented for selenium which is widely used in combination with vitamin E. There is poor response of systemic selenium and glutathione status on elevated selenium intake but a response of keratinised tissue; selenium competes with sulphur during the keratin formation. The depression of sulphur inclusion results in lowered formation of disulfide bounds. This may impair physical properties of horn. There is a lack of work on those aspects.

Feed additives

Not the least because of the large use of concentrates there is an interest to improve gut health as well as metabolic functions by several feed additives. This stimulates the search for successful candidates for probiotics (Laukova *et al.*, 2008). The discrepancies in the results regarding the effects and efficacy of probiotics show that there is no strong evidence for a benefit of probiotic utilisation in horses (Jouany *et al.*, 2008; Köpke, 2009; Laukova *et al.*, 2008; Mackenthun, 2009; Respondek *et al.*, 2008; Swyers *et al.*, 2008; Van Loo, 2007; Xu *et al.*, 2008). Benefits of methyl sulphonyl methan supplementation to prevent oxydative stress (Maranon *et al.*, 2008) or of chromium supplementation to improve insulin responses in insulin resistant ponies (Vervuert *et al.*, 2009a) indicate the detailed search for practical solutions but also the small borderline between feed additive utilisation and drug administration.

The intensity of supplying herbs and spices in equine nutrition is in contrast to the volume of scientifically elaborated knowledge about their effects and efficacy. Fortunately since some years precise studies have been initiated (Colas *et al.*, 2008; Hsia *et al.*, 2007; Pearson *et al.*, 2007b; Williams and Lamprecht, 2008). Partly we receive from *in vitro* work a confirmation about the potential to activate proteins which are involved in xenobiotic management or modify inflammatory responses (Hellum and Nilsen, 2008; Pearson *et al.*, 2007). Promising work *in vivo* shows a beneficial modulation of respiratory dysfunction in horses by herbal products (Pearson *et al.*, 2007a).

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Practical and controversial news on the relation between nutrition and sports performance: 2007 to now – part 2

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In this presentation I will first mention a recent position statement on the relationship between nutrition and athletic performance in humans, and then discuss in more detail two studies examining the effects of nutrition during recovery on muscle glycogen resynthesis in horses, a subject that remains of interest to researchers, nutritionists and trainers given the relationship between muscle glycogen content and athletic performance.

Rodriguez NR, Di Marco NM, Langley S (2009) Joint Position Statement: American College of Sports Medicine, American Dietetic Association, and Dietitians of Canada. Nutrition and Athletic Performance. Med & Sci Sports & Exerc 41:709-731.

Objectives

To employ evidence-based analysis of peer-reviewed literature to address the following topic-specific questions:

- Energy balance and body composition – What is the relationship between energy balance, body composition, and/or weight management and athletic performance?
- Training – What is the evidence to support a particular meal timing, caloric intake, and macronutrient intake for optimal athletic performance during training?
- Competition – What is the evidence to support a particular meal timing, caloric intake, and macronutrient intake for optimal

athletic performance during competition during the 24 hours prior to competition? What is the evidence to support a particular meal timing, caloric intake, and macronutrient intake for optimal athletic performance during competition?

- Recovery – What is the evidence to support a particular meal timing, caloric intake, and macronutrient intake for optimal athletic performance during recovery?

Practical interest

I chose to mention this paper for two reasons: first, the questions posed by the authors are highly relevant to equine sports nutrition, and second, it serves to highlight one of the difficulties that we, as equine nutritionists and veterinarians, face when making recommendations for the feeding management of athletic horses – namely, the lack of evidence-based information concerning optimal nutrition for equine sports performance. The recommendations set forth in this position statement are based on a rigorous, systematic, evidence-based analysis of the nutrition and sports performance literature. Contrast this situation with that in the equine field where all too often recommendations are based on little to no data or, worse, extrapolated from the results of studies in other species. This problem is particularly prevalent in the marketing of nutritional ergogenic (performance-enhancing) supplements for horses. In this context, I draw your attention to the following quote from the position statement: 'The majority of (nutritional) ergogenic aids on the market do not perform as claimed'. The exceptions were 1) creatine (effective in repeated short bursts of high-intensity activity such as sprinting and weight lifting), 2) caffeine (obviously illegal in horses!), and 3) sports drinks and gels that help maintain hydration and fuel supply during endurance exercise.

Waller AP, Heigenhauser GJF, Geor RJ, Spriet LL and Lindinger MI (2009) Fluid and electrolyte supplementation after prolonged moderate-intensity exercise enhances muscle glycogen resynthesis in Standardbred horses. J Appl Physiol 106:91-100.

Objective

To determine the effect of administering a hypotonic electrolyte solution immediately after prolonged exercise, and before feed is provided, on rehydration and muscle glycogen and electrolyte recovery in horses. It was hypothesized that provision of a balanced electrolyte solution, immediately followed by a typical hay and grain meal, after glycogen-depleting exercise, will result in a faster rate of muscle glycogen synthesis than a grain/hay meal with voluntary access to water alone.

Background

Glycogen stored within muscle is an important fuel source during exercise. In horses, previous studies have shown that low muscle glycogen content prior to exercise results in decreased athletic performance during moderate and high-intensity exercise. Furthermore, postexercise muscle glycogen synthesis appears to be quite slow in the horse, with as much as 72 h required for complete replenishment of glycogen stores. Inadequate muscle glycogen replenishment could adversely affect the performance of horses participating in several events in one day or on consecutive days. Therefore, there is interest in the development of nutritional strategies that optimize muscle glycogen synthesis in horses after exercise.

Overview of the study

Six moderately fit Standardbred geldings were used in a crossover design in which each horse performed a Competition Exercise Test (CET) twice in randomized order, an electrolyte treatment and control. The basic diet of the horses was 6 kg mixed hay and 4 kg oats daily. The CET, designed to simulate the second day (speed and endurance test) of a one-star CCI 3-day event, consisted of the following exercise phases on a treadmill: 10-min walk (1.7 m/s), 10-min trot (3.7 m/s), 2-min gallop (10 m/s), 20-min trot (3.7 m/s), 10-min walk (1.7 m/s), 8-min canter (8.0 m/s), and 10-min walk (1.7 m/s). Ten minutes after completion of the CET, the horse either 1) received by nasogastric tube a commercially available electrolyte solution designed to replace an 8-liter sweat loss (12 g Na, 24 g Cl, 9 g K, 1 g Ca and Mg), given in 8 liters of water (osmolality = 212 mosmol/kgH_2O), or 2) stood in stocks for an equivalent amount of time. Muscle biopsy samples (*gluteus medius* m.) were collected before exercise, 15 min after cessation of exercise (0 h), and at 4 h and 24 h post-treatment. In both treatments, horses were given 2 kg hay and 2 kg oats at 20 min after completion of the CET, along with free choice access to water. Horses were given a further 2 kg oats and 2 kg hay at 6 h of recovery, and 2 kg of hay at 12 h of recovery. Muscle samples were analyzed for glycogen and a number of other metabolites as well as electrolyte contents.

Main findings

The CET resulted in decreased total body water and muscle glycogen concentration of 8.4±0.3 liters and 22.6%, respectively, in the control treatment, and 8.2±0.4 liters and 21.9% in the electrolyte treatment. When compared to the control trial, electrolyte treatment resulted in faster restoration of hydration, evidenced by faster recovery of plasma protein concentration, maintenance of plasma osmolality and greater muscle intracellular fluid volume. Additionally, electrolyte treatment enhanced the rate of muscle glycogen resynthesis; at 4 h of recovery muscle glycogen was not different from pre-exercise in this group and was significantly higher than in control at 4 h and 24 h of recovery. Muscle electrolyte (Na, K, Cl and Mg) contents did not differ between treatments. Total water intake, including the 8 liters given via nasogastric intubation, was higher in the electrolyte treatment; at 24 h of recovery total water consumption in the electrolyte and control treatments was, respectively, 49.5±4.2 and 34.8±2.2 liters.

Practical interest

This study examined a novel approach to the post-exercise recovery of muscle glycogen content in horses. Previous studies (e.g. Davie *et al*., 1994; Lacombe *et al*., 2004) have focused on the provision of glucose polymers or starch-rich meals during the recovery period after glycogen-depleting exercise, with the main finding of minimal impact of these nutritional treatments on rate of muscle glycogen resynthesis.

The authors of these studies focused on the potential influence of muscle hydration on glycogen resynthesis, reasoning that exercise-associated dehydration might constrain glycogen resynthesis because this process requires adequate intracellular water and K^+. Within muscle and liver, glycogen is stored in a hydrated form associated with about 3 g water and 0.5 mmol K^+ per gram of glycogen. As glycogen is utilized there is concurrent release of water and K^+ from the glycogen store and one previous study in rats demonstrated that cell shrinkage, as occurs with dehydration, is associated with decreased glycogen synthesis in skeletal muscle (Low *et al.*, 1996). The findings of Waller *et al*. (2009) suggest that post-exercise rehydration strategies may be useful when prompt restoration of muscle glycogen content is a goal. Nasogastric intubation is not a convenient (or desirable) means for the administration of a electrolyte solution but the quantity of electrolyte given in the present study can be administered another way, e.g. as a slurry via dosing syringe – and previous studies have shown that electrolytes given in this manner do enhance rehydration post-exercise providing horses have access to water.

Comments on the study

An advantage of the present study was the use of an exercise protocol that simulated actual competition exercise. Earlier studies that examined post-exercise muscle glycogen resynthesis employed arduous exercise protocols (multiple sprints after prolonged endurance work; multi-day protocols) with a primary aim to deplete muscle glycogen to 30-40% (or less) of its initial value. A drawback of these protocols is that the exercise does not simulate the real world and, as a result, the practical application of the results is questionable. Of interest and potential concern, the total feed provided during 12 h of recovery (6 kg oats, 6 kg hay) exceeded that provided in 24 h (4 kg oats, 4 kg hay) during the 2 week period prior to the study. It may be argued that this sudden change in diet (i.e. a 50% increase in the quantity of oats, shortened intervals between oat feedings) is risky from the perspective of gastrointestinal health although adverse events were not reported.

What next?

A study that uses hay only as the base diet during recovery would be an interesting follow-up. Some horsemen prefer to feed only hay during the early phase of recovery from competition exercise, so the question arises: Does electrolyte supplementation enhance rehydration and muscle glycogen resynthesis when horses are fed only hay during the early (e.g. 12 h) post-exercise recovery phase?

Waller AP, Geor RJ, Spriet LL, Heigenhauser GJF and Lindinger MI. Oral acetate supplementation after prolonged moderate-intensity exercise enhances early muscle glycogen resynthesis in horses. Experimental Physiology (May 29, 2009 – epub ahead of print).

Objective

To determine the effect of oral acetate administration after glycogen-depleting exercise on plasma acetate appearance, skeletal muscle acetate metabolism and glycogen resynthesis. It was hypothesized that (1) administration of an oral sodium acetate/acetic acid solution with a typical grain and hay meal after glycogen-depleting exercise will result in a rapid appearance of acetate in blood with rapid uptake by skeletal muscle; and (2) within skeletal muscle, acetate will be converted to acetyl-CoA and acetylcarnitine, which will be metabolized to CO_2 and water via the TCA cycle, generating ATP within the mitochondria and thereby allowing glucose taken up by muscle to be preferentially incorporated into glycogen.

Background

It has been suggested that low availability of lipid metabolites during the post exercise recovery period may limit muscle glycogenesis by redirecting glucose away from glycogen synthesis to support immediate energy needs through the tricarboxylic acid cycle (Hyyppa *et al.*, 1997), i.e. available glucose is partitioned to oxidation rather than storage. Therefore, post-exercise feeding strategies that directly or indirectly (e.g. via production of volatile fatty acids by microbial fermentation) increase the availability of alternative substrates for the TCA cycle (such as acetate) could support glycogen synthesis. Interestingly, the ingestion of a glucose/acetate solution increased glycogen replenishment and glycogen synthase activity in rats after exercise when compared to glucose alone (Fushimi *et al.*, 2002).

Overview of the study

Eight Standardbreds and 1 Thoroughbred horse (7 geldings, 2 mares) were used in a crossover design in which each horse performed a Competition Exercise Test (CET) twice in randomized order, an acetate treatment and a control trial. The base diet was 6 kg mixed hay and 4 kg of commercial sweet feed. The CET was as described in the preceding paper. Within 10 min after completion of the CET, the horse either 1) received by nasogastric tube a sodium acetate-electrolyte solution containing 500 g sodium acetate (134 g Na, 366 g acetate), 250 ml acetic acid (250 g acetate), 32 g KCl, and 300 g glucose in 8 l of water (acetate treatment), or 2) stood in stocks for equivalent amount of time (Con). The acetate-glucose dosage was calculated on the basis of the number of glucosyl units needed to replace an approximately 40% decrease in muscle glycogen content. Muscle biopsy samples (*gluteus medius* m.) were collected before exercise, 15 min after cessation of exercise (0 h), and at 4 h and 24 h post-treatment. Blood samples were taken at 20-60 min intervals up to 8 h of recovery, and again at 24 h of recovery. In both treatments, horses were given 3 kg hay and 2 kg sweet feed at 20 min after completion of the CET; a further 2 kg sweet feed and 3 kg hay was given at 6 h of recovery, then 2 kg hay at 12 of recovery. Analytes measured in muscle samples included glycogen, acety-CoA and acetylcarnitine contents, and pyruvate dehydrogenase activity. Plasma acetate and glucose concentrations were measured in blood samples.

Main findings

Similar to their previous study, Waller *et al*. observed an approximately 20% decrease in muscle glycogen content after the CET protocol. Acetate treatment resulted in a rapid and sustained increase in plasma acetate concentration and skeletal muscle acetyl-CoA and acetylcarnitine concentrations were significantly increased at 4 h of recovery, suggesting substantial intestinal uptake and followed by tissue (skeletal muscle) extraction of the supplemented acetate. The rate of muscle glycogen synthesis during the initial 4 h of recovery was higher in horses supplemented with acetate. However, from 4 to 24 h of recovery glycogen synthesis was greater in the control treatment such that by 24 h of recovery muscle glycogen content was similar in both treatments. Some of the acetate treated horses exhibited signs of mild gastrointestinal disturbance (poor appetite and transient loose feces). In the control treatment horses finished all of the offered sweet feed by 20 min post feeding (40 min post exercise), and had consumed all of the hay provided by 3-4 h post feeding. In the acetate treatment, the horses consumed all hay by 4-6 h post feeding but only consumed ¼ of the sweet feed within 20 min post feeding and did not eat remaining grain from both meals until between 12-24 h of recovery. Plasma glucose concentrations did not differ between treatments and in both groups remained significantly increased from pre-exercise during most of the recovery period.

Practical interest

The addition of acetate to post-exercise meals may be beneficial in regards muscle glycogen replenishment but further studies are needed before this strategy is ready for 'prime time'. Although not assessed in the present study, the sodium acetate-acetic acid solution has low palatability and is unlikely to be voluntarily consumed even when mixed with other feedstuffs. The clinical signs of gastrointestinal disturbances are a further concern; these may have been related to the acetate or acetic acid or due to fluid shifts into the gut and changes in hydration associated with the administration of a very large sodium load (134 g). Other forms of acetate (e.g. triacetin) should be studied.

Comments on the study

In theory, the glucosyl units provided in the acetate treatment (glucose/acetate) were more than sufficient to replace the glycogen degraded during the exercise protocol and this supplementation likely accounted for the early enhancement in the rate of muscle glycogen storage. The rate of resynthesis then slowed (lower than the control treatment between 4-24 h), perhaps the result of the poor feed (sweet feed) consumption and therefore ongoing availability of glucose for glycogen synthesis. Studies in human athletes have demonstrated that small, frequent doses of carbohydrate (1.0-1.8 g/kg/h) increase muscle glycogen resynthesis rates compared to a single dose that delivers the same total amount of carbohydrate (Jentjens and Jeukendrup, 2003).

What next?

As mentioned, a more palatable form of acetate or acetate/glucose combination is needed. To be practical, this supplement must be palatable and readily consumed when mixed with other feed, e.g. chaff or hay chop. With this formulation in hand, next steps might be to repeat the present study and/or evaluate the efficacy of smaller, frequent doses given over a 6-8 h recovery period. A study design that compares hay only vs. hay plus acetate/glucose supplement would more directly address the question: does supplemental acetate (or a combination of glucose/acetate) enhance post-exercise muscle glycogen resynthesis?

Summary

The two papers discussed above add to the body of knowledge concerning the effects of post-exercise feeding on muscle glycogen replenishment in horses. So far, it is clear that glycogen resynthesis in horses is quite slow when compared to other species and only modestly influenced by post-exercise feeding strategy. In my opinion, the post-exercise diet should not be radically different (if at all) from the regular diet, with an emphasis on providing good-quality forage rather than 'pushing' cereal grain. The studies by Waller and colleagues indicate that attention should be given to replacement of water and electrolytes (rehydration) and, potentially, the use of substrates other than glucose (e.g. acetate) for enhancement of muscle glycogen resynthesis.

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What's new in Equine Nutrition since 2007?

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We are responsible not only for our own diet but for the diet of the animals under our care. This means that trends apparent in the human nutrition field often are paralleled by changes in the feeding practices and management of domestic animals. The three areas chosen for discussion here highlight this: increased use of supplements, quality of the diet, and obesity.

NRC Committee (2009) Safety of dietary supplements for horses, dogs and cats. National Academies press Washington.

Objectives

'The committee's assignment was to assess the safety of three dietary supplements (Lutein, evening primrose oil and garlic) offered to horses, dogs and cats. During its review of the data for each of the three supplements, the committee was asked to examine general considerations that need to be taken into account in determining the safety of animal dietary supplements. The committee was to describe its findings and conclusions about the safety of the three supplements and provide recommendations for factors to consider in future analyses'….'the committee did not assess the validity of utility or efficacy claims'.

Why chosen

We are all aware that there has been an increased use of 'specialised supplements' and 'nutraceuticals' in horse diets with very few feed rooms not containing one or more products. In fact it seems that very few horse feeders today provide their animals with a simple, single cereal or compound feed plus roughage diet. Many add other separate feeding stuffs and supplements in order to personalise the diet, add that little bit extra to boost performance, improve health, correct an imbalance etc. However, there is a great deal of, often heated, discussion over this whole topic covering areas such as how do we define a supplement, are they needed, do they work and are they legal? Unfortunately an important consideration, often forgotten with respect to supplements is 'safety'. Typical arguments are that the ‘only thing hurt by trying a product that doesn’t work is your wallet’, or ‘its been fed for years it must be safe’ – but this is not always true.

Background

The exact expenditure on dietary supplements for horses is unknown, however a USDA survey undertaken over ten years ago (NAHMS 1998) suggested that around 5% of horses on US farms were fed herbal supplements and nearly 4% were fed a joint support preparation. It is likely that these numbers will have increased dramatically over the last 10 years. Some nutritionists are of the opinion, for both humans and animals, that supplements are unnecessary because a balanced diet will provide all the nutrients required. However, one of the tenets of good feeding is to feed the horse as an individual, and supplements are a means by which the diet can be tailored. Not all animals can be provided with a balanced diet nor can the ‘feeder’ always be certain of the quality of the diet (in particular the forage component) and in such instances the supplement may act as an ‘insurance cover’. Whilst this might seem reasonable, even under these circumstances safety considerations obviously need to be taken into account. Taking into consideration all sources of a particular nutrient in the ration, for example, can be very important when considering adding in a ‘supplement’ or a mixture of different ‘supplements’ all containing that nutrient. For selenium, for example, the differences between adequate, optimum and potential toxic intakes are fairly small, and levels of toxicity could be approached if multiple selenium containing ‘supplements’ are used.

Then there is the issue of the more specialised products or nutraceuticals, which are suggested to have positive effects on, for example, health, performance and behaviour. These supplements may contain nutrients in amounts far greater than minimal nutritional requirements (e.g. high doses of Biotin, and antioxidants such as Vitamin E) or other components for which there are no scientifically established dietary requirements (e.g. chromium, various herbs, chondroitin sulphate, glucosamine). Supplements may consist of single ingredients, simple combinations or more complex cocktails of feed ingredients and additives. Increasingly we are seeing novel materials being marketed, with particular health claims, but little evidence of efficacy nor safety. For some animals using a product that does not work will mean that their clinical condition remains unresolved and even get worse, or it might result directly in an adverse effect through toxicity, interaction with other components of the ration or medications etc. The nature of the supplement, how it was made or processed, what it is being fed with, the status of the horse etc., all may influence whether a particular supplement is safe or not. Depending on the material there may be a risk of contamination by unwanted and/or potentially toxic materials. It is important to realise, for example, that cross-reactions are known to occur between certain medicinal/drug therapies and herbal/spice preparations.

Overview of the review

This report was intended to help form the basis of a more general framework for evaluating animal dietary supplement safety. The committee was faced with trying to provide recommendations for dietary supplements for which there are insufficient safety data of a quality normally required for animal drugs and animal food additives (as is the typical situation for many 'supplements'). They approached this problem by reviewing all the literature available (in the English language) relating to the supplements, concentrating on any published scientific reports relating to safety in the target species and other related species, as well as humans and rodents. Additional information was also received directly from pet food and animal dietary supplement industry representatives. The review provided guidance on the factors that should be considered when evaluating the safety of any dietary supplements for horses, dogs, and cats.

Main findings

One of the initial outcomes was the committee's working definition of an animal dietary supplement (ADS) as currently there is no legal definition in the EU or the US of a supplement. The definition was 'a substance for oral consumption by horses, dogs or cats, whether in/on feed or offered separately, intended for specific benefit to the animal by means other than provision of essential nutrients recognised as essential or for provision of essential nutrients for intended effect on the animal beyond normal nutritional needs but not including legally defined drugs'. The other key findings were:

- Confirmation that there are insufficient safety data to define a no observed adverse effect level (NOAEL) or even a safe upper intake level (SUL) for the materials in question. The committee therefore provided guidance as to their ranking when assessing acceptable and relevant data, noting that on a 7 point scale the majority of the data available and used was in the lower classes (4,5,6). With such limited data, the committee could only report historical safe intake and estimate a presumed safe intake (PSI) e.g. for the horse:
  - for lutein: PSI of 8.3 mg/kg BW when fed as a forage or natural sources; no data to support supplementation;
  - for evening primrose oil: PSI of 400 mg/kg BW, assuming the level of total fat < ~23% of diet by weight;
  - for garlic: historical safe intake of 15 mg/kg BW of dried garlic powder: PSI levels of up to 90 mg/kg BW may be OK in healthy non-stressed horses; levels of 90-200 mg/kg BW may be threshold for toxicity in some.
- There is a clear need for a comprehensive adverse event reporting system.
- There needs to be clarification of the regulation of animal dietary supplements.

Practical interest

This work confirms that just because ingredients have been fed for centuries it does not mean that they are always safe to be fed at any intake level, as is illustrated by garlic. It also illustrated that apparent safety in one species does not automatically mean that the material in question is safe in others. Safety, however, can be difficult to determine absolutely under all circumstances. Whilst it is likely that this report may result in some increased regulation of supplements, at least in the US, linked to safety, it is important that the defining bodies for such regulations accept that traditional toxicological evaluations (such as the dose at which 50% of the animals die) are not going to be performed in horses (or dogs/cats). There is therefore a need to determine practical guidance as to the means by which novel ingredients can be determined as safe without very expensive studies having to be undertaken or studies requiring significant adverse signs to occur before safety can be 'defined'. Companies and researchers in the future would benefit from considering more closely monitoring the animals used in their trials for specific adverse event signals. These should be based on a thorough review of the available literature.

Comments on the study

It is unfortunate that the committee (of which the author was a member) was not instructed to recommend a framework for the assessment of supplements in the future.

What next

The veterinary, feed and supplement industry need to consider very carefully their response to this review.

Martin L, Geffroy O, Bonneau A, Barre C, Nguyen P, Dumon H (2008) Nutrient intake in high performance show-jumping horses in France. In: Nutrition of the exercising horse. Saastamoinen MT, Martin-Rosset W (eds). EAAP publication no 125. Pages 333-340.

Objectives

To describe dietary practices in show jumping horses and compare them with the NRC recommendations.

Why chosen

This work was presented as a short presentation during the 4th European Workshop on Equine Nutrition (EWEN) held in Forssa

Finland in 2008. This presentation promoted significant thought provoking discussion – not because the data was flawed or the study incorrectly undertaken but because it highlighted, to the author at least, the difficulty in persuading people of the value of nutrition when high performance horses can apparently perform very well on apparently/theoretically inadequate diets.

Background

Few published data are available on feeding and management practices in performance horses in general and the show jumper in particular. The new NRC 2007 provides recommendations as to the minimal levels for various nutrients in the diet but there is little data available looking at how these levels are reflected in typical practice in the field.

Overview of the Study

Data from 17 elite show jumping horses was collected. These horses were visited once in their stable and their general health, weight, body condition etc. was determined. Details of the feeding and management practices were obtained and the feeds being fed were analysed.

Main findings

All of these elite horses were housed in individual stalls and were never grazed. The average forage to concentrate ratio was 55: 45 and the dry matter ingested ranged from 1.2-2.4% BW. All the horses were fed some commercial feeds although for 10 of the horses these were mixed with oats and/or barley. Mean DE intake was 1.4 +/- 0.1 times the maintenance requirement but ranged from 1- 1.9 times. Crude protein intake ranged from 84-147% of the recommendations for exercising horses. Although the mean mineral intake (Ca, P, K, Cu, Zn Fe) exceeded the NRC recommendations, not all animals were being fed above the minimal level recommended. The authors report that only one diet was balanced for Fe, Cu, Cl and Zn. The sodium intake was below the NRC recommended allowance in 9 horses but was very high in one horse. The authors noted that, 'in the present study we found that mineral intake varied widely between horses depending on ration composition and the resulting diets were sometimes very unbalanced. Some of the identified imbalances, if continued could explain the chronic fatigue or decreased appetite sometimes observed in exercise horses'.

Practical interest

The authors conclude by saying that 'the present study underlined the interest of determining and correcting the nutrient concentrations of rations. The analysis should be repeated several times per year to take into account the variability of the feed sources'.

This is where the controversy starts – these horses were all apparently performing at the elite level – and therefore unless they develop a problem the owners/feeders could assume that all is well for their individual animal. For the majority of the horses in the study (except perhaps the one with chronic muscular problems) it could be assumed that any imbalances were either irrelevant or manageable for that horse. However, no-one knows whether their performance could be enhanced if the imbalances were not present – or maybe those horses that reach the elite level of any discipline are those that can 'cope' with the demands of that discipline despite the feeding and management practices they experience. Often nutrition is only considered 'seriously', with the owners/feeders willing to make substantial changes, once a problem arises – which might be too late unfortunately. Once a problem starts – if nutrition is considered to play a potential role – then analysis at that time may or may not identify a possible nutritional cause *but* it does not reflect the nutrient intake over the previous 'x' months or years. Forage in particular changes in its micronutrient content and therefore, without the regular evaluations that the authors suggest, it is difficult to assess actual intake over time. This is then the dilemma – how to justify the cost of regular analysis and evaluations when many animals might perform well for years despite a mild nutritional imbalance causing minor limitations the results of some imbalances may be long term rather than short term in nature; are we sure our recommended intake levels are ideal for all horses under all circumstances etc. etc. What is optimal nutrition?

If the researchers had been able to take half the study group and correct the imbalances and then show an improved performance over the short and long term then perhaps the show jumpers might pay attention. This, however, rarely happens and is fraught with study design issues and questions *but* somehow we need to overcome these so that we can show in the field the true short and long-term value of feeding appropriate balanced diets to horses, in particular high performance horses.

What next

If a study could be undertaken that supported the value of good nutritional practices for long-term performance it could be to the benefit of nutritionists all over the world.

Dugdale AHA, Curtis GC, Knottenbelt DC, Harris PA, Argo CMcG (2008) Changes in body condition and fat deposition in ponies offered an ad libitum chaff based diet. In: Proc 12th Congress Europ Soc Vet Comp Nutr. Page 39.

Objectives

To characterise concurrent changes in voluntary food intake (VFI), body condition score (BCS), direct (ultrasonographic) and indirect (deuterium oxide dilution technique) measures of body fat in ponies fed *ad libitum* during summer and winter.

Why chosen

In both the UK and the USA obesity appears to be a growing concern for horses as well as humans. In a recent cross-sectional study of 300 mature horses in the US 19% were reported to be obese with body condition scores (BCS) of 8 or 9, and 32% were overweight (BCS 6.5-7.5) (using a 9 point BCS system: Thatcher *et al.*, 2007). In Scotland, a recent review (Wyse *et al.*, 2008), of 319 pleasure riding horses using a 6-point scoring system, suggested that 32% would be considered to be obese (BCS 6) and a further 35% would be considered fat (BCS 5). There is increasing evidence of an association between obesity, altered metabolic function and disease. Whilst it is obviously very important to develop effective strategies to manage the obese pony it may be equally important to understand why they become obese in the first place. In addition, it will be valuable to know how best to monitor, more accurately and appropriately, changes in body condition, given that the current body condition scoring systems were developed for light breed horses.

Background

Obesity has been associated with insulin resistance in horses and ponies, and insulin resistance in the presence of obesity has been associated with altered reproductive activity in mares and an increased risk of and predisposition for pasture-associated laminitis. For many horses, energy intakes above their own individual requirements will result in either an increase in weight and/or a change in behaviour and most cases of obesity will be linked to an energy imbalance. Genetics may be another factor in the predisposition to obesity. The term 'easy keeper' is often used to describe a horse or pony that has a tendency to be overweight and appears to require fewer calories than most horses to maintain condition. One hypothesis is that certain lines of horses and ponies have inherited genetic traits that have facilitated survival on poor quality forages and/or in the face of limited feed availability – the so-called 'thrifty genotype'. Native UK pony breeds for example retain strong seasonality with respect to their appetite and under 'feral' conditions tend to gain weight during the summer months when food is abundant before losing it again during the winter. Such cyclical changes may not occur when food intake and quality is maintained during the winter, resulting in further weight gain.

Overview of the study

Mature (5-19 year old), Welsh Mountain pony mares were studied in either summer (n = 6, 244±20.3 kg) or winter (n = 5, 221±21.2 kg). Each group comprised 2 animals in thin (BCS1-3), moderate (BCS 4-6) and fat (BCS 7-9) condition. They were then monitored over twelve successive weeks during which all animals had *ad libitum* access to a chaff-based, complete diet (GE 16.8 MJ/kg DM). Ponies were weighed daily (±1kg). Body condition score was subjectively appraised each week and the depth of subcutaneous fat at both the 12th intercostal space and the middle gluteal (rump) region were measured ultrasonographically. Estimations of body fat percentage were made at the beginning and end of the 12-week study period using the deuterium oxide dilution technique.

Main findings

Appetites became maximal by week 5 for both groups. Appetite was greatest in summer ponies in thin and moderate BCS. Conversely, the appetite of fat ponies was similar between seasons. Summer ponies of thin and moderate body conditions in week 1 had the greatest average daily gains (ADG) in body mass (0.73±0.04 kg/d); whereas thin and moderate winter ponies gained only 0.47±0.05 kg/day. Fat ponies tended to gain very little body mass in summer (0.01±0.06 kg/d), but showed ADGs of 0.33±0.2 kg/day in winter. BCS changed little throughout the studies in fat ponies, but increased significantly (c. 3 points on a 1-9 scale), in the thin animals in both summer and winter. Ultrasonic fat depth at the 12th intercostal space but not the rump correlated well with total body fat percentage derived from deuterium oxide dilution (12th intercostal space, r = 0.82; gluteal region, r = 0.59).

Practical interest

The average daily intake of the fat ponies did not seem to change significantly even when provided a long-term *ad libitum* diet. Their actual intake was around 2% of BW on a Dry matter basis and this resulted in either no or a slight increase in body weight. This confirms that restricting obese ponies to a hay diet of around 2% BW on a DM basis is unlikely to result in weight loss. Season does appear to influence weight gain and appetite as well as the BCS of the pony. Rib fat (12th intercostal space), may be a more reliable predictor of total body fat in ponies than rump fat.

Comments on the study and associated literature

This study and associated literature prompt many questions and comments including:

- Should we try and have a harmonised body condition scoring system?

  There are numerous body condition scoring systems used throughout the world e.g. the 1-9 point Henneke *et al*. (1993) system used by Thatcher *et al*. (2007) and many researchers in the UK and US; the 1- 6 point Webb and Weaver (1979) system used by Wyse *et al*. (2008); the 0-5 point system used in France and many European countries (INRA1990) and the 0-5 point Caroll and Huntingdon (1988) adapted system often used in Australia. This potentially could cause confusion when looking at results, although very fat/ obese animals at the top of any of the scales are likely to be very fat/obese.

- Can a BCS system be the same between all breeds of horses and ponies?

  The Henneke 9-point system was originally developed from work carried out predominantly using Quarter Horse broodmares and is therefore perhaps more suited to a lighter breed type of animal, including the Thoroughbred. Use of the systems described above prompted a group of workers in Germany to adapt the system for the Warmblood horse (Kienzle and Schramme, 2004). They scored (and took ultrasound measurements as well as measurements of skin fold or tissue thickness and neck fat) from around 180 Warmblood horses and produced an amended 9-point system.

  Certainly ponies are not miniature thoroughbreds, and they differ not only in their general appearance but often their natural tendency to accumulate fat and the preferred areas for fat deposition. This suggests that the existing horse BCS are likely to be inappropriate for many pony breeds. Therefore it may be very important to develop a more pony orientated scoring system – as noted by Argo (2009), 'great improvements would be achieved by establishing and promoting a pony-specific body condition scoring (BCS) system which would facilitate the expedient detection and correction of insidious weight changes'.

- What exactly does a body condition scoring system measure and how does it respond?

  Body condition scoring is, in part, a measure of how fat or thin a horse is. Much of a horse's body fat is subcutaneous but it is not spread evenly around the body and therefore tends to accumulate in specific areas. It is these specific and identifiable areas that are examined more closely when using a BCS system. Improved information as to the link between body condition score and internal as well as external fat deposition is required. Many of the body condition scoring systems were based on indirect indices of body fat. Recently more in depth evaluations involving cadaver work has been undertaken to start to provide more objective support for the BCS systems. In France a study of 20 adult horses was undertaken involving body weight and condition scoring (INRA 1990) pre slaughter and comparison with total fat tissue as well as subcutaneous, internal and intermuscular fat *post mortem* (Martin- Rosset *et al.*, 2008). Although the numbers were relatively small the authors reported a good correlation between the body condition score and the total fat tissue. This work needs to be expanded to other breeds as well as the pony and as Argo 2009 states 'however, our current knowledge of pony body composition is sparse and poorly documented and a much greater understanding of the endogenous, intensely seasonal, physiological control of fat deposition and mobilisation from different adipose tissues is required. The significance of discrete regional deposits to health also warrants investigation'. The above study and others that are being carried out in this key area may help us to understand what exactly an external Body condition scoring system can tell us and how it can be used to monitor changes.

- Is body condition scoring the best way to predict prediction to disease?

  This is another complex area – work in humans has shown that there are people with a normal body weight but a high total body fat mass, which have an increased risk of developing insulin resistance and other obesity related metabolic diseases. Ideally we need a scoring system that enables the detection of physiologically or pathologically relevant fat deposits. A recent publication, based on work previously discussed in this Nutrition news section (Bailey *et al.*, 2007), showed that there are insulin resistant, laminitis predisposed ponies which phenotypically cannot be distinguished from their non insulin resistant, apparently not predisposed to laminitis, cohorts. The importance of where fat has been deposited has been highlighted in human obesity work (apple vs. pear body shapes) and has been explored further in the pony by recent work in America (Carter *et al.*, 2009) given the suggestion that ponies with cresty necks are more likely to be at risk of laminitis. It is therefore likely that current BCS systems will not be able to differentiate between the different areas of regional adiposity that may indicate increased risk in the horse.

  This means that we might need to develop alternative or additional methods for evaluating disease risk. These might include additional objective evaluations of body type/fat mass such as the use of bioimpedance analysis (BIA) as illustrated by Van der Aa Kuhle *et al*. (2008), and deuterium oxide dilution as used in the above study. In addition, means to identify particular regional depositions of fat associated with increased risk such as the cresty neck scoring system proposed by Carter *et al*. (2009) may prove invaluable. The relative value of various fat ultrasound determinations needs to be considered for different types of animals (as discussed above and in Martin-Rosset *et al*. (2008). Overall all of these and other modalities need to be explored further in various breeds and types of ponies but they offer potential for the future.

- How do we tackle this increasing problem of obesity in horses? Although much more research is needed for us to understand the drivers of obesity as well as how best to safely encourage and monitor weight loss, the education of owners, feeders, feed companies, veterinarians and nutritionists is key to this question. In the Wyse *et al*. study – there was only a fair agreement between the owner's perception of obesity and the body condition score. The authors note that 'the owners of fat horses were most likely to score their horse incorrectly, and only 50 per cent of the owner estimates for fat horses were in agreement with the body condition score'. Such education needs not only to concentrate on an increased awareness of body condition scoring and weight determination but also on an improved awareness of some of the key drivers for weight gain e.g. excessive energy intake in respect of the level of exercise and therefore requirements.

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Dressage and show jumping

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Training for elite dressage and show jumping horses is driven by improved performance, both optimal short term performance and longevity of high performance. Recent research therefore includes investigation of current practices, detection of risk factors for injury and methods to improve training practices to achieve maximal performance. In addition, increased public concerns over welfare have led to investigation into some specific training techniques.

Warm up

Warm up during a training session and warm up in a competition environment has been shown to have both physiological and psychological benefits in other sports. However, for power sports, which would include show jumping and dressage at elite levels, warm up may need to take into account adequate physiological and psychological warm up requirement compared to muscular fatigue prior to performance. Recent papers investigating current practices in UK dressage and show jumping horses have been published. These looked at the effects of competition level and type on warm up total time and breakdown by pace or jumping efforts. In addition, the relationship between warm up duration (including the breakdown by pace) and dressage performance outcome was investigated.

Murray RC, Mann S, Parking TDH (2006) Warm-up in dressage competitions: association with level, competition type and final score. Eq & Comp Exercise Physiol 3(4):185-189.

Warm up of 266 competitors at British Dressage affiliated competitions was observed, including competitors at Novice (N) (n = 104), Medium (M) (n = 65), Prix St Georges (PSG) (n = 59) and Grand Prix (GP) (n = 38) levels. Competitions were classified as local (n = 102), regional championship (n = 54) and national championship (n = 106) events. Overall, the mean warm-up duration for competitors at dressage competitions was 29 min 53 sec. Total warm up time and proportions of walk, trot and canter at each level were recorded. There was no effect of rider experience, but level and type of competition affected the proportion of time spent in different paces and total time, which was increased at higher levels and championships. Increased warm-up time and specific warm-up design were positively associated with final score at Novice and Prix St George level.

Whitaker TC, Mills A, Duxbury LJ (2008) Horse warm-up regimes at two different competitive levels of show jumping: a pilot study. Comp Exercise Physiol 5:105-106.

This study compared warm up practices for 87 competitors between lower level and intermediate level show jumping. The results showed that competitors warmed up significantly longer overall at intermediate compared to lower level competition. More jumping efforts and more successful jumping efforts were observed at intermediate compared to lower level competition.

Comments

The results of these studies indicated that warm-up time appears to increase with competitive level in both show jumping and dressage. The results of the dressage warm-up study supported the hypotheses that competitors at higher competitive levels warm-up for longer than competitors at lower levels, and that warm-up at a championship was longer than at local level. At novice and Prix St Georges level, the results supported the hypothesis that increased warm-up time was associated with better performance, but this was not supported at other levels. Mean warm-up time was less than in a previous report of dressage horses in Germany which may reflect difference in practices between countries. However, details of movements performed during warm-up or of marks for individual movements in the tests were not recorded so muscular fatigue may not have been detectable using this study design. In addition, muscular fatigue may be more important at higher levels so further investigation of threshold for muscular fatigue in GP horses would be useful. Increased jumping efforts were observed at the higher level of competition, but only intermediate level competition was evaluated. It will be important to determine whether at elite level competition the number of jumping efforts increased from the intermediate level, whether this influences performance and whether there is a threshold beyond which performance may be compromised.

Dressage training practices

In order to understand ways to improve training practices, it is important to create a baseline of the current practices for training horses. There has been investigation into dressage training and management practices in the UK, as described below but there has been little reported from other countries which would be useful as a comparison.

Walters JM, Parkin T, Snart HA, Murray RC (2008) Current management and training practices for UK dressage horses. Comp Exercise Physiol 5:73-83.

It was hypothesized that there would be an effect of horse competition level on management and training. The objectives of the study were to record rider, horse, management and training information from UK dressage horses and to investigate relationships between the variables. Data from 2,554 respondents to a questionnaire was used to determine distribution of rider level and horse features (height of 164-171 cm and warmbloods were predominant), time in training, which increased by level up to Prix St Georges and frequency of competing, which was less at Grand Prix level. Management factors including turn out (most horses had 15-30 h turnout per week), nondressage exercise and dressage training (3-4 times per week). Mean warm-up of 16 min and cool-down was 11 min at all levels, and mean training duration was 36 min. Preliminary, Novice, Intermediare I and Grand Prix horses trotted most and cantered least in training. Overall, time spent in transitions, specific movements, and different types of paces varied between levels; elite horses spent more time in collected and less time in working than non-elite. Overall, there were differences in training and management practices between competition levels.

Comments

This is the first large descriptive survey in dressage horses. The findings indicate the patterns of management and training in dressage horses, which can be used as a baseline to investigate the effects of interventions.

Risk factors for lameness in dressage horses

Prevention of lameness is an essential component in training horses to elite level and staying at elite level. This study investigated the risk factors for lameness in relation to horse features, health factors, training and management and training surface features.

Murray RC, Walters JM, Snart H, Dyson SJ, Parkin TD (2009) Identification of risk factors for lameness in dressage horses. Vet J Apr 13. [Epub].

The aim of this study was to describe the prevalence of illness and lameness at different anatomical sites in registered United Kingdom dressage horses and to identify risk factors for lameness. The findings were based on a questionnaire sent to 11,363 registered members of British Dressage in 2005. Four multivariable logistic regression models were developed for each section of the questionnaire. A final mixed effects logistic regression model was developed which combined the results from all prior models. Owners reported that 33% of horses had been lame at some time during their career, with 24% of these within the previous 2 years. Various factors were associated with the occurrence of lameness in the last 2 years, including age, height, indoor arenas, horse-walkers, lunging (as protective), back problems, arenas that become deeper in wet conditions and sand-based arenas.

Comments

This study clearly showed association between training factors and lameness. The findings indicated that there was a potential protective effect of some types of non-dressage exercise and dressage training, possibly associated with improved proprioceptive or strength training. In addition there was evidence that adaptation to the training surface reduced lameness risk. However, certain types of training surfaces and those which changed under different conditions or across an arena surface increased the risk of lameness, as did some other health problems and individual features of the horse.

Position of the head and neck during dressage training

There has been much media attention in relation to head and neck positions in dressage training. However, there remains limited scientific information on the effects of different positions. Various different studies have concluded that there is minimal stress associated with a hyperflexed neck position (Sloet van Oldruitenborgh-Oosterbaan *et al.*, 2006; Van Breda, 2006) and subjectively improved responsiveness to the rider. In contrast another study suggested that horses choose to avoid this position or that is makes them more fearful (Von Borstel *et al.*, 2009).

An international collaboration investigating the effects of head and neck position on various aspects of locomotion and horse/saddle/rider interaction has led to various publications, which provide some guidance on the biomechanical effects and potential benefits and limitations of different head and neck position. These papers indicate that there are benefits and limitations to use of various head positions and that there is no clear biomechanical or kinematic evidence to suggest that a hyperflexed head and neck position is associated with detrimental effects. In general, the most negative effects on various different outcomes was in association with a hyperextended head and neck position.

Rhodin M, Gómez Alvarez CB, Byström A, Johnston C, van Weeren PR, Roepstorff L, Weishaupt MA (2009) The effect of different head and neck positions on the caudal back and hindlimb kinematics in the elite dressage horse at trot. Eq Vet J 41:274-279.

Objective

To evaluate the kinematic effects of different head and neck positions in 7 elite dressage horses ridden at trot on a high speed treadmill. The results indicated that an FEI position or hyperflexion increased back vertical excursion and sacral flexion compared to free trot on loose reins, while an extremely elevated neck position increased lumbar back extension. It was concluded that movement of the horse is different between being ridden on loose reins and in collected trot, but the exact degree of neck flexion was not consistently correlated to the movements of the horse's limbs and trunk at collected trot. Although an extremely elevated neck position produced some effects commonly associated with increased degree of collection, the increased back extension could be detrimental which riders and trainers should be aware of during training.

Comments

This study, along with other associated studies indicates that there may be some benefits for development of core muscle strength to riding with the head and neck in a flexed rather than hyperextended position.

References

Sloet van Oldruitenborgh-Oosterbaan MM, Blok MB, Begeman L, Kamphuis MC, Lameris MC, Spierenburg AJ, Lashley MJ (2006) Workload and stress in horses: comparison in horses ridden deep and round ('rollkur') with a draw rein and horses ridden in a natural frame with only light rein contact. Tijdschr Diergeneeskd 131:152-157.

Van Breda, E (2006) A nonnatural head-neck position (Rollkur) during training results in less acute stress in elite, trained, dressage horses. J Appl Anim Welf Sci 9:59-64.

Von Borstel UU, Duncan IJH, Shoveller AK, Merkies K, Keeling LJ, Millman ST (2009) Impact of riding in a coercively obtained Rollkur posture on welfare and fear of performance horses. Appl Anim Behaviour Sci 116:228-236.

Practical and controversial news on racehorse training

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Over the past 5 decades enormous progress has been made in our understanding of the physiology and mechanics of horse performance. This has had profound consequences on many aspects of horse management including veterinary diagnosis and treatment of exercise-associated conditions, nutrition and stable management. However, despite this huge accumulation of knowledge, principally by those in academic institutions, very little has changed in how horses are prepared for the rigours of competition. Many would agree that this is disappointing, since it is likely that there is much room for improvement. While there would be no great benefit to be had from making horses run faster, run further or jump higher, it is salutory to consider that high rates of injury and horses failing even to make it to the start have not fallen over the past 20 years or more.

This review covers findings that are relevant to the training of horses for racing and eventing. The studies discussed have been published between 2007 and July 2009 and cover a wide range of areas. These include the few that provide practical information on how training can be managed. Many others will inform the reader who is interested in maximizing not only the welfare of the horse in sports, but also the return on the investment of the owner, without whom of course there would be no horse sport.

The first section of this review addresses the more practically-relevant studies that have been published and information on products that are currently commercially available for use in monitoring and managing day-to-day training. The second section discusses studies that have added to the understanding of different aspects of horse training, with varying degrees of practical relevance. As someone working to make a living as a licensed racehorse trainer, but with a veterinary and exercise physiology background, I have selected information that I considered worthy of comment. This is not intended to be an exhaustive coverage of every study published.

Findings of practical relevance to training horses

Heart rate during TB racing

Mukai K, Takahashi T, Eto D, Ohmura H, Tsubone H, Hiraga A (2007) Heart rates and blood lactate response in Thoroughbred horses during a race. J Eq Sci 18:153-160.

Many would find it surprising that up until this study was published there had only been one report of heart rates in TB horses during a race (Krzywanek *et al.*, 1970). Mukai's study provides useful descriptive data of heart rates before, during and after 1,200 m simulated race in 3 year old TB horses. The race times and post-race blood lactate concentrations provide validation that the exercise load was similar to that in actual races. It is disappointing that no speed data were provided in association with HR data (achievable using GPS or RF systems for speed measurement), but the authors point out that this report should lead on to more wider-ranging studies. It would be very helpful to extend this investigation to measure workload of horses racing over a range of distances (TB horses race over distances from 800 m to 7,200 m) and on different surfaces. Workload measured during competition in other equestrian disciplines including eventing and endurance has provided useful information for designing appropriate training regimes.

Blood lactate measurement

The following two studies provide separate validations of a novel portable blood lactate meter designed for use on human subjects ('Lactate Pro'). This meter might have benefits over previously available portable devices since it uses an amperometric (rather than colorimetric) method for lactate concentration measurement, and as such may be less prone to interference from high PCV's in equine blood during intense exercise. It also requires a smaller sample size, thereby making it more amenable for use with skin puncture rather than venepuncture sampling (which might be more acceptable for repeated sampling in day-to-day practical field use).

Sloet van Oldruitenborgh-Oosterbaan MM, van den Broek ETW, Spierenburg AJ (2008) Evaluation of the usefulness of the portable device Lactate Pro for measurement of lactate concentrations in equine whole blood. J Vet Diagn Invest 20:83.

Blood samples were collected from horses jumping in an arena, horses running at racing speed on a track and from horses affected by colic. Data from the Lactate Pro were closely correlated to those from an ABL 'wet chemistry' analyzer, although slightly underestimating values by a mean of 6.6%. Lactate values collected during exercise were all lower than 8 mmol/l, although values in colic cases were as high as 18.6 mmol/l. The authors did not attempt to investigate the effect of elevated PCV on lactate values and stated that this needed further study.

Kobayashi M (2007) Simple lactate measurement in horses using a portable lactate analyzer with lancet skin punctures under field conditions. J Eq Sci 18:5-11.

This study set out to compare blood lactate concentrations measured by a standard (YSI) laboratory method and the Lactate Pro and Accusport portable meters, and to compare concentrations in blood from jugular venepuncture with samples obtained by lancet skin puncture.

This is only the second study to examine sampling by lancet skin puncture (the first was by Lindner and Birks, 1994). This approach is very relevant since such a method could potentially increase the widespread use of lactate-guided training in horses: in many countries blood sampling can only be undertaken by qualified veterinarians. The Accusport meter had been previously validated for horse blood (only able to cope with PCV up to 53) and required 15-50 µl of blood whereas the Lactate Pro only required 5 µl.

While the repeatability of the YSI analyzer was higher than either portable device, that of the Lactate Pro was greater than the Accusport and was acceptable over a range of values. Up to 10 mmol/l lactate the portable meters were closely correlated to the YSI, but at higher concentrations the Lactate Pro correlation weakened while the

Accusport lost its correlation. The Lactate Pro appeared to be reliable up to a PCV of 57, slightly higher than the limit of the Accusport which was at 54. There was close correlation between values from cervical skin puncture samples and those from the jugular vein.

This study provides some very useful information in that it reasserts the validity of taking samples from skin puncture, which is apparently technically easy to perform and well accepted by subjects. Promotion of this technique could help open up more widespread use of blood lactate measurement in guiding training regimes. However, although these data suggest that the Lactate Pro performs better than the Accusport, both are limited in that at the higher values of lactate and PCV encountered in intense equine exercise their accuracy becomes poor. This could be overcome by making measurements on plasma rather than whole blood and on diluted samples, but these procedures would preclude the practice of blood lactate monitoring becoming more widespread.

Practical equipment for performance monitoring

Combined speed and heart rate monitors

There are several commercially available products that can be used for monitoring work effort in horses. Speed is measured by a GPS receiver, and heart rate by 2 electrodes placed under the saddle/girth connected to a wireless transmitter. These can be used to:
1. set work rate by riding at target speed or heart rate;
2. make a precise record of work undertaken;
3. provide information regarding distances and gradient of terrain;
4. check that work riders rode to instruction;
5. provide real performance data when horses are being ‘tried’.

The products include the Polar RS800 G3, the Etrakka and the series of Garmin Forerunner units (301, 305, 405). Each of these provides a display on the rider’s wrist, or mounted on the head piece of the bridle behind the poll. While the first two have separate GPS antennae, the Garmin products have the antenna built into the watch receiver (Figure 1). These are human products and the human chest-belt electrode/transmitter unit has to be adapted for horse use (I can be contacted for details, www.naylor-racing.com).

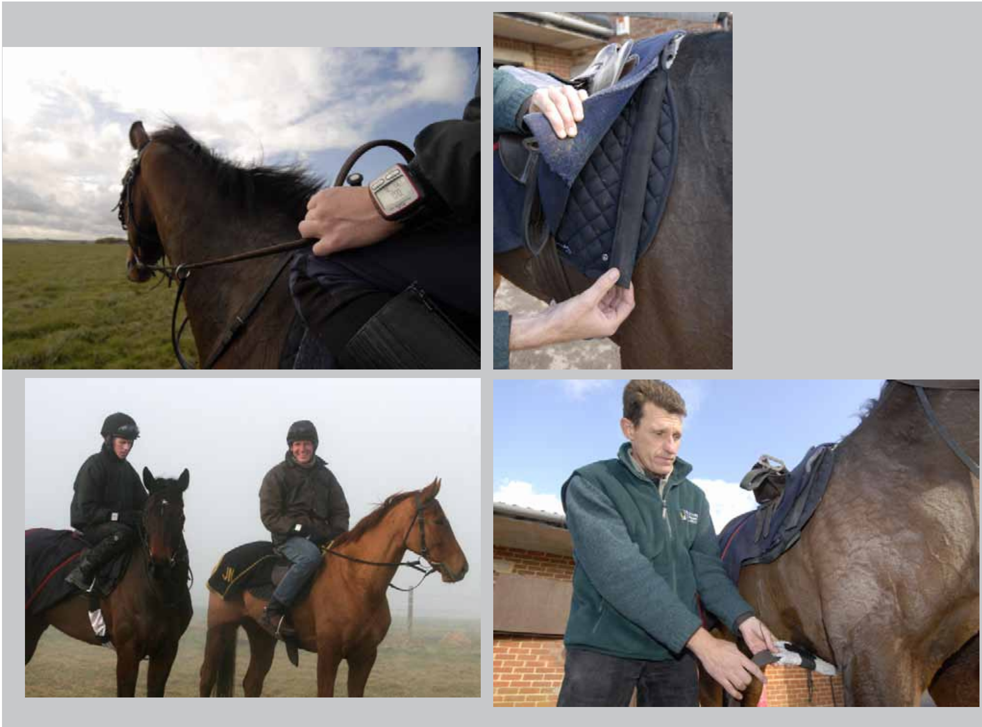

Figure 1. Garmin 305 in use.

A very exciting new product is in development (www.gmaxequine.com) that has been designed specifically for use with horses. It will include speed and heart rate monitors, it will have options for measuring stride characteristics and metabolic indices, and provision for real-time monitoring of horses from distant locations.

Software for analyzing performance data

Each product has its own software for downloading and analyzing the data. The Etrakka unit arguably has the best package to include calculations such as Vhrmax (speed at which maximal heart rate is reached) that may be used as an index of fitness. The standard Garmin software provides little analytical capacity but provides an easily viewed record of workouts (Figure 2).

A number of options are available for more detailed data analysis. Most proprietary exercise monitors can be downloaded into software

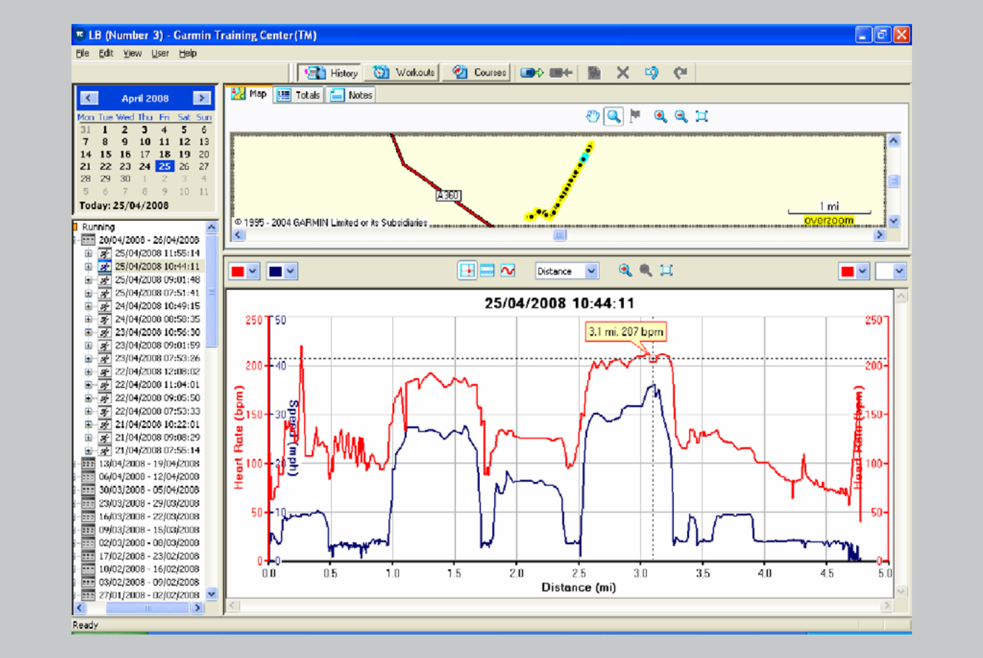

Figure 2. Garmin download, map above, exercise data below.

such as 'Sport Tracks' (Figure 3), which is available as donation-ware. This is an extremely versatile package that allows for a range of ways of examining data and allows for export to other analytical software (which some other systems do not allow for).

Portable blood lactate analysers

These products have been discussed above.

Stride measurement

A product has been launched recently for commercial use that has been independently validated. This provides data for stride length and stride frequency using a tendon-boot-mounted accelerometer and a hat-mounted GPS receiver. A recent addition to these services includes a similar system with accelerometers on each limb that provides information on limb phasing (www.etb-pegasus.co.uk).

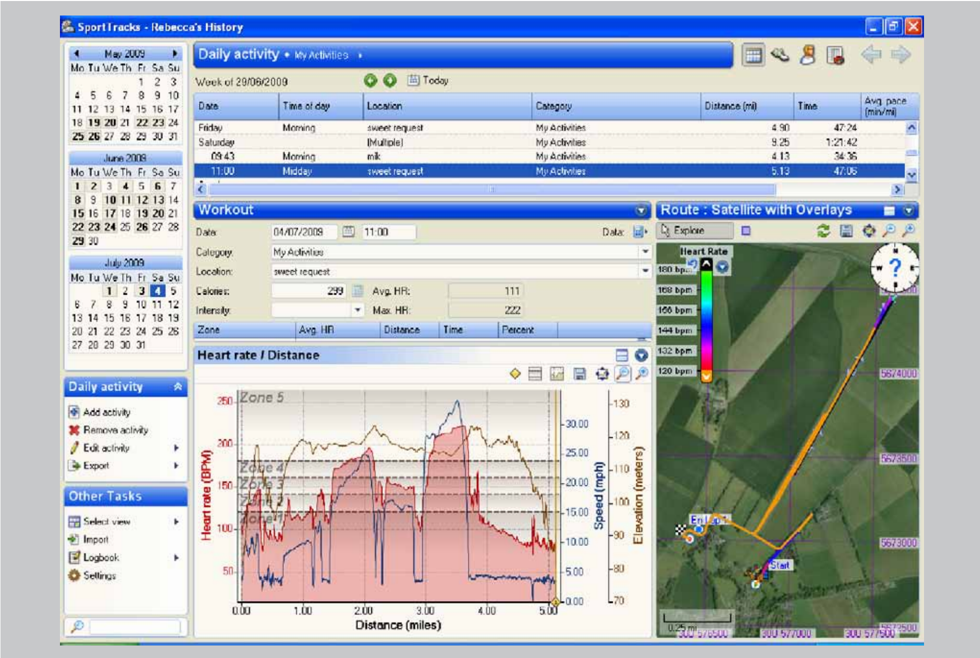

Figure 3. Sport Tracks download, map to right, exercise data to left. Incremental exercise workout: blue line speed (the lowest one when figure is not shown in full color), red line heart rate (the second lowest when figure is not shown in full color).

Alternative training equipment

A very innovative range of equipment to help in training racehorses has been developed by a Turkish company (www.kurtsystems.com). This includes a vehicle into which an individual horse is harnessed. The speed of the vehicle is controlled by a driver in the cab mounted behind the horse. A track system has been developed in which 4 horses can work at the same time and this may even be suitable for the early conditioning of foals (Figure 4).

Studies that have contributed to understanding how training might be improved

Training and injury

The evidence continues to mount for the need for closer monitoring of horses' work in training. Several studies are discussed below that have provided further information on this and other issues over the

Figure 4. Kurt Systems' horse training vehicle and track system for training horses without riders.

past three years. It has long been known that there is a high rate of 'wastage' in horseracing. Recent studies have suggested that in the UK as many as 40% of 2 year old TB horses in training and 25% of 3 year old in training do not make it as far as starting in a single race. This is principally due to musculoskeletal disease (Wilsher *et al.*, 2006). It becomes more clear as time passes that attention needs to be given to exactly how horses are trained and if current regimes can be improved. This can be achieved by quantifying work effort and then modifying as necessary, based on sound scientific understanding of performance and injury risk.

Dyson PK, Jackson BF, Pfeiffer DU, Price JS (2008) Days lost from training by two- and three-year-old Thoroughbred horses: A survey of seven UK training yards. Eq Vet J 40: 650-657.

Dyson *et al*. provide a survey of the causes for loss of training days that contribute to the stark statistics presented previously by Wilsher *et al*. (2006). Their data showed that 20% of all days available for training were lost due to lameness, and they concluded that there has been no improvement in the rate of injury to young horses over the past 20 years.

This injury rate remains high despite the much more widespread use of synthetic all-weather training surfaces and supposedly improved turf management and provision of watering. While the authors suggest that injury rates have not declined because horses may be trained harder than previously, this view would not be consistent with the failure of race times to improve to any great extent over the same period. A major hurdle that emerges in many studies is that there has been little effort to quantify work effort objectively in the horses and training regimes in question.

The most common cause of lameness was stress fractures, followed by carpal and fetlock injury and sore shins.

There were highly significant differences in injury rate between trainers. Days lost to training in relation to total days available for training ranged from 11 to 31% for 2 year old and 4.5 to 34% for 3 year old horses. This further adds great weight to the contention that *how horses are trained* has an enormous bearing on their risk of injury. There was no information provided by the authors regarding success rates of these trainers. Such data would be very useful in considering the implications of these findings in regard to a 'cost-benefit ratio': do successful trainers have higher or lower rates of injury?

Verheyen KLP, Price JS, Wood JLN (2009) Exercise during training is associated with racing performance in Thoroughbreds. Vet J 181:43-47.

Did the authors of this study really consider their title carefully? It is glaringly obvious to anyone with even very little knowledge of horse sport that exercise is likely to have a major impact on performance.

The philosophy that it is 'unsporting' to train because it gives one an unfair advantage was abandoned in human athletics after the Victorian age! (ref Preface to The physiology of training, G. White (ed), Pub Elsevier, 2006).

This study, despite its approach arguably being flawed in many ways, did make an attempt to consider the relationship between risk of injury and racing success. Their conclusion was that 'decreasing canter exercise during training in favor of more high-speed exercise' (an approach suggested to decrease risk of fracture and DMD) 'is unlikely to adversely affect race performance in flat races'. However, their conclusions must be considered in light of the observations set out below.

This work is hampered by a number of important flaws in the approach to testing the hypothesis that success in races is related to work done in 30 days prior to a race:

- Work was simply divided in to 'canter', speed of <14 m/s (approx. 31.5 mph) or 'gallop', speed of >14 m/s. This division is arbitrary, and there is no mention in the methods referred to as to whether or not speeds of typical working paces were ever recorded in the training yards in question. Gradient of gallop tracks was not considered either here and is a critical factor in dictating how fast trainers will work their horses (for instance on my own training gallops, a similar work rate is achieved by galloping 32 mph on a level track as is achieved by 28 mph on an inclined track with the same grass surface). This omission of providing more useful description of quality vs. quantity of work invalidates many of the study's conclusions.
- The approach does not take into account the stage of season for any horse. The conclusion that horses were more likely to win having raced in the previous 30 days may have as much to do with the experience from their first run (72% of horses were 2 and 3 year old), and in any older horses where many will run better having had their first run of the season.
- The type of racing will affect the results: this study has considered that all races are alike. The first 3 races of any horse's career in the UK are required to achieve a handicap rating, and most horses in the UK TB population will progress handicap racing. Only horses of the highest ability will continue to run in 'weight for age' or

'conditions' races. As such, in handicaps, weight is allotted according to the horse's ability in order that every horse theoretically has a chance of winning a race (because a horse of low ability is given less weight to carry than one of high ability).

- Following on, the use of prize money as a measure of success is a very insensitive measure of a horse's performance. It would be more useful to measure a horse's collateral form rating (many are available provided by professional race reading services including the British Horseracing Authority BHA) which gives a more objective measure of a horse's performance irrespective of type of race or weight carried.

Considering these important limitations, it would appear unwise for the authors to be making such simplistic recommendations to trainers as 'it therefore seems reasonable to advise trainers to try to adjust training schedules in favor of more high-speed and less canter exercise, in order to both reduce the risk of injury and create a successful racehorse'. Without any background knowledge of an individual trainer's facilities or regime, such a blanket statement is at best poorly conceived and at worst dangerous.

With the current ability to measure speed and work rate in horses in training, such statements are of no material use whatsoever to trainers. What would be more useful would be objective information of the type of build up of speed and distance over time (in relation to gradient) that would be appropriate for conditioning of bone and other tissues.

Parkin T (2007) Epidemiology of training and racing injuries. In: Havemeyer Workshop Report. Eq Vet J 39:466-469.

Here, Parkin summarises the discussions of the Havemeyer Workshop on training and racing injury.

Their recommendations included:

- *Prospective daily recording of training data*, in order to be able to analyse long and short term trends.
- *Type of exercise*: 'use of binary classification of pace of exercise is appropriate, i.e. fast work >14 m/s vs. other'. While this approach does not take into account many factors such as gradient, going,

weight carried etc., the report does go on to suggest that GPS speed and HR measurements should be made for describing work effort, and taking account of topography, surface type and weight carried. Patterns that are used within training regimes should also be considered.

- *Case definitions*: should be tightened up, since these are very wide in many studies and preclude making precise conclusions from data sets.
- *Condition of racing surface* merited its own comment, referred to as 'going' or 'track rating'. They concluded that current descriptions are not reliable due to lack of objectivity and wide variation around tracks on the same day.

Although it was not stated, this is an area that must be developed before much sense can be made of relative risk factors and performance data. Neither penetrometers nor the 'Goingstick' which is currently used on all UK turf tracks provide reliable objective data and there is so far no means for quantifying going on synthetic surfaces. It might be possible that horse-mounted devices are needed.

Cogger N, Evans DL, Hodgson DR, Reid SW, Perkins N (2008) Incidence rate of musculoskeletal injuries and determinants of time to recovery in young Australian Thoroughbred racehorses. Aust Vet J 86:473-479.

This study followed 248 2 and 3 year old TB horses in Australia and set out to describe the incidence of musculoskeletal (MS) injury and the time to recovery. Unlike many previous similar studies, an injury was only considered as such if it resulted in the horse leaving the stable for ≥ 7 days and time to recovery was considered to be from the onset of the MS injury to the next race (not resumption of training).

Key findings included that shin soreness was the most common injury, 2 year olds were 2.99 times more likely to experience a first injury than 3 year olds, that 70% of injured horses returned to training with 55% recovered within 6 months. Although there were important differences in methodology and classification, the injury sites and rates were largely consistent with the findings of Wilsher (2006) and Dyson (2008) and others. Similarly there was a relatively low incidence of soft tissue injuries. Those horses that had been training at high speed (>14.8 m/s) or had raced already were considerably (almost twice and four times respectively) more likely to have a successful return to training than those that had not been forward enough to be working at that pace. There was a clearly significant effect of trainer on risk of injury and outcome.

These latter points are of interest because the ability to have worked at higher speed may be consistent with Verheyen's work in that higher speed work may have provided some protection from injury or at least greater chance of recovery, and that individual training stable factors played a significant role, further adding to the requirement for more detailed understanding of training practices.

One aspect of this study that is worthy of note is the authors' attempt to consider the pace at which horses are trained. Rather than simply using a cut off point of 14 m/s for fast or slow work, they divided galloping exercise into 5 categories: 'trot and/or canter' at <10 m/s, 'Evens' or '¾' pace at 13.3-14.8 m/s, 'home on the bit' at >14.8 m/s, 'trial' at or near race pace and 'race' at race pace. These categories of work rates were based on instructions commonly given by trainers to work riders, but did not appear to be validated by measurement. However, at least this approach acknowledged that some trainers were capable of adopting more than two speeds in training their horses, unlike in Verheyen *et al*.

Cogger N, Perkins N, Hodgson DR, Reid SW, Evans DL (2008) Profiling training preparation in young Australian Thoroughbred racehorses. Austr Vet J 86:419-424.

This study aimed to examine the impact of injury on training programs of 2 and 3 year old TB horses.

Here survival analysis was used to examine training and its relationship to injury and time in training before and after racing, in terms of 'preparations'. These were defined as groups of days leading up to an injury or a series of training days up to a period of >7 days away from the stable. The conclusion was that injuries in 2 and 3 year old in Australia did not have a long-term impact on the performance measures used in the study.

Jackson BF, Dyson PK, Lonnell C, Verheyen KLP, Pfeiffer DU, Price JS (2009) Bone biomarkers and risk of fracture in two- and three-year old Thoroughbreds. Eq Vet J 41:410-413.

Despite large amounts of research funding directed to studies of bone metabolism over the past 20 years, this short communication burst the bubble of one key promise, that blood tests would soon be available to determine which horses were affected by subclinical stress fractures that might progress to catastrophic breakdown. The authors followed 529 2 year old and 326 3 year old horses to relate the concentrations of blood-borne markers of bone metabolism with confirmed fractures associated with exercise. 11.3% and 9.2% of 2 and 3 year old, respectively, sustained fractures but there was no relationship between incidence and any bone marker concentration. What was of interest was a large range of fracture incidence between 7 trainers (0-19%). The authors concluded that of all the possible contributory factors, 'the way that horses are trained is likely to be one of the most important'.

Further weight is given to the importance of well managed training programs by the following studies. Many soft tissue injuries are the result of the accumulation of microtrauma over time as a result of repeated exercise loading. The aim of a successful trainer is to maximize fitness while minimizing risk of injury by utilizing the least possible amount of work to achieve that fitness.

Patterson-Kane JC, Firth EC (2009) The pathobiology of exercise-induced superficial digital flexor tendon injury in Thoroughbred racehorses. Vet J 181:79-89 (Review).

This is a comprehensive review of the current state of knowledge of the function and repair of tendon in relation to superficial digital flexor tendon injury in Thoroughbred racehorses. Tendon injury arises from a progressive accumulation of microdamage related to age and exercise trauma. Excessive repetitive loading of the tendon is associated with progressive damage to the matrix that cannot be repaired adequately by tenocytes in the time available or in the face of overwhelming challenge to cellular restorative processes due to the continuing exercise burden.

While providing an up to date review of the knowledge of tendon pathophysiology, it concludes that we are still a very long way from overcoming this spectre that haunts all forms of high intensity equine sports.

Ely ER, Avella CS, Price JS, Smith RKW, Wood JLN, Verheyen KLP (2009) Descriptive epidemiology of fracture, tendon and suspensory ligament injuries in National Hunt racehorses in training. Eq Vet J 41:372-378.

This report provides data for rates of injury by fracture and to flexor tendon and suspensory ligament (TLI) of a sample of 1,223 jump racehorses over a 2 year period, and provides important new information of rates of injury to older horses in training and racing.

Overall, in contrast to the studies of flat racehorses discussed above, the rate of TLI was nearly twice that of fracture, at 1.9 and 1.1 per 100 horse months, respectively. Of the fractures the most common sites were the pelvis and fore cannon bone (17% each). Of the racing fractures, 41% occurred after falling at a jump. Of all nonfatal fractures, at least 77% returned to racing. Of the TLI, 89% were of SDFT and 11% were of SL. Of all nonfatal TLI, at least 70% returned to racing.

It is interesting to note that the fracture rate during training, at 0.6 per 100 horse months, was considerably lower (by 36%) than that in flat horses presented by Verheyen and Wood (2004), where the rate was 0.94 per 100 horse months. The authors attribute this to 'differences in training regimens, training surfaces and the occurrence of jump schooling'. While the latter factor would be worthy of further investigation (it is intriguing to consider that it might be protective for flat race horses to do some jump training to improve bone strength), this does not sit well with the theories put forward that more high speed exercise is protective against fracture risk. Many jump trainers in the UK now use uphill training tracks for most of their work, where lower speeds are required to work the horses at high physiological work rates. Lower speeds will not elicit the high strain rates in bone that are considered necessary for improving bone strength.

Unfortunately the authors do not provide any information about the training terrain or intensities used by the trainers in the study, and it is perhaps surprising that this paper has been published with such important information not being available. Until such time, it is difficult to interpret the relevance of these findings in relation to optimal training regimes for bone health.

Singer ER, Barnes J, Saxby F, Murray JK (2008) Injuries in the event horse: training vs competition. Vet J 175:76-81.

There is little data currently available regarding the incidence and prevalence of injuries sustained by event horses. This study reported injury data from the 2002 season for British Eventing one day events (ODE) and FEI CCI competitions, and from the 2003 season during training for a CCI event.

This study is maybe the first that actually describes the fitness preparation of high level eventing horses in the UK for competition. It achieves this secondarily, in attempting to identify risk factors for injury in training. The authors deserve credit here since, compared to the more numerous studies that consider TB racehorses, they have actually attempted to describe the training variables used (even if, by their own admission, the data are 'poor'). They later conclude that more specific information is required in regard to how horses are trained and the impact on SDFT and SL injury.

During ODE competitions, the majority of injuries were superficial trauma (59%), a high proportion affecting the carpus and stifle. In the CCI group, significantly higher incidence of SDFT injury (17%) and exertional rhabdomyolysis (17%) were reported.

Of particular interest here is the report of the training of the CCI horses. Of the 403 being trained, 38% did not start. Of these, 61% (21% of the total) were unable to compete due to injury. These injuries were 43% tendon or ligament, 11% foot, 11% back, shoulder, pelvis, 5% exertional rhabdomyolysis, 2.4% EIPH and 27% other undiagnosed lameness/illness. In the majority of horses injured, the injury became apparent during (65%) or within 24 hours (21%) of a fitness work session.

When questioned, most of the riders (86%) stated that they used interval training for fitness preparation, with 34% using 3 bouts, 30% using 2 bouts and 24% using 1 bout of exercise. Since interval training necessitates repeated bouts of exercise with partial recovery in-between, these findings illustrate the misunderstanding of this form of exercise by many of the riders.

This study provides some very useful background information regarding the fitness preparation of eventing horses for competition, illustrating that despite the high incidence of training-associated injury, the knowledge of fitness regimes is moderate.

Early training and its consequences

It has been proposed that early training of horses might be beneficial for long-term soundness since certain musculoskeletal tissues are more capable of responding to loading during the growth phase than when they have fully matured. These studies are part of a continuing large scale series investigating outcomes of early exercise imposed on TB horses.

Rogers CW, Firth EC, Mcilwraith CW, Barneveld A, Goodship AE, Kawcak CE, Smith RKW, van Weeren PR (2008) Evaluation of a new strategy to modulate skeletal development in Thoroughbred performance horses by imposing track-based exercise during growth. Eq Vet J 40:111-118.

This report describes the protocol for conditioning foals from 3 weeks of age until 19-21 months when they were broken in. Exercise was imposed on an oval track 515 m in circumference with a sand over grass surface. Exercise was increased progressively from 5.36 m/s to 9.62 m/s with sprints included later on of 12.5 m/s. The cumulative work index (CWI) estimated that the conditioned group did 30% extra physical work over the control pastured group. Key conclusions were that neither body condition nor behaviour were significantly affected by conditioning, nor were there differences in clinical scores for joint effusions between conditions. Thus it appeared that the foals in this study were able to sustain the increased workload without suffering adverse affects. No claims were made that structure or function of musculoskeletal tissues were significantly affected.

Rogers CW, Firth EC, Mcilwraith CW, Barneveld A, Goodship AE, Kawcak CE, Smith RKW, van Weeren PR (2008) Evaluation of a new strategy to modulate skeletal development in racehorses by imposing track-based exercise during growth: The effects on 2- and 3-year-old racing careers. Eq Vet J 40:119-127.

This study followed on from the previous one and reported on the subjects' 2 and 3 year old racing careers. All horses were trained by the same trainer, blinded to the rearing history, using conventional Australasian methods. Key findings included the conclusion that early conditioning did not have any negative consequences for the 2 and 3 year old careers. The workload required to reach race fitness was not different between groups, although the conditioned horses tended to reach race fitness earlier. There was no difference in the incidence of orthopaedic problems but the conditioned individuals tended to show signs later than those pasture reared only. There was no significant effect on race performance of early conditioning. The authors accept that with the small numbers of the study few hard and fast conclusions could be drawn, but their study shows some encouragement for early conditioning and did not show any detrimental effects. It is interesting to note that only one 2 year old developed sore shins, and the authors attributed this to the progressive increase of training load during the training program. It might also suggest that the training did not closely represent real commercial training since a higher incidence would have been expected.

Muscle training

Rivero JLL (2007) A scientific background for skeletal muscle conditioning in equine practice. J Vet Med A 54:321-332.

This is a review of the current state of knowledge of muscle conditioning in athletic horses. It provides a useful summary of the studies that have provided an understanding of muscle responses in a range of breeds including the TB.

Rivero JLL, Ruz A, Marti-Korff S, Estepa J, Aguilera-Tejero J, Werkman J, Sobotta M, Lindner A (2007) Effect of intensity and duration of exercise on muscular responses to training of Thoroughbred racehorses. J Appl Physiol 102:1871-1882.

There are few studies in TB horses that provide useful information on how different durations and intensities of exercise promote muscular responses.This study aimed to measure the effects on short term muscle adaptation of running TB horses at speeds eliciting blood lactates of 2.5 (v2.5) and 4 mmol/l (v4) for 5, 15 and 25 min. This used a novel approach of a 6 × 6 randomised Latin Square design for each horse experiencing each period of 22 days training under each combination of speed and duration, with 10 days rest in-between each. Muscle responses were quantified by evaluating fibre type ratio changes and changes in myosin heavy chain (MHC) isoforms, fibre hypertrophy, oxidative and glycolytic enzyme activities, and capillarisation. Both intensity and duration of exercise were critical in determining responses. Overall it appeared that changes in fibre types IIx to IIa and hypertrophy were more dependent on intensity, whereas oxidative capacity and capillarisation were more dependent on duration of exercise.

The authors deserve credit for putting their results into perspective of real-life TB training. Their suggestion is that the aerobic capacity of muscle can be developed in young TB horses by moderate intensity exercise, whereas higher intensities of exercise for the same duration will develop strength and power.

This might potentially include use of lactate-guided training, or use of other monitoring methods. However, Figure 5 shows plasma lactate curves for a horse on my gallops and illustrates that v2.5 and v4 from this plot would not be appropriate intensities for up to 25 min of exercise. Even at v2.5, the speed was approximately 12 m/s (27 mph), which could not be sustained for that time. This compares with a speed of approximately 8 m/s in their treadmill study.

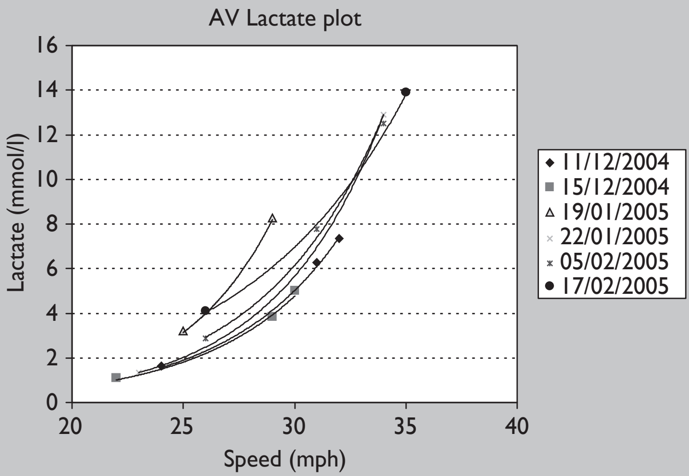

Figure 5. Plasma lactate curves.

Comment of the editor

Good point! Rivero *et al.* guided the exercise intensity of their horses by blood lactate concentrations determined with a standardized exercise test done before commencing a conditioning period. The blood lactate concentration after the exercise was not considered at all. This is important because this approach has to be differentiated from another way of guiding exercise using blood lactate concentrations which is to run horses at defined speeds (and duration of exercise) and then measuring the blood lactate concentration after exercise aiming at maintaining the workload of the horses within defined ranges of blood lactate concentration. (This would be the approach of the author.)

Furthermore, it has to be created awareness towards the effect of the standardized exercise test prescription on the blood lactate concentrations (Lindner A 2007) News on equine sports science (2005-06). In: Applied equine nutrition and training – ENUCO 2007. Lindner A (ed). Wageningen Academic Publishers, Wageningen, the Netherlands. Pages 173-189). Thus for example, when advising on conditioning horses at their v4 for 15 minutes it is mandatory to also use the same or a very similar exercise test prescription as the one used in the study!

Detraining

Graham-Thiers PM and Bowen LK (2009) Ability to maintain fitness improved during large pasture turnout. J Eq Vet Sci 29:430-432.

This short communication is of interest since it aimed to examine the effect on fitness of turn-out into a large paddock versus stable rest or stable rest with moderate exercise. While horses at stable rest lost fitness over a 14 week period, indices of fitness during and after a standardized exercise test at pasture turnout appeared to improve, as did bone density scored radiographically. These findings will be of interest to those who might consider the management of horses during breaks from training and even those considering turn out for horses while they are in training.

Gastric ulceration and performance

Nieto JE, Snyder JR, Vatistas NJ, Jones JH (2009) Effect of gastric ulceration on physiologic responses to exercise in horses. Am J Vet Res 70:787-795.

Gastric ulceration is a ubiquitous problem in all types of domesticated horse and particularly those involved in competitive sport. It has long been known that the condition can be responsible for poor performance. This study is the first to illustrate that this may have a contribution from impaired physiological responses to exercise and not only the horse being affected psychologically through discomfort or general malaise. Here the authors used a group of treadmill exercised horses as their own controls by using an exercise protocol to induce ulceration and omeprazole to eliminate the condition reversibly. The principal finding was that mass specific oxygen consumption increased to a lesser extent in the horses when they were affected by ulcers than when not. This failure to improve aerobic capacity with training would undoubtedly have a detrimental effect on performance (and did so here although not significantly), but the underlying reasons were not determined. It was interesting to note that stride length increased during training in both conditions but to a lesser degree in the ulcer affected horses (2.3 vs. 4.8%). This might have affected tidal volume and hence alveolar ventilation for a particular speed of exercise.

Stride characteristics

Ferrari M, Pfau T, Wilson AM, Weller R (2009) The effect of training on stride parameters in a cohort of National Hunt racing Thoroughbreds: A preliminary study. Eq Vet J 41:493-497.

There is currently a good deal of interest in regard to aspects of locomotion and stride strategy of exercising horses. This is an area of study that has so far received relatively little attention. This investigation examines if stride characteristics in NH TB racehorses were affected by training. While there were significant differences in stride parameters before and after training, these were of very small magnitude (at 11 m/s SF increased from 2.160 to 2.167 Hz after training), equivalent to a SL decrease of approx 2 cm on a stride of 5.09 m. However, although this was a statistically significant difference, is it likely to be of biological or practical significance? This issue was not addressed by authors. It is interesting that this change in stride length was in the opposite direction and to a lesser extent than that measured by Nieto *et al*. in the treadmill study discussed above.

Nankervis K, Hodgins D, Marlin D (2008) Comparison between a sensor (3D accelerometer) and ProReflex motion capture systems to measure stride frequency of horses on a treadmill. Comp Exerc Physiol 5:107-109.

This brief communication is relevant because it is the first publication to provide an initial validation of a novel accelerometer device designed specifically for ease of use in studying aspects of equine gait. The 'Pegasus System' consists of a triaxial accelerometer coupled with a data logger. This study illustrates that it provides accurate information of stride frequency in horses at different gaits on a high speed treadmill when compared to an optical gait analysis system. Further studies are in preparation that provide a full validation of its use for horses during overground locomotion when combined with a GPS receiver.

Hoof-ground interaction

The basis for these studies was that the effects of different types of track surfaces on which high speed exercise is performed are poorly understood, despite the widespread use of many different products.

These studies are part of a larger project to study the effects of different surfaces on hoof acceleration, ground reaction forces, tendon load and limb and trunk kinematics.

Chateau H, Robin D, Falala S, Pourcelot P, Valette JP, Ravary B, Denoix JM, Crevier-Denoix N (2009) Effects of a synthetic all-weather waxed track versus a crushed sand track on 3D acceleration of the front hoof in three horses trotting at high speed. Eq Vet J 41:247-251.

This study investigated the effects of two different but commonly used training surfaces, crushed sand and a waxed sand surface, on hoof acceleration. A triaxial accelerometer was mounted on one hoof and measurements made of three horses trotting at 10 m/s over both surfaces. Repeated measurements made on the same horse showed good repeatability although there were large differences between horses and between strides. There were large differences between tracks. The waxed track appeared to reduce the amplitude of the deceleration at impact by about 50%, reduced the vibrations associated with impact and caused a more gradual deceleration of the hoof during the braking phase. These findings would suggest that the waxed surface subjects the distal limb to less mechanical stress than does the crushed sand and so might be a better training suface. However overall performance might be impaired since hoof acceleration at breakover was reduced, as was stride length on the waxed surface.

Robin D, Chateau H, Pacquet L, Falala S, Valette JP, Pourcelot P, Ravary B, Denoix JM, Crevier-Denoix N (2009) Use of a 3D dynamometric horseshoe to assess the effects of an all-weather waxed track and a crushed sand track at high speed trot: Preliminary study. Eq Vet J 41:253-256.

This study employed a shoe instrumented with four triaxial piezoelectric transducers for measuring ground reaction forces in three planes. The measurements were made at the same time as those in the above study. The results corroborated those above in that there were smaller vertical and horizontal forces at impact on the waxed surface, suggesting that it had better shock-absorbing properties. In addition, the amplitude of the maximum horizontal breaking force was lower and occurred at a later stage. These findings would suggest that the waxed surface might be a safer surface on which to train horses.

Together these studies illustrate that the accelerometer and dynamometric horseshoe are tools with adequate sensitivity to quantify the effects of different track surfaces on biomechanics of the distal limb in horses moving at high speed. It is clear that tools such as this should be utilized on a range of track surfaces used already in SB and TB racing, and on different training track options. They could possibly also be used on turf to quantify the differences of states of going on hoof-ground interaction and provide a more objective measurement of 'going'.

Conclusions

A review of studies published over the past three years provides a good deal of information relevant to those involved in managing horses in training for high intensity competition. It is sobering to consider that we have been aware of high injury rates in racehorses for over 20 years, yet there has been no improvement in these statistics during that time. While we have a greater knowledge of some of the important risk factors, many of the recent studies illustrate the very wide range of incidence of injury between trainers. An essential element here must be the differences in training regimes used. However, many within the academic community appear largely unaware of exactly how horses are trained. This area clearly needs much wider study.

Recommendations have been made in regard to maximizing bone health in racehorses. Reducing the volume of 'canter' work and increasing that of 'gallop' has been suggested, and there is some corroborating evidence from other studies for this approach. However, more useful guidelines will be needed before these recommendations are of much practical benefit. On the other hand, some very useful information has emerged in relation to muscle training, the recommendations from which might be viewed as contradicting those regarding bone fitness. At least the authors here made every effort to put their suggestions into a practical framework, by using blood lactate concentrations to guide training intensity. Many researchers should take note of this helpful approach.

One issue that has not been addressed at all is the behavioural effects of different conditioning programs for high intensity exercise. The suggestions regarding more 'gallop' work early in training might well have consequences on young horse behaviour. With the advent of 'equitation science' in recent years, maybe we can look forward to such studies in the near future?

It has been of interest to have the differences confirmed in incidence of injury in young (flat) vs. older (NH) TB racehorses. Young horses are clearly at greater risk of bone injury (stress fractures, sore shins) than are their older NH counterparts, who are at greater risk of tendon and ligament injury. This goes hand-in-hand with our growing knowledge of how tissues are affected by repetitive high load exercise. It is of note that it would appear that the risk of fracture in training in the older horses was considerably lower than that of the younger ones. A series of studies is well under way to determine if early conditioning from birth will help ameliorate the high rates of injury.

Not before time, the biomechanical properties of different track surfaces are starting to receive more attention. Many products are marketed for training and racing, but there is no objective data available on their safety. These studies illustrate that such data may become available in the foreseeable future. One of the greatest controversies in UK racing is the reporting of grass track condition. Neither penetrometers nor the 'Goingstick' which is currently used on all UK turf tracks provide reliable objective data and there is so far no means for quantifying going on synthetic surfaces. Racing administrators are guilty of failing to address this issue adequately and this area needs much greater attention from the academic community.

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Effects of diet and feeding regimens on performance

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Virtually all individuals that compete with their horses want the performance of their animals to be as good as possible. While genetics plays a large role in whether their horse will likely be successful, their feeding program can certainly have a tremendous impact. A good feeding program will not make a horse with poor genetic potential a star athlete, but a poor feeding program can make a horse with good genetic potential not able to achieve its maximum potential. A great deal of equine nutrition research that has been down through the years has focused on improving performance and has provided valuable guidance to develop sound nutrition for athletic horses.

Energy

The biggest nutritional difference between exercising and non-exercising horses is the increased energy demands that are typically associated with exercise. While a short-term inadequacy of the intake of most nutrients will not have a noticeable impact on a horse's performance, a horse that does not regularly consume an adequate amount of energy will fatigue faster and not perform as well compared to one that is consuming sufficient quantities. What frequently determines how a horse performs in competition is how quickly it fatigues (Worth, 1985). While there are many biochemical and physiological reasons why fatigue occurs, one of the reasons is a depletion of glycogen stores that could be used to generate energy. Some research has suggested ways to increase glycogen stores through dietary manipulation such as feeding diets containing elevated amounts of oil or fat (Potter *et al.*, 1992; Geelen *et al.*, 2001), but before a person starts to worry about which form of dietary energy is needed, it is important that they simply determine if their horse is receiving enough dietary energy.

Determining whether requirements are being met for most nutrients is difficult without analyzing the diet and comparing the contents to the animal's nutritional requirements. However, it is possible to evaluate whether energy requirements are being met simply by examining the animal. Some individuals suggest that an animal's behavior can indicate if they are getting too much energy in their diets or the wrong type of energy (for example, some people consider corn to be a 'hot' feed and causes their animals to be unruly). Behavior problems involving an unruly horse are more often related to a horse not enjoying the work it is being asked to do – often because of a training or soreness issue. In contrast, a horse that has a 'lack of energy' and seems sluggish certainly might not be receiving enough calories to meet its energy demands.

Rather than relying on behavior and attitude to determine if energy requirements are being met without being exceeded, a better method is simply to body condition score (BCS) the horse (Henneke *et al.*, 1983). The BCS can be a useful way to determine if the horse is meeting its caloric requirements without exceeding them. A horse that has a decreasing BCS is using more calories than it is taking in and a horse that has an increasing BCS is taking in more calories than it is using. This is the simplest and probably best way to see if you are meeting the energy requirements.

Even if a horse's BCS is not increasing or decreasing, it does not mean that the animal is at the ideal BCS. For instance, a horse can be at a constant BCS but that BCS could be too high or too low. A BCS between 4 and 6 should be the goal for most athletic horses (Becvarova *et al.*, 2009) as some research has suggested that glycogen stores are not maximized when horses are in a thin condition (Jones *et al.*, 1992; Scott *et al.*, 1992). Simply put, the 'gas tank is not full'. Considering glycogen is a preferred substrate for energy production during short-term, high-intensity anaerobic exercise such as in Thoroughbred racing, cutting, or jumping, having a 'full gas tank' will allow the horse to perform for a longer time without slowing down. As the duration or length of competition increases, there is reason to believe that a somewhat lower BCS could be an advantage. During most equine competitions, there is insufficient time to mobilize significant amounts of body fat stores for energy. As a result, higher body fat stores act simply as additional weight that the animal needs to carry around without providing much benefit in terms of immediate energy supply. That is why it is not uncommon to see endurance horses competing at distances of 100 miles or greater with a BCS under a 5 and still doing quite well as compared to ones that have a greater BCS (Lawrence *et al.*, 1992). Similarly, the body mass index (the system that suggests what an ideal weight is for a given height) of elite human marathon runners would be considered to be low by most standards – though obviously having such a low body fat content does not impair the performance of those distance runners.

Slight over-consumption of most nutrients (but not so much as to cause a toxicity) can simply be wasteful and usually is unlikely to impair performance. However, regularly consuming too much dietary energy will likely result in excessive fat deposition. That extra body fat can certainly be detrimental to performance. A horse that is greater than a BCS of 5 is carrying around extra weight and that will impair the performance of most athletic horses. For example, a Thoroughbred racehorse at 500 kg with a BCS of 6 probably weighs about 16 to 20 kg more than a similar-built horse at a BCS of 5. Thus, it is easy to imagine what a difference that could make to its race performance. Other issues that arise because of horses being too fat include an increased risk of injury due to the extra weight they carry causing greater forces on their legs. Additionally, the extra fat serves as insulation and makes it challenging for the horse to dissipate heat (Morgan, 1997). While running out of substrate to generate energy is one reason horses fatigue, overheating is another. When muscle temperatures get too high, enzymes involved in muscle contraction no longer work as efficiently – effectively limiting how much work the animal can do. While carrying extra fat is beneficial to animals living in the artic because of its insulating properties, it certainly can decrease the performance of an athletic horse. As previously mentioned, muscle glycogen seems to decrease when a horse is thin, but it does not seem to increase when a horse progresses from a moderate BCS to a fat BCS. Thus, there is seemingly little benefit to athletic horses to be much above a BCS of 5. In contrast, one could make an argument that some horses do fine having a BCS a bit lower than a 5 – especially if the distance they compete at is great enough. That being said, it is often a challenge to get enough calories into those hard working horses and it is not being suggested that one try to restrict the feed given to athletic horses in order to achieve a BCS much less than 5 as that certainly could impair performance.

When it comes to nutritional requirements, meeting the energy requirements of athletic horses can be much more challenging than meeting the requirements for other nutrients. It is quite simple to meet the requirements for protein, minerals, or vitamins simply by altering what you feed the horse or supplementing more of those nutrients into their ration. However, with energy, often a horse cannot consume enough calories to meet their energy needs, particularly with a high forage diet (Harris, 2009). Unfortunately, not providing enough dietary energy can have a relatively immediate impact on performance. The positive thing is that energy is the one nutritional requirement where one only needs to monitor their animal to see if the dietary energy intake is matching the energy expenditure. If they match, and if the BCS is in the appropriate range, it increases the chance that your horse will perform well.

The inclusion of dietary fat (or oil) has been one of the biggest changes witnessed in the feeding of horses in the past three decades. While many horses can benefit from the inclusion of fat into the diet to reduce the amount of soluble carbohydrates being fed, athletic horses can benefit the most from dietary fat. One undisputable benefit is the increased caloric density of fat as compared to carbohydrates. As a result, it allows an animal to meet its dietary calorie requirement while consuming a smaller quantity of feed. This becomes especially beneficial in intensely exercising horses that have a high calorie requirement that they might not be able to meet if consuming a traditional diet of hay with a couple of kilograms of a commercial grain mix. Without the inclusion of fat, sometimes horses such as these are simply unable to eat enough feed and end up losing weight as a result.

Another benefit of feeding fat is that it is a safer method to provide large quantities of calories than feeding large quantities of grain containing substantial amounts of starch. When too much starch from grain is provided, it can escape undigested from the stomach and small intestine into the hindgut (Julliand *et al.*, 2006). When there, the microbial population has access to it and the fermentation of these products could lead to problems with colic or laminitis. By contrast, if excess fat were to escape undigested into the hindgut, similar microbial action would not take place, thus preventing the horse from being put in danger.

Additionally, the fermentation of carbohydrates can be accompanied by the release of a substantial amount of heat. While this can be beneficial in keeping a horse warm in cold climates, the need to get rid of extra heat can be challenging for athletic horses, especially as they produce tremendous amounts of heat while exercising or are in a hot climate. As a result, by reducing the carbohydrate fraction of the diet by replacing calories from carbohydrates with those from added fat, less heat is produced in the hindgut. This makes it easier for a horse to cool itself (Potter *et al.*, 1990; Kronfeld, 1996).

One more benefit of feeding fat is debatable, but worth considering. Some research has suggested that inclusion of fat into the diet of athletic horses at the rate of up to 10% of the total diet has the potential to have a glycogen-sparing effect (Treiber *et al.*, 2008). As horses perform their daily work, most of it is conducted aerobically (at a slow pace). During that exercise, the fat from the diet, which requires oxygen in order to utilize it for energy production, could be used preferentially instead of glycogen. By virtue of using the fat from the diet on the 'slow' days, glycogen is spared for use on days when competition at a high rate of speed is needed.

Regardless, it needs to be mentioned that a large amount of calories typically comes from the roughage portion of a horse's diet. There are times that athletic horses can obtain all of their energy requirements (and other nutrients) strictly from the roughage they are provided. However, the ability to meet the energy requirements through roughage alone typically decreases as the intensity of training increases. While horses seem to have less health and behavioral issues when on a high roughage diet, there are many athletic horses that simply cannot meet their requirements unless a more energetically-dense concentrate mix is provided.

Protein requirements

Many individuals believe that protein requirements increase dramatically when horses enter athletic training due to the anabolic changes to muscle that often occur at the commencement of training and the need to repair muscle damage or increase muscle hypertrophy (Custalow, 1991). Though protein requirements do increase with training, the increase is not as dramatic as most people would believe. Often, the additional protein needs are met when the horse consumes more feed to meet the increase in caloric requirements. However, if one chooses to use added-fat to meet the increased demand for calories, feed intake may not increase and the requirements for protein may not be met unless the concentration of protein is increased in the feed.

To determine whether a horse's protein requirements are being met, one must determine how much protein the animal is receiving. To do that, the amounts of protein received from all segments of the diet must be determined – including both the concentrate (grain) and roughage portions. Too often, horse owners consider only the amount of protein received from the concentrate the horse is fed and ignore the amount received from pasture or hay. Given that the amount of protein in good quality hay (particularly legume hay) can be substantial, having a concentrate high in protein is often unneeded for an athletic horse. However, the only way to fully determine if a horse is receiving sufficient protein is to calculate the amount provided by the concentrate and roughage and compare that to the animal's requirement.

It should be stated that the horse really does not have a protein requirement, but an amino acid requirement instead. Unfortunately, there is a lack of research on the amino acid requirements of horses (Antilley *et al.*, 2007). This is especially true for the exercising horse. Until such research is conducted, it is not possible to accurately say what are the true amino acid requirements for exercise.

Mineral requirements

The mineral requirements of athletic horses can vary dramatically depending upon several factors. These can include stage of training, management of the animals during training, and the intensity of training (Nielsen *et al.*, 1997). Often calcium is the first mineral that is thought of and its importance to bone is well known. However, what is often not recognized is that bone is a very dynamic tissue that responds fairly rapidly to forces that are applied to it. When horses are sprinted (whether free-choice or forced), bone responds by either becoming stronger or, if already strong, maintaining its strength. By comparison, when there is no high-speed exercise, there is no demand for bone to be strong so the bone does not increase in strength (having bone strength maximized is important for horses that compete at high speeds such as race horses or that place large loads on their legs such as jumpers – it is less critical for horses that only compete at slow speeds). During training periods when the amount of high-speed exercise is increasing, the demand for calcium is greater. By decreasing the forces placed upon the skeleton (such as by stalling without any high-speed exercise) the demand for calcium would typically be decreased (Nielsen *et al.*, 1998). This is just one example of how mineral requirements can vary substantially depending upon what the animal is doing. Guidelines established by the NRC (2007) attempt to ensure minimum amounts are met for most minerals regardless of stage of exercise and the type of management used. To determine if the required amounts are being fed, it is important to analyze the feed being fed. Generally, if a commercial blended concentrate is being fed to an athletic horse, the chance for that animal being deficient in minerals is decreased.

Exercising horses have a greater requirement for electrolytes (sodium, chloride, and potassium). This increased requirement is primarily associated with needing to replace electrolytes lost in sweat (Coenen, 2005). Thus, quantifying additional requirements needed for exercise primarily requires determining how much sweat is lost during exercise. The best way to do this is to weigh a horse before and after an exercise bout as the weight loss reflects primarily sweat loss. By using the concentrations of the electrolytes found in sweat, estimates for the amount of additional electrolytes required can be determined. It is recognized, however, that weighing the horse each exercise bout is impractical so guidelines are provided by the NRC 2007 depending upon the perceived intensity of exercise that the horse is performing. While being very subjective, it also does not take into consideration the ambient conditions in which the exercise takes place. For instance, a horse exercising in a very hot climate will sweat more (and thus have greater requirements for electrolytes) than one exercising at the same intensity in a very cold climate. Fortunately, if a horse is allowed free access to salt (sodium chloride) and also is fed a good quality forage (typically high in potassium), most horses will consume enough electrolytes to meet their needs. Although there are certainly cases, especially with extremely hard working horses in hot climates, where this is not the case and additional electrolyte supplementation is necessary. Regardless, it is critical that adequate water be available to a horse to allow excretion of extra electrolytes if excess is consumed.

Vitamin requirements

If being fed good quality hay or allowed access to pasture and exposed to sunlight, it is unlikely a horse will be vitamin deficient. Furthermore, commercial, blended concentrates that are often fed to athletic horses are typically fortified with vitamins. Such fortification greatly decreases the chance of a deficiency being present. That being said, it is speculated that in some hard working horses that have quit eating, supplementation with B-vitamins, particularly thiamin, may help restore their appetite and improve energetic metabolism (Carroll *et al.*, 1949; Wolter, 1987). While normally synthesized by the microflora in the hindgut in sufficient quantities, given the requirement in certain co-factors involved in energy metabolism, supplementation in hard working horses may be justified if an animal's appetite seems to be depressed.

There has been some research into the role certain vitamins play as anti-oxidants in the exercising horse – particularly those competing in endurance competitions. It is unclear at this time if supplementation with these vitamins in a horse fed a balanced diet is warranted.

Timing of meals

Obviously, providing a balanced diet to maximize athletic performance is critical. Some have suggested that timing of feeding can also influence performance (Harris and Graham-Thiers, 1999). For instance, feeding a grain meal within three hours of performance appears to result in elevated insulin and decreased glucose concentrations. Feeding hay with the grain appears to blunt the response but did not prevent the decrease in blood glucose. By contrast, feeding the meal 8 hours prior to exercise did not seem to have the same effect. One could question whether the psychological response to actual performance might override any negative effects on blood glucose that accompany feeding a grain meal within a few hours of competition. Even if that were true, other physiological changes that accompany meal feeding may influence performance. For instance, up to a 24% loss of plasma volume within one hour of feeding has been reported (Clarke *et al.*, 1990). Pagan and Harris (1999) concluded that grain should be withheld from horses before exercise but small quantities of hay should be fed.

While meal-feeding is a practice that tends to be inherent when athletic horses are maintained in stalls, it is critical that caretakers of horses recognize that such a practice is not ideal for the horse. If grain is to be fed, the amount should be divided into as many small meals as is practical. Doing so helps to eliminate boredom and greatly reduces the risks associated with grain overload and likely diminishes any detrimental effect a meal may have on performance.

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Evaluating recent equine research

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Each year, much research is done in the area of equine nutrition to better refine the nutritional requirements of horses. While important, it is rare for dramatic changes in nutrient recommendations to be suggested based upon that type of work. Other studies examine feeding management techniques. The first paper addresses an approach to feeding horses that would be dramatically different than would be commonly recommended for horses but that would be the commonplace for other livestock species. Other research tends to be focused on evaluating different forms or sources of nutrients. This type of research often has great appeal to those in the equine industry, as individuals are interested in providing nutrients to their animals in a form that can be best utilized. Two of the papers that will be discussed have taken that approach. The final paper examines whether a commonly used supplement is efficacious. With most research papers, there are limitations to the studies that may not be clear to a casual reader. Besides bringing your attention to some new research, a further goal is to illuminate what some of these limitations may be to encourage similar scrutiny when evaluating other research.

Henneke DR, Callaham JW (2009) Ad libitum *concentrate intake in horses. J Eq Vet Sci 29:425-427.*

Objectives

To establish if horses with *ad libitum* availability of a moderate energy dense concentrate and hay would self-regulate feed intake.

Why chosen

This study illustrates that horses are similar to other livestock species in that they can be provided an unlimited amount of grain without dramatic health problems if acclimated to the diet appropriately.

Background

Domestic horses are often fed grain meals twice a day in addition to hay. It is commonly recognized that providing too much grain in a meal can lead to health problems such as colic or laminitis. While most other domestic livestock species can be fed unlimited quantities of grain, such a practice is rarely attempted with horses and there are no known published studies that have specifically examined whether such a practice can be done.

Overview of the study

Two trials were conducted to examine this issue. In the first trial, six aged geldings were removed from pasture and put into stalls two weeks prior to the onset of the study. They were fed about 3.2 kg of a 12.5% CP, 20% CF, and 3.5% fat pelleted concentrate and 9 kg of hay per day divided into two equal feedings. Horses were weighed and body condition scored throughout the 35-day trial. During the second week of the trial, the amount of concentrate was increased at a rate of 5% per day per horse and the volume of hay was increased to provide unlimited access. At day 15, horses had unlimited hay and grain throughout the duration of the trial. In the second trial, six horses were removed from pasture where they had not been receiving grain, divided into two groups and placed into two dirt paddocks. During the first week of the trial, horses received 2 kg/head/d of concentrate. In the second week they received 3 kg/head/d, and in weeks three and four they received 4.5 and 6 kg/head/d of concentrate, respectively. Unlimited access to grain began in week five and continued until the end of the 133-day trial. A round bale of coastal hay was available to horses in each pen. At the onset of week 14, horses were moved to a 5.5 hectare pasture for the remainder of the study.

Main findings

In the first trial, average consumption of grain began at 5 kg/hd/d, peaked at 19 kg on day 25, and then declined to 10 kg on day 35. Average hay consumption began at 6.8 kg/hd/d and declined to 1.0 kg. Total feed consumption began at 2.5% BW (1% concentrate, 1.5% hay), rose to a peak of 3.25% BW (2.5% concentrate, 0.75% hay) on day 25, and declined to 2.4% BW (2.1% concentrate, 0.30% hay). The body condition score of the horses increased from 5.8 at the beginning of the study to 6.7 at the conclusion. In the second trial, during the paddock phase, average daily concentrate consumption peaked at 22.45 kg during week 6, declined to 15.46 kg during week 8, increased to 20.5 kg on week 12, and then declined to 16.15 kg during week 14. When moved to pasture, concentrate consumption declined to 5.5 kg during week 18. Concentrate consumption averaged 1.2% BW during week 4, rising to 3.3% during week 7, and declined from 2.5% to 1.5% during the time the horses were on pasture. During this study, horses gained nearly 50 kg and body condition score increased from 4.7 at the beginning of the study to 6.5 at the completion.

Practical interest

Most equine nutritionists would not encourage allowing horses *ad libitum* access to grain for fear of horses developing colic or laminitis. This study demonstrates that horses can be gradually adjusted to such a diet without encountering such problems. This is probably not all that surprising as foals are often creep-fed in a similar fashion (i.e. allowed unlimited access to grain) without problems. Typically, only if a creep-feeder has been allowed to become empty and foals gorge themselves when it is refilled are health issues encountered. Likely, if one were to continually allow unlimited access to grain throughout a horse's life, horses would respond no differently than other livestock species on a free-choice grain diet.

Comments on the study

Though this study demonstrates one can provide free-choice grain to horses, one can still question whether it is advisable to do so. Though the average feed intake typically stabilized at around three weeks, the body condition score of horses in both studies ended up being greater than what most individuals would suggest as being ideal for the majority of horses. There are times when such an approach may be appropriate though. While the authors suggest this approach may be a safe and reasonable alternative to meal feeding in maintenance or lightly exercise horses, I would argue that most of those horses may not need a meal of grain anyway. In contrast, hard working athletic horses that have a high energy demand may be able to benefit from such an approach. Those horses are often fed large amounts of grain in meals and they may be better off having grain in front of them at all time. However, this would seemingly only apply if their body condition scores do not become elevated past a point where it hinders performance. Further, while no problems other than having excess body fat was observed in the study horses, this does not guarantee that problems would not be seen if tested in a larger population of animals. For example, if this study was replicated with ponies or horses that were insulin resistant, health problems such as laminitis might become apparent. Hence, simply because there were no health problems with these twelve animals, it does not guarantee complete safety. It simply illustrates that nutritional management of this type can be done without serious health issues in at least some animals.

Besides that observation, this study demonstrates that while intakes may initially increase dramatically when horses are allowed unlimited access to feed, high intakes (i.e. greater than 3% of BW) will likely not persist after horses become acclimated to the new diet. Thus, if feed intake is only measured at one point, it may not adequately reflect feed intake over a larger time period.

Also, it was interesting to note that while consumption of grain was extremely high at times when horses were consuming hay, the grain consumption decreased to what would be considered a more traditional amount when horses were allowed access to pasture. Most horse owners have noted that even when plentiful hay is available, horses will often ignore that in order to graze fresh pasture – even when the quantity of forage in the pasture would not be adequate to sustain them. It would appear that this study confirms just how strong the desire to graze is as they will eat less grain to consume more fresh pasture when it is available. Along those lines, it is worthy to note that all horses did consume at least some forage each day. That being said, this study would suggest that studies could safely be designed that would allow concentrate-only diets to be fed that would allow the digestibility of nutrients from grains to be calculated. Currently, the digestibility of nutrients from different forages can be calculated by feeding forage-only diets. This study suggests the same can be done for grains, which would allow more accurate diet formulation by more precisely estimating digestibility of all components of a ration.

O'Connor CI, Nielsen BD, Woodward AD, Spooner HS, Ventura BA, Turner KK (2008) Mineral balance in horses fed two supplemental silicon sources. J Anim Phys Anim Nutr 92:173-181.

Objectives

To investigate the effects of two different silicon (Si) supplements on mineral absorption and retention in horses.

Why chosen

Many companies are adding supplemental Si to supplements as a method to improve bone and cartilage health. However, many of the Si sources have had little research done on them to ensure, at the very minimum, that they can be absorbed.

Background

Silicon has been shown to be required for normal growth and development. In the early 1990's, one study showed decreased injuries rates in horses in race training when providing a supplement that contained Si that could be easily absorbed (Nielsen *et al*., 1993). While Si is common in the environment, most of it is relatively unabsorbable. Hence, it is critical to know if a Si source can be easily absorbed. If it is relatively unabsorbable, it is unlikely that it would have any effect on injury reduction.

Overview of the study

Eight geldings were randomly placed in one of two groups: control or supplemental Si, which was provided by one of two supplements.

The first, sodium aluminum silicate (SA), contains a bioavailable form of Si and is high in aluminum (Al). The second supplement contains oligomeric orthosilicic acid (OSA). All horses received textured feed and had *ad libitum* access to hay. Supplemented horses received either 200 g of SA or 28.6 ml of OSA per day to provide an equivalent amount of Si. Following a 10-day adaptation period, the horses underwent a 3-day total collection. Blood samples were taken on days 0 and 13. The two balance studies were conducted four months apart to reduce carryover effects.

Main findings

Intakes of Al and Si were greater with SA supplementation than were received by the controls. Sodium aluminum silicate increased fecal and urinary Si excretion. Additionally, Ca retention and apparent digestion were also increased by SA. Supplemental OSA increased retention of Ca and boron (B) and apparent digestion of B. OSA tended to increase Si retention and apparent Si digestion. Both SA and OSA supplementation resulted in higher plasma Si concentrations than was seen with the control groups.

Practical interest

Sodium aluminum silicate is the only Si supplement shown to reduce injuries to performance horses (Nielsen *et al.*, 1993) though it did not, despite popular claims, reduce the incidence or size of osteochondrotic lesions in blinded, placebo-controlled study (Turner *et al.*, 2007). However, feeding it also results in relatively high amounts of Al being consumed which has been shown to result in increased Al being deposited in various tissues in dairy calves (Turner *et al.*, 2008). This increase in tissue Al would be considered to be undesirable by many. Supplementing with OSA allows a bioavailable form of Si to be provided without accompanying high amounts of Al.

Comments on the study

The results of this study could certainly be considered positive for the supplementing of OSA as a means of providing additional Si to horses. However, it would be wrong to assume that just because Si was absorbable, OSA provides a means to reduce injuries in performance horses. The study conducted with SA was an FDA-blinded and controlled study utilizing 53 horses. Horses were put into race training and each horse was raced for a total of nine races or until they experienced some type of injury necessitating removal from training. The results of that study showed a reduction in injuries to supplemented horses and it suggests providing supplemental Si may have been the reason for the reduction. However, the additional dietary Si came from sodium aluminum silicate (sodium zeolite A) which is a molecule containing other elements besides Si. Hence, while it likely is the Si causing the beneficial effects (based upon known roles that Si plays in bone and cartilage development), it is wrong to automatically assume that such is the case. Hence, though OSA had beneficial effects on Ca retention and digestion, and supplementation of OSA resulted in greater plasma Si concentrations than were reported in the control group, it is wrong to assume that such positive findings will automatically result in reduced injuries to performance horses when they are supplemented with it.

There are many silicon-containing supplements currently being marketed with the intent of reducing injuries. Only a few contain SA and even fewer contain SA in a dosage that was deemed to be optimal through previous work. While some of the products contain Si previously shown to be unavailable (though this would likely be unknown to consumers), there are some products that contain Si that may be beneficial (such as OSA). However, until efficacy studies are done on these compounds that actually show they reduce injuries, it is wrong to assume they do simply because they contain Si.

Calamari L, Ferrari A, Bertin G (2009) Effect of selenium source and dose on selenium status in mature horses. J Anim Sci 87:167-178.

Objectives

To determine the effects of either dietary selenium (Se) source or dose on the Se status of horses.

Why chosen

This study illustrates that the source of a mineral can influence the status of that mineral. However, other than providing evidence to suggest one source is more available than another, the study fails to show any marked benefits in health factors.

Background

A clear dietary need for Se exists in the horse. While often incorporated into the diet through the use of sodium selenite, it can also be provided through an organic form produced by growing specific strains of yeast in a Se-enriched environment. In this process, selenomethionine is the predominant form produced. While inorganic Se, such as that found in sodium selenite, is absorbed through passive diffusion, selenomethionine is actively transported through intestinal membranes. This suggests the absorption of Se can be enhanced when the Se is provided as selenomethionine.

Overview of the study

Twenty-five mature horses were blocked by bodyweight and allocated to one of five treatment groups. The groups included a negative control receiving 0.085 mg of Se/kg of DM from the basal diet, three groups receiving supplemental organic Se (0.2, 0.3, and 0.4 mg of total Se/kg of DM), and a positive control receiving 0.3 mg of total Se/kg of DM that received the supplemental Se as sodium selenite. Horses were adapted to the basal diet for 56 days and then were on their respective treatments for 112 days. Blood samples were taken on d 0, 28, 56, 84, and 112. Whole blood and plasma were analyzed for total Se and glutathione peroxidase activity (GPX-1). Plasma samples were analyzed for the thyroid hormones (thyrozine and triiodothyronine).

Main findings

Total Se in blood and plasma and GPX-1 activity were greater in all supplemented horses, with a linear dose effect of Se yeast for whole blood and plasma Se and a quadratic dose effect for whole blood GPX-1 activity. A plateau for total Se in plasma was achieved within 75 to 90 days, though this was not observed in blood total Se or GPX-1 activity.

On d 84 and 112, horses supplemented with Se yeast showed greater total Se in blood compared with horses supplemented with sodium selenite at the same dietary concentration. Selenocysteine (the predominant form of Se in whole blood and plasma) increased in all horses supplemented with Se. The selenomethionine content of whole blood and plasma increased in horses supplemented with Se yeast, but not in horses supplemented with selenite. The study concluded Se yeast was more effective than sodium selenite in increasing Se in blood, but it did not modify GPX-1 activity.

Practical interest

Within the European Union, selenium yeast has recently been authorized as a feed additive. Given the increased availability, the use of Se yeast to provide supplemental Se to horses may be worthy of considering. However, while this study clearly demonstrates the Se from Se yeast is more available, it fails to demonstrate any other benefits. Hence, from a practical standpoint, one needs to question whether the increased cost could be justified. Quite possibly, simply adding more sodium selenite may accomplish the same goal of increasing the Se status of the animal. However, there is little available research that supports having the Se status of a horse dramatically higher than what is normally accomplished with supplementing Se via sodium selenite. Hence, one could question whether the extra expense of using Se yeast is justified.

Comments on the study

This study was reasonably well-designed with an attempt being made to balance horses within each treatment group and to have the horses all consuming the same diet for a fairly long period before being placed on their respective diets. While the results of the study showed the source of Se had an effect on plasma and blood Se, no other clear benefit was shown indicating that Se yeast was better at improving health parameters than was the inorganic selenium. That being said, one cannot conclude that there are not any such health benefits – it is just that none were found in this study. Obviously, there are two ways the results of this study can be used incorrectly. First, if one were to say that supplementing with Se yeast is a better way to provide additional Se in an equine diet, this study could support the opposing side and say that a better (i.e. more economical) way to provide supplemental Se is simply to provide a bit more sodium selenite. It would likely cost less and still yield the same results as feeding a bit less of the Se yeast. One must also recognize that simply because no health benefits were shown, it does not guarantee that none exist. In pretty much all research, scientists attempt to examine the physiological parameters most likely influenced by a treatment and that are economically feasible to measure. However, many other parameters are never measured and these may have been ones that were altered. Simply put, while this study answers the question it set out to answer (is the selenium from Se yeast more available than the Se from sodium selenite), it does not answer the more important question which is whether this is of much benefit. From all indications, it seems that providing Se from this source yields little other health benefits so trying to justify its use on that basis would certainly be limited. One benefit that may exist, but which was not addressed in this study, is whether there may be environmental advantages by being able to feed less to have the same effect. Whether or not this benefit justifies the added cost is a question that consumers may have to face. An even more important question may be whether there are health concerns (at least in humans) when using an organic Se source as compared to an inorganic form (Scientific Committee on Food, 2000) given the improved or altered bioavailability.

Wigley R (2009) The glucosamine debate: is it better than placebo? Inflammopharmacol, 17: 191-192.

Objectives

To evaluate the recent studies that examined the efficacy of glucosamine in treating or preventing osteoarthritis.

Why chosen

This paper illustrates how results of studies can be influenced by the placebo effect. As a result, it serves to illustrate the importance of proper experimental design to avoid such problems.

Background

Large sums of money are spent every year on various supplements to treat or prevent osteoarthritis. One of the most common is glucosamine. Its use is widely recommended in horses, dogs, and humans. Despite common use, data demonstrating efficacy are limited. However, to better evaluate whether its use is warranted, greater familiarity with the research in this area is needed.

Overview of the study

Several studies examining the use of glucosamine in randomly controlled trials are reviewed including the following:

- Clegg *et al.* (2006) Glucosamine, chondroitin sulfate and the two in combination for painful knee osteoarthritis. New England J Med 354:795-808.
- Wigley (2007) When is placebo effect not an effect? Clin Med 7:450-452.
- Suarez-Almazor *et al*. (2007) A randomized controlled trial of osteoarthritis of the knee: effects of provider communication style. Arth Rheum 56 (suppl 19):S315.
- Sawitzke *et al*. (2008) The effect of glucosamine and/or chondroitin sulfate in the progression of knee osteoarthritis. Arth Rheum 58:3183-3191.
- Rozendaal *et al*. (2009) Effect of glucosamine sulphate on joint space narrowing, pain and function in patients with hip osteoarthritis; subgroup analyses of a randomized controlled trial. Osteoarthr Cartil 17:427-432.

Main findings

The Clegg *et al*. (2006) study examined 1,583 subjects and found that glucosamine chloride or chondroitin sulphate, administered by themselves, did not differ from a placebo in their ability to have test subjects report favorable results. In contrast, 66.6% of the subjects receiving the combination of the two agents reported favorable improvement. However, there was only a trend ($P = 0.09$) for this to be different from the placebo as the control group reported a 60.1% improvement. This paper questions whether there was only a 6.5% improvement in the individuals receiving the combined treatment

(66.6 - 60.1% = 6.5%) and whether that small improvement could be attributed to the subjects receiving six tablets per day compared to three tablets with the other two treatments. It was not reported if, truly, the number of tablets differed between groups, but if it did, the perception by subjects that taking 6 tablets per day is better than taking 3 could have contributed to the small improvement. Subjects from this study were followed for two years (Sawitzke *et al*., 2008) and no differences were seen between joint spaces in any treatment groups compared with the control group. This strongly suggests glucosamine does not physiologically treat osteoarthritis as the placebo effect should have no influence on this measure. Likewise, Rozendaal *et al*. (2009) reported that in a two-year study of 222 patients with hip osteoarthritis, there were no differences in symptoms or in radiographic progression between those taking glucosamine sulphate or placebo.

There are two examples of the influence a placebo can have in arthritis. Wigley (2007) found that replacing a positive suggestion with a neutral suggestion eliminated the placebo response for vitamin E for hand pain associated with rheumatoid arthritis. Likewise, Suarez-Almazor *et al*. (2007) showed that a positive suggestion increases and that neutral suggestion decreases apparent treatment gain when acupuncture is used for knee osteoarthritis.

Practical interest

This paper has two very important practical take-home messages for the equine community. First, the supplementation of glucosamine likely has little benefit for the treatment or prevention of osteoarthritis in horses. Hence, spending money on such a treatment and possibly avoiding a more efficacious treatment would not be advised. Furthermore, it is important to recognize that a placebo effect can greatly impact the results of a study. This emphasizes the need to avoid anecdotal evidence as if someone believes a product will work, it greatly increases the likelihood they will claim that it works. In several blinded and placebo-controlled studies, I have had trainers claim that the product they were using was great and that their horses were performing much better than they had been performing. Ironically, at the time they were making their claims, their horses were being supplemented with a placebo – wheat flour. It is extremely doubtful that the small amount they were receiving – an amount to replicate the amount of the test substance – had any effect on their horse but the trainers believed it did. If the studies had not been blinded and placebo-controlled, the findings may have been very misleading.

Comments on the study

While this article is published in a scientific journal, it is intended only as a 'point of view'. As opposed to presenting the results of a study, the author critically analyzes some of the more commonly cited recent studies that examined the efficacy of glucosamine. While concluding that 'glucosamine chloride has not been shown to be more effective than placebo in any trial', the author also concludes that glucosamine sulphate has had no effect on hip osteoarthritis and that 'the only trials that report an effect greater for the sulphate than placebo for knee osteoarthritis were supported by the manufacturer'. While that, in itself, does not disqualify those studies, having independent confirmation would be important before one could conclude it is efficacious. Even if it is independently confirmed, one needs to question whether using large numbers of subjects to find statistically significant improvements that might be of limited clinical value is justifiable. When 1,583 humans are needed to find a trend for a 6.5% improvement by supplementing both glucosamine and chondroitin sulphate (which again may have been influenced by the treated individuals consuming twice as many tablets) and for that trend to have a *P*-value of 0.09, it is highly unlikely equine researchers will be able to find significant treatment differences for this product in well-controlled and blinded studies. Granted, the reviewed studies were all in humans and may not be applicable to the horse – but it is highly suggestive that glucosamine may have either no effect, or only a minor one, on osteoarthritis in the horse.

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How not to train sport horses: detrimental regimens and exercises

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Take home messages

- Racehorses should start to exercise as two-year olds.
- Slowly introduce faster work during the first months of training.
- A lot of high speed work is likely to be detrimental.
- Doing zero fast work during training will not prepare the musculoskeletal system for racing.
- Between 4 and 7 furlongs (800-1,400 metres) of fast work per week is probably best.
- Well maintained turf is probably the safest surface on which to train and race.
- Warm up and cool down periods are important in all sports horses.
- Further studies on the influence of different surfaces on injury rates in all sports horses are on-going.

Introduction

This paper summarises some of the recent research into training methods that have been shown to be detrimental to sports horses. The vast majority of this work has been performed in the racehorse but more recently work has been completed in dressage horses and is underway in show jumpers.

In order to provide an 'evidence-based' set of recommendations this paper focussed on the most robust research in this field. In doing so the review is restricted to studies with large study populations and avoids references to small studies with insufficient power to detect statistically significant associations.

This paper addresses the following questions for racehorse training:
- When should full/pre-training commence?
- How much should a horse train per week?
- On what surfaces should a horse train?

In addition some aspects of dressage horse training relating to exercise regimens and training surfaces are investigated.

When should full/pre-training commence?

It has long been suggested, indeed some in the industry still hold the view, that starting race training at the age of two years is too early. With this in mind the RSPCA, in the UK, commissioned research to specifically test this (and other) hypotheses. Wood *et al*. (2001) in fact demonstrated that starting to train and race as a two-year old was beneficial to the long-term survival of the horse. Horses that started racing over the age of two were more likely to suffer a fatality during a race than those that started racing at two years of age. This finding was supported by Parkin *et al*. (2004, 2005) who showed that horses that first raced at the age of two were less likely to sustain a catastrophic distal limb fracture or a lateral condylar fracture of the distal third metacarpus at any point in their subsequent career.

Both of these studies also demonstrated a significantly increased risk of fatality or catastrophic fracture during the first races or first year of a horses racing career. The relationship between the risk of catastrophic fracture and first year of training was present regardless of the age at which the horses started training. This perhaps suggests that younger horses are more ‘fragile’ and should therefore be slowly eased into training and racing.

How much should a horse train per week?

There is clearly a balance between cardiovascular fitness and skeletal adaptation/pathology that needs to be considered. Horses must have the required cardiovascular fitness to enable them to perform at their best during a race. However, overtraining can result in excessive strain on the musculoskeletal tissues that may result in an inability to train and race or even in a catastrophic breakdown.

Work from Hong Kong would suggest that getting this balance correct is especially crucial in the first few months of racing. Lam *et al*. (2007) demonstrated that horses that raced further in their first six months of training after import into Hong Kong were more likely to eventually retire due to a tendon injury. For example, a horse that raced a total of 10,000 metres, during the first six months in Hong Kong, was approximately three times more likely to eventually retire due to a tendon than a horse that raced a total of 4,000 metres, during the first six months in Hong Kong.

Once the first year of training has been successfully negotiated it is still important to balance the needs of the cardiovascular system and the musculoskeletal tissues. A number of studies have been able to draw conclusions about the level of exercise required to avoid fracture or tendon injury.

For example, work from the UK has shown that horses that exceeded 220 furlongs (44 km) at canter (less than or equal to 14 m/s) and 30 furlongs (6 km) at gallop (greater than 14 m/s) in a 30-day period were at the highest risk of fracture of any bone (Verheyen *et al*., 2006a). This level of gallop distance exercise equates to approximately seven furlongs per week which compares closely with that reported in California (25 furlongs per 30 day period) by Estberg *et al*. (1998).

Together these, and other, studies provide good evidence of the association between increased amounts of high-speed exercise and the risk of musculoskeletal injury. These findings are consistent with the hypothesis that horses doing a lot of this type of exercise are also accumulating sub-clinical or clinical bone damage that may result in a catastrophic outcome.

There are some studies that have failed to identify significant associations between increased exercise distance and the risk of injury. In fact one of these studies actually demonstrated that horses with catastrophic musculoskeletal injury accumulated significantly less high speed exercise in the 30 day period before the injury date compared to uninjured controls (Cohen *et al*., 2000). Cases were also more likely to have done zero high-speed exercise than controls before the injury date. This finding was also identified when risk factors for catastrophic distal limb fracture (Parkin *et al*., 2004) or lateral condylar fracture of the third metacarpus (Parkin *et al.*, 2005) were investigated in the UK. The bones of the horses which were doing no high-speed exercise in these studies may not have been optimally adapted to the loads they would experience under racing conditions and were therefore more likely to fracture.

The final results produced for catastrophic distal limb fracture and lateral condylar fracture suggested that the risk was highest for horses doing zero fast work. For horses doing between 4 and 10 furlongs of fast work per week the risk was reduced and thereafter the level of risk did not alter (Parkin *et al.*, 2004, 2005). Very few high load cycles have been demonstrated as being sufficient to induce an osteogenic response in avian ulnar bones (Rubin and Lanyon, 1984). Therefore relatively short distances of gallop work during training may be protective against fracture during racing. Biologically plausible findings such as these could contribute significantly to the formulation of training regimens designed to reduce the risk of injury.

On what surfaces should a horse train?

In many racing jurisdictions the majority of training occurs at the same location as racing and sometimes on the racetrack itself. However, in others such as the UK the vast majority of training occurs on private or public gallops away from racecourses. The former scenario makes it difficult to compare the effects of different surfaces as most, if not all, horses will be using the same surface. In the later scenario there are so many other factors that may contribute to differences in injury rates on different surfaces (e.g. training regimen, topography and population demographics) that identifying definitive associations with surface type have proved difficult. Nevertheless, some authors have reported significant differences in training injury risk. For example, dirt training surfaces have been associated with a higher incidence of dorsal metacarpal disease than wood fibre training surfaces in the USA (Moyer *et al.*, 1991) and horses that spent more time exercising on Equitrack® were less likely to fracture in a study on a number of yards in the UK (Pickersgill, 2000).

A further study conducted in the UK showed that horses that predominantly used a particular unnamed all weather surface, were eight times more likely to have sustained a pelvic or tibial stress fracture (Verheyen *et al.*, 2006b). In these analyses a horse was defined as predominantly using a particular surface if it had spent more than 70% of its time on that surface during the 30 or 60 days prior to fracture date. Horses that did not spend more than 70% of their time on one surface were defined as using a mix of training surfaces and were classed as the reference group, with which other surfaces were compared. The authors recognise that using a 70% cut-off was arbitrary and also that many other factors were not accounted for such as incline and maintenance of the training surface, indicating that further investigations measuring other training variables are required before definitive statements about the effect of training surfaces on injury and lameness can be made.

Work in other sports horses

In comparison to the amount of work performed in the Thoroughbred racehorses there is limited evidence from other disciplines. Some work has been completed in the dressage horse (described below) and work is currently on-going in show jumpers. This work is being led by Lars Roepstorff at the University of Uppsala and initial results should be available during 2010.

Dressage horses

These findings come from a questionnaire study conducted by Murray *et al.* (*in press*). More than 12,000 registered members of British Dressage were asked to complete a questionnaire designed to identify training and training surface characteristics associated with injury or lameness.

The amount of time spent exercising on a horse walker was associated with lameness. Horses that did seven hours per week compared to one hour per week were almost twice as likely to have been lame. It is however important to remember that the most likely explanation of this finding was that horses that spent more time on the horse-walker were doing so because they were recovering from an episode of lameness. As such use of a horse-walker is a result of lameness rather than a cause of lameness.

In contrast lunging as part of a normal exercise regimen was associated with a reduced likelihood of lameness in the previous two years. Horses that were regularly lunged were 0.8 times less likely to have been lame in the previous two years, than horses that were not normally lunged. This association may be due to adaptation of the musculoskeletal system to different types of exercise and potentially improved proprioceptive conditioning. It is also possible that horses being lunged were fitter or maybe warmed up and cooled down more effectively than those that are not lunged, thus reducing their predisposition to lameness.

Horses that trained most often on outdoor arenas (whatever they were made of) were less likely to have been lame in the previous two years compared with horses that most often trained in indoor arenas. This may reflect various factors such as arena size, surface characteristics or arena maintenance that require further investigation.

Horses that most often used an arena with a sand-based surface were more likely to have been lame compared with horses that used other surfaces. However, for horses that trained on sand-based surfaces there was also a small but significant negative association between the number of times per week that horses trained on these surfaces and the likelihood of lameness. This finding probably illustrates the process of adaptation in bones, tendons, joints and muscles. In other words, keeping to a particular surface type (even those that may inherently be more risky such as sand-based surfaces), may be the better injury prevention strategy than continually changing training surface type. To maximise this beneficial effect the best option would be to continually train on the type of surface on which the horse would spend most of its time in competition.

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Plyometric training for the development of strength in humans: principle and practice for its application in horses

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Take home message

Strength is a critical physiological ability for the optimal performance in equine disciplines. The total force exerted across a muscle is the sum of active force generated by contractile proteins and passive elastic energy provided by interstitial structures of the muscle-tendon complex. The horse is an extreme (superior) athlete which has a considerable number of physiological adaptations to produce greater amount of force in a very efficient manner: its increased muscle mass, its high proportion of fast-twitch muscle fibres with a shortening velocity higher than expected according with its considerable body size, and its great locomotor efficiency, which typically consists of exercises based upon the stretching-shortening cycle (henceforth, SSC), in which the elastic energy is stored in the passive elements of the muscle-tendon complex during the lengthening (eccentric) phase of the cycle, and then reused and added to the active energy produced in the shortening (concentric) phase. Probably because of these unique physiological features of equine locomotion, training strength in horses is much more difficult than training strength in other athletic species.

Unfortunately, there is a lack of scientific studies determining the optimal training methods for the specific development of strength and power for improving dynamic athletic performance in horses.

Nonetheless, from the extensive human literature available, there is now sufficient evidence that plyometric training (henceforth, PT), which basically consists of SSC exercises such as those that naturally and spontaneously occur in equine locomotion, represents a method of choice when aiming to improve athletes' explosiveness and strength. Together, these results suggest that the increase in muscle strength seen after PT is attributed to a number of specific adaptations of the musculoskeletal system: increased motor unit recruitment, single muscle fibre hypertrophy, increased muscle fibre shortening velocity, rise of myofibre stiffness and improvements of the mechanical properties of the muscle-tendon complex. Overall, these adaptations underlie a significant enhancement of jumping performance, running economy and running distance performance. Recent well-documented studies have partially documented some of these physiological adaptations to several training tools in horses, providing valuable information about what training stimulus is necessary for improving muscle strength in these athletes.

For example, weight bearing, as a form of progressive resistance plyometric exercise training, can improve strength through a selective type II muscle fibre hypertrophy. In sport horses, jumping training, as a natural stimulus of SSC exercises, and riding school dressage-type exercises on ground with different slopes, can also improve muscle strength significantly. In racehorses, significant adaptations in strength can be obtained with interval training by means of high-intensity (about 80 to 100% of the maximal aerobic capacity) and short-duration (5-10 minutes per session), 3-5 days per weeks for 8-12 weeks. To reduce the risk of injury into the musculoskeletal system inherent to these high-speed gallops, similar adaptations in strength can be obtained galloping the horses at lower speed on a slopped track (3°), probably because the hindlimb muscles may be fully recruited during these gallops. In designing the training program, it is important to understand that the larger adaptations in strength occur in the six to eight first weeks of training, and that further adaptations cannot be induced prolonging and (or) intensifying training load beyond this initial 12-16 weeks period. Trainers, riders and practitioners should be able to recognize when signs of overtraining are present in these animals. Finally, regular and progressive exercise training can induce in young animal (less than 2 years old) muscle fibre hypertrophy and larger and stronger tendons, which can improve in later strength performance and reduce the risk of training-induced injuries.

Introduction

Strength, defined as the capacity to develop muscular tension, is one of the most critical factors for the optimal dynamic athletic performance in both human and equine athletes. Thus, performance of a number of individual and team human sports requiring jumping, kicking, and sprinting relies heavily on explosive leg strength and power acceleration capacity (Markovic, 2007). Similarly, although most equestrian disciplines, including Thoroughbred racing, are largely aerobic in nature, they also depend of very high rates of force produced in a very short time (e.g., push-off for a jump or specific dressage exercises), or to be maintained over a longer period of time (e.g., the maximum running speed phase of a race). It has been demonstrated in humans that peak vertical ground reaction forces during foot contact in maximal sprinting (less than 100 ms) can be higher than 5 times body weight (Mero *et al.*, 1992), equivalent to about 1,675 N (Weyand *et al.*, 2000), advocating the importance of both high force and high power generation. In a show jumper of about 600 kg of body weight, the total peak force produced by each forelimb during the trailing phase of a free jump over a vertical fence of 1.15 m height has been estimated to be about 8,160 N (Bobbert and Santamaria, 2005). In a Thoroughbred racehorse of about 475 kg of body weight galloping in field conditions at 17 m/s, the peak vertical limb force loaded during each foot contact (ranging between 75 and 90 ms) is about 12,000 N for the forelimbs, and about 7,250 N for the hindlimbs (Witte *et al.*, 2006). These data clearly demonstrate that equine dynamic movement performance requires both high and explosive force generation of the musculoskeletal system.

In human sport medicine, much effort from both coaches and researchers has been focused over the past decades on determining the optimal training methods for the specific development of strength and power for improving the dynamic athletic performance. Although several training methods have merged for this goal (i.e., heavy resistance, explosive-type resistance, weight training, etc.), most human researchers and practitioners seem to agree that plyometric training (henceforth, PT), which typically consists of stretch-shortening cycle (henceforth, SSC) exercises characterized by multi-joint actions, rapid eccentric phases and explosive concentric muscular contractions potentiated by the stretch reflex, represents a method of choice when aiming to improve athletes' explosiveness and dynamic performance (Komi, 1992; Markovic, 2007). Numerous human studies have sufficiently informed that this training method improves significantly high-power dynamic movement performance, particularly jumping and sprinting abilities, by inducing specific neuromuscular and musculotendinous adaptations related with strength and power, as well as it reduces the risk of musculoskeletal injuries by minimizing muscle soreness and enhancing functional joint stability (Chimera *et al.*, 2004; Malisoux *et al.*, 2006a,b; Kubo *et al.*, 2007; Impellizzeri *et al.*, 2008).

Unfortunately, scientifically-based and optimal training methods for the specific development of musculoskeletal strength and power have not been introduced in the equine sport practice, even when there is an extensive specialized literature focused on equine musculoskeletal adaptations to training (e.g., see Rivero and Piercy, 2008; Smith and Goodship, 2008 for recent reviews). Investigators in these fields have traditionally been more interested to know how plastic the horse's musculoskeletal system is rather than optimizing practical methods. For example, few studies have compared the effects of different training protocols varying in the type, intensity, duration and frequency of the exercises (e.g., Sinha *et al.*, 1993; Gansen *et al.*, 1999; Rivero *et al.*, 2007). Furthermore, most of these earlier training studies failed to offer an appropriate description of the training design, and (or) used very limited methodologies to accurately describe strength and power training-linked adaptations. As a consequence, these studies are of little, if any, application in practice. Recent years have seen, however, the publication of some well-documented training studies in both sport and racing horses providing valuable information about (1) what strength-related physiological parameters can be modified in horses with training, and (2) how can this be achieved in practice.

Interestingly, most equine natural gaits (walk, trot, gallop, jumping) consist of SSC exercises, in which lengthening (eccentric) and shortening (concentric) actions of the muscle-tendon complex in the limbs are repeated during each cycle (Bobbert and Santamaria, 2005; Butcher *et al.*, 2009). As most of equine training methods used in practice consist of natural gaits at different intensities and durations, it can be assumed that these procedures represent a natural stimulus of PT in the horse. It can then be speculated that, although still not demonstrated in horses, many of the specific neuromuscular and musculotendinous adaptations already demonstrated in humans after PT might also occur in horses. This paper will discuss the literature in relation to key factors of both strength and power adaptations in the horse. The major emphasis will be on the equine discipline; however, in areas where equine literature is lacking, directly relevant literature from these studies in the human area will be presented. The review will focus on peripheral neuromuscular and musculotendinous adaptations to strength training and the level of stimuli required to trigger these adaptations. Also some recommendations relating the requirements for adaptation to the design of training programs will be offered. But as key players involved in equine training in practice (practitioners, trainers and riders) usually have limited information about the scientific approach to training, this review will begin with some relevant physiological considerations responsible for strength and power, in order to provide an appropriate scenario for subsequent discussion on experimental results. For a more general scientific background of equine conditioning in practice, the readers of this article can be referred to previous reviews (Davie, 2006; Rivero, 2007; Rogers *et al.*, 2007).

Main physiological components responsible for strength

The horse is an extraordinary (supreme) athlete that can run at high speeds (18 m/s, 65 km/h), maintained for about 1-2 minutes over distances of 800-1,550 m or clear a fence of more than 2.30 m height with a rider on its back. This superior athletic ability is attributable to a number of physiological adaptations: its high maximal aerobic capacity, the ability to increase oxygen-carrying capacity of blood at the onset of exercise through splenic contraction, its increased muscle mass, larger intramuscular stores of energy substrates (glycogen in particular), high mitochondrial volume, high proportion of fast-twitch muscle fibres with a shortening velocity higher than expected according with its body size, and efficiency of gait (see Hinchcliff and Geor, 2004, for a mini-review). Probably because of these unique physiological features, training horses is much more difficult than training other athletic species.

The horse's skeletal musculature is highly developed, particularly in athletic breeds. In contrast to most mammals, in which 30-40% of body weight consist of muscle, or in non-athletic horse breeds (42%), more than half (about 55%) of a mature Thoroughbred's body weight comprises skeletal muscle (Gunn, 1987). Low body fat and a large amount of muscle likely reflect adaptation and selective breeding of animals used for elite endurance and sprint racing (Kearns *et al.*, 2002). Equine locomotor muscles are mainly located proximally on the appendicular skeleton with their tendon located distally, which has the effect of reducing the weight of the lower limb and decreasing the energy to overcome inertia when the limb swings back and forth (Wilson *et al.*, 2001). As already mentioned, equine natural gaits consist of plyometric muscular contractions based upon SSC exercises. During these contractions, the elastic energy is stored in tendon structures and intramuscular connective tissues during the lengthening (eccentric) phase of the cycle, and then reused and added to the active energy produced in the shortening (concentric) phase. Thus, the total force exerted across a muscle is the sum of active force generated by the contractile machinery and passive force provided by elastic structures that are arranged either in parallel or in series with the contractile proteins in the muscle-tendon complex.

Even when the contractile behavior differs considerable from muscle to muscle, in the superficial digital flexor muscle of the forelimb, slight fascicle shortening occurs just prior to or immediately after contact in walking (Figure 1A; Butcher *et al.*, 2009). The shortening is followed by substantial fascicle lengthening over the majority of the remaining functional contact period as peak force is achieved. Average muscle stress and force in this muscle ranges between 0.16-0.173 MPa and 3,873-4,117N, respectively (Butcher *et al.*, 2009). With increased speed at trot, muscle force increases substantially while average and peak fascicle strain decreases relative to walking (Figure 1B). During trotting, the superficial digital flexor fascicles shorten to a greater degree (up to 1 mm) just prior to and immediately following hoof contact, as force levels rise rapidly. The period of fascicle shortening is followed by a very brief period of isometric contraction near peak muscle force before lengthening, while in others shortening is followed by 1.5-2

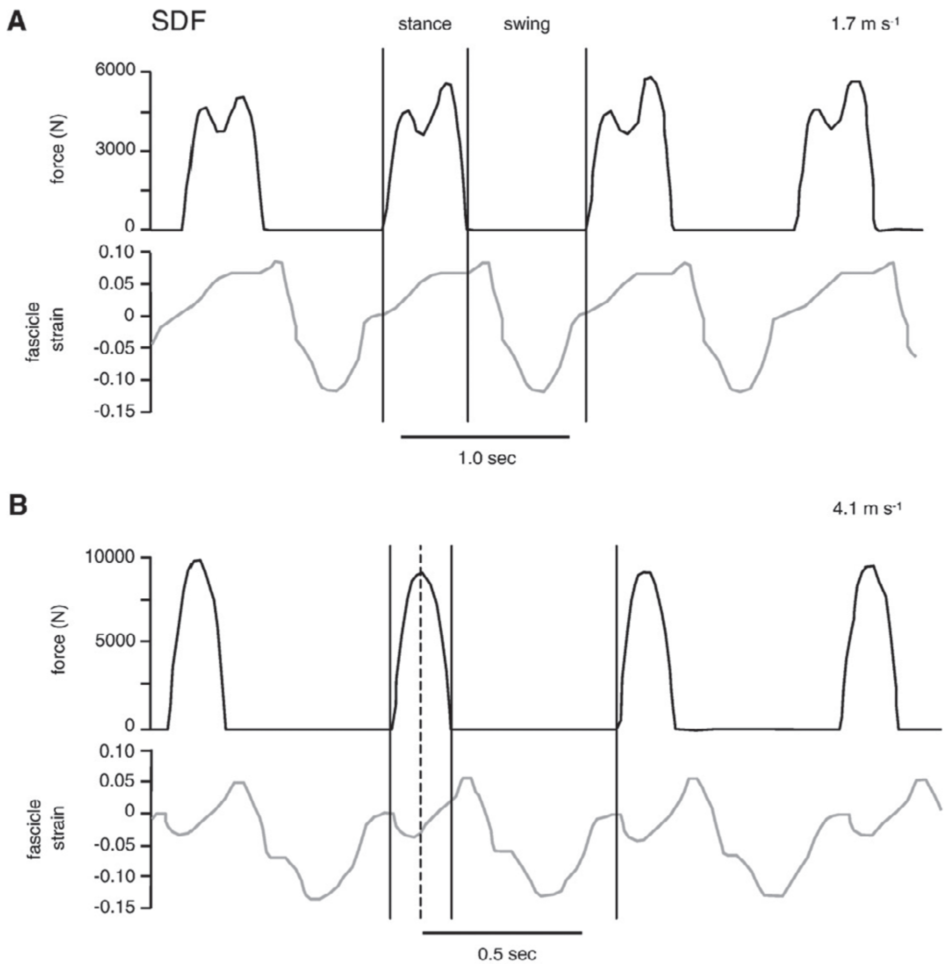

Figure 1. Representative in vivo *recordings of muscle force and fascicle strain from the superficial digital flexor muscle of the forelimb during walking (A) and trotting (B). Solid vertical lines mark stance phase (functional contact period) and wing phase of the stride. Muscle force occurs during the stance phase, being higher during trotting than during walking. Fascicle (grey lines) shortening occurs about the functional contact point and continues during early stance for walking and even further for trotting. More explanations in the text. Source: Butcher* et al.*, 2009.*

mm of fascicle lengthening corresponding with high peak force levels (Figure 1B; Butcher *et al.*, 2009). The importance of SSC exercises for the equine locomotion is clearly illustrated by analyzing the contribution of the forelimbs and hindlimbs of the horses to mechanical energy changes in jumping (Figure 2; Bobbert and Santamaria, 2005). During the forelimb push, the total energy first drops by 3.2 J/kg (stretching phase) and then increases by 4.2 J/kg (shortening phase; Figure 2A). The energy stored in elastic components during the lengthening phase is released again and contributes to the gain of total energy during the second half of the forelimb push, and is added synergistically to the energy regenerated in concentric contractions of proximal limb

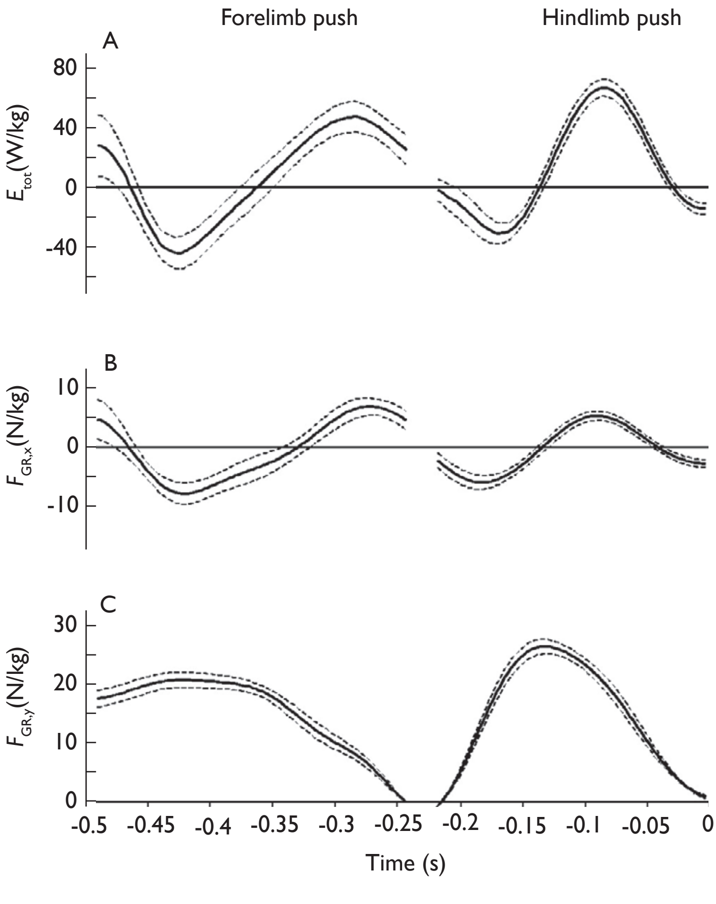

Figure 2. Average time histories of (A) total power output, (B) the horizontal component of the calculated ground reaction force, and (C) the vertical component of the calculated ground reaction force in forelimbs and hindlimbs during the push-off for a free jump over 1.15 m high fence. Solid lines indicate averages of 23 horses; broken lines indicate 95% confidence intervals. Explanations in the text. Source: Bobbert and Santamaria, 2005.

muscles. During the hindlimb push, the total energy also first drops, but only slightly by 1.6 J/kg, and then increases again by 4.1 J/kg (Figure 2B). These differences between limbs clearly illustrate that during the hindlimb push, the muscles of these limbs act primarily producing active energy (positive work).

The efficiency of the SSC mechanism is admirable in a cursorial quadrupedal animal like the horse. Thus, the force required to elevate the centre of mass is provided when the limbs are in contact with the ground, but movement of distal limbs is mainly passive, and results from the release of elastic energy when the limbs are unloaded, and previously stored in the muscle-tendon complex during weight bearing (Wilson *et al.*, 2001). Wilson and Watson (2003) magisterially linked thoracic horse limb protraction at high speeds to a catapult mechanism in which energy is stored in elastic tissues (the internal tendon and lacertus fibrosus of the biceps brachii muscle) during limb load, but released rapidly when the toe leaves the ground, thereby quickly protracting the limb. This effective utilization of elastic strain energy explains much of the extra locomotor capacity of the horse which consumes less metabolic energy than would be expected based on the substantial energetic demands on this animal, particularly when moving at fast gaits. Moreover, metabolically economical force production may also be increased through eccentric muscle contraction, where muscle length increases while active due to externally applied forces. The resisting force of an eccentrically loaded muscle can be double than that of maximal isometric force. In fact, several studies in humans have shown that training by means of SSC exercises improve significantly running economy and distance performance (Spurs *et al.*, 2003; see below).

Active (contractile) components of the force

More than 90% of a muscle is made up of muscle fibres, with the rest consisting of nerves, blood vessels, and the fat and connective tissue that separate the individual fibres (endomysium), the fascicles (perymisium) and the whole muscle (epimysium). The connective tissue merges with both the origin and the insertion tendons of the muscles, as well as with internal tendons in compartmentalized muscles. The sarcomere, composed by different contractile proteins, is the unit of muscular contraction which occurs within each sarcomere, when thin myofilaments slide over thick myofilaments. Two of the features of a skeletal muscle that have been shown to be important in determining its contractile force production are its fibre type characteristics (i.e., fibre type composition and fibre size) and architectural design (i.e., angle of fibre pinnation and fibre length). Both factors have a considerable impact on both the maximum force production and maximum rate of shortening (Roy and Ishiara, 1997).

Contractile force for a particular muscle is partly regulated by the rapidity of α-motor neuron discharge; muscle fibres contract with a twitch following a single discharge of an α-motor neuron, but sustained contractions result from repetitive neuron firing. The force of contraction increases with the rate up to a maximum limit that is determined by the contractile (i.e., intrinsic velocity of shortening) and morphological (i.e., cross-sectional area) characteristics of recruited muscle fibre types. Furthermore, a process known as motor unit recruitment, which reflects the gradual inclusion of larger motor neurons and muscle fibres as greater force is required, also regulates active force production. In general, dynamic and rapid movements, such as those demanded for jumping and sprinting, rely on recruitment of larger diameter α-motor neurons and fast-twitch fibres. The horse in general, and the Thoroughbred in particular, have a considerable number of these fibres (about 40-50% of the fibre type composition in propulsive muscles), which are considered as a pool of reserve only recruited during maximal (or supra-maximal) exercise intensities (Yamano *et al.*, 2006; Rivero *et al.*, 2007). In applying the overload principle of the training (see below), it is important to understand that the level of the training stimulus must be enough to recruit these very fast fibres.

Fibre types can be differentiated by analyzing the specific myosin isoform they express, since this contractile isoprotein clearly reflects each fibre's phenotype. Adult equine skeletal muscles express three myosin isoforms designated as types I, IIa and IIx (Rivero *et al.*, 1999). Their differential distribution at the cellular level defines three pure (i.e., expressing a unique myosin isoform) fibre types (designated as I, IIA and IIX), and a hybrid fibre type co-expressing isoforms IIa and IIx (type IIAX; Figure 3). Overall, the intrinsic capacity of each fibre type to generate tension increases in the rank order I→IIA→IIX (Figure 4). There are two important reasons for this: (1) the speed at which

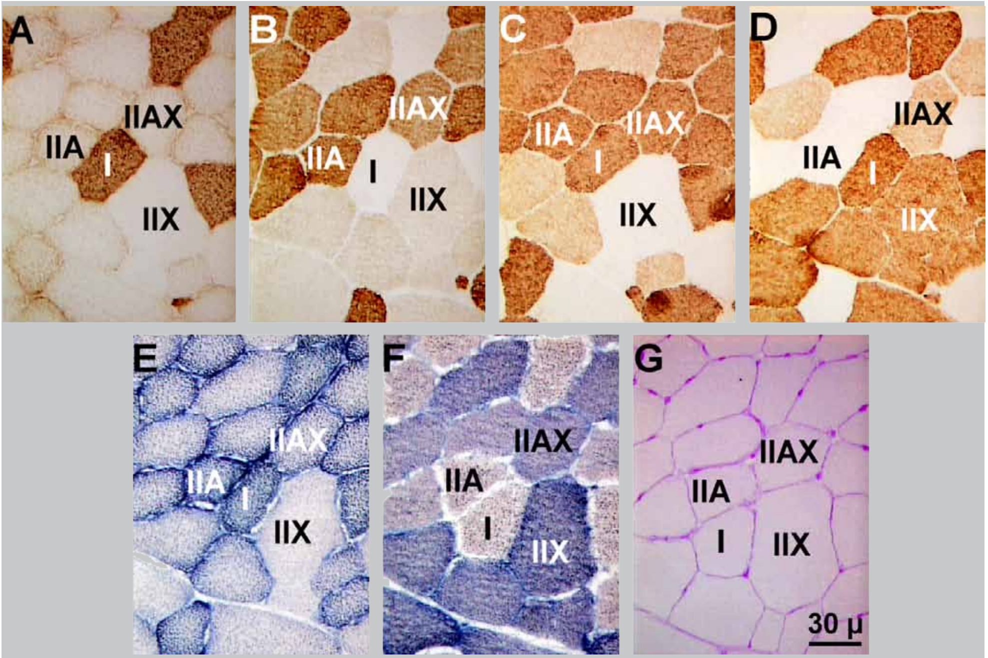

Figure 3. Serial frozen sections of gluteus medius muscle from a horse stained for immunocytochemistry and enzyme histochemistry. A-D, sections are stained with monoclonal antibodies against specific myosin heavy chain isoforms: BA-D5 (A, anti-I), SC-71 (B, anti-IIa), BF-35 (C, anti I+IIa), and S5-8H2 (D, anti I+IIx). Other sections were stained for quantitative histochemistry of succinate dehydrogenase (E, used for estimating fibre aerobic capacity) and glycerol-3-phosphate dehydrogenase (F, for estimating anaerobic capacity). G: alpha-amylase perdiodic acid Schiff for visualizing capillaries. The four myosin-based muscle fibre types are labeled in all serial sections: three of them as fibres expressing a unique myosin isoform (i.e., I, IIA, and IIX fibres), and the fourth as a hybrid phenotype co-expressing myosins IIa and IIx (i.e., type IIAX fibres). Bar, 30 µm. Note how fibre cross-sectional area increases in the rank order I→IIA→IIAX→IIX. Source: Rivero et al.*, 2007.*

each sarcomeric myosin isoform hydrolizes ATP; and (2) differences in fibre size between fibre types (Ducheteau *et al.*, 2006). Explanations are simple. (1) As the ATP hydrolysis' increases (i.e., I→IIA→IIX) → greater energy production → greater and faster actomyosin cross-bridge cycles production → faster shortening velocity → greater tension. And (2), as fibre cross-sectional area increases (i.e. I→IIA→IIX) a greater number of sarcomere is arranged in parallel → greater tension. Interestingly, the shortening velocity of the horse's muscle fibres is significantly faster than of the human fibres, despite the fact that the

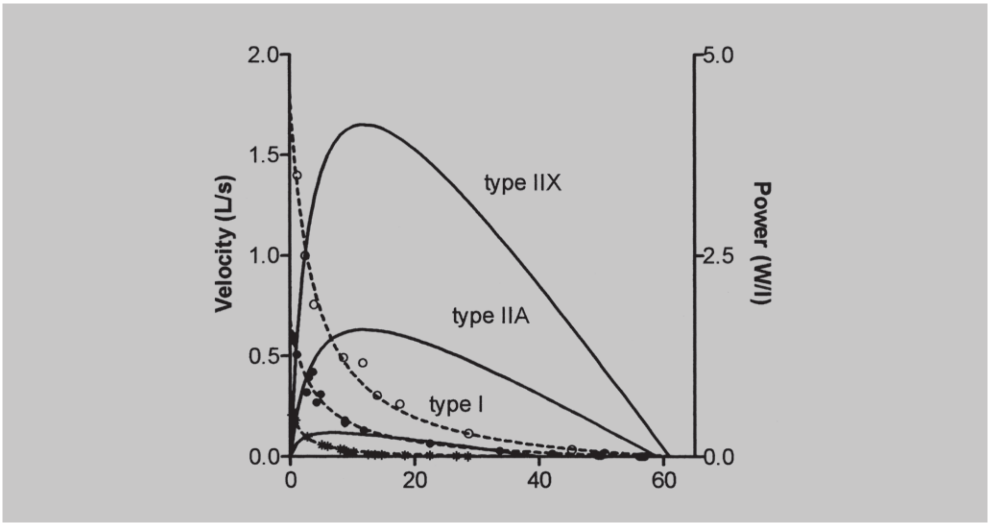

Figure 4. Force-velocity curves (dotted lines) and power-force curves (solid lines) of representative type I (stars), type IIA (full circles), and type IIX (open circles) human skeletal muscle fibres. Source: Bottinelli and Reggiani, 2000.

horse body mass is 5-fold greater than in humans (Marx *et al.*, 2006). As fibre size is body-mass-independent (Hoppeler and Flück, 2002), this physiological difference means that equine muscle fibres can generate much more tension per unit of muscle area than expected according with the great body size of this species.

In consequence, strength training should be aimed to a double purpose: (1) to enhance quantitative patterns of recruitment of fast-twitch fibre types, and (2) to stimulate a rise of muscle fibres' shortening velocity or a selective hypertrophy of type II fibres, or both. But because the unique features of the horse's physiological components of contractile force production (i.e., its very high reserve pool of type IIX fibres and the considerable high shortening velocity of their muscle fibres), it seems clear to understand that it is not easy to achieve these adaptations in practice.

As already anticipated, architectural features of muscle (e.g., fibre length and pinnation angles) also affect the force production in horses (Butcher *et al.*, 2009). In general, muscles or neuromuscular compartments of muscles with very long fibres (sarcomeres arranged in series), and small pinnation angles (fibres arranged in parallel from the origin to the insertion tendon of the muscle), which usually also contain a great proportion of type II fibres, are better designed for the development of high rates of dynamic forces. Consequently, training stimulus for the development of strength and dynamic performance in horses should be focused for stimulating adaptations in these muscles. On the contrary, muscles with short fibres arranged in a pinnate form, which usually also contain a high number of aerobic type I fibres, are better designed for generation of low intensity isometric tension, so they are less important for improving strength and power with training.

Passive elastic components of the force

The force that is generated in the cross-bridge cycle of myofibre sarcomeres is transmitted via the contractile apparatus to intermediate filament proteins that act to maintain and stabilize the muscle fibre's shape during contractions. These intermediate filaments provide a structural link first to the sarcolemma and then to the extracellular matrix via a group of proteins known as the dystrophin-associated protein complex. These elastic elements can be arranged according to two different models: one with the parallel elastic component arranged in parallel with both the contractile element (sarcomere) and the series elastic component (Figure 5A), and the other in which it is in parallel with only the contractile element (Figure 5B). Contractile forces are transmitted from each muscle fibre via the extracellular matrix and the connective tissue of tendons, and ultimately to the bones of the skeleton. The structural associations of these various elastic components are not entirely known, but in humans it is often assumed that titin, membrane and interstitial connective tissue contribute the majority of the passive force (MacInstosh and MacNaughton, 2005).

The horse has adapted the basic muscle-tendon units of the distal limbs in order to withstand the high weight-bearing loads and to act as elastic structures for energy storage and efficient locomotion. For example, the superficial digital flexor muscle has a great amount of connective tissue within it and has an accessory ligament that 'bypasses' the muscle belly to insert on the distal radius. These two adaptations allow the musculotendinous unit to withstand greater loads and to store larger amounts of elastic energy than would be possible by the

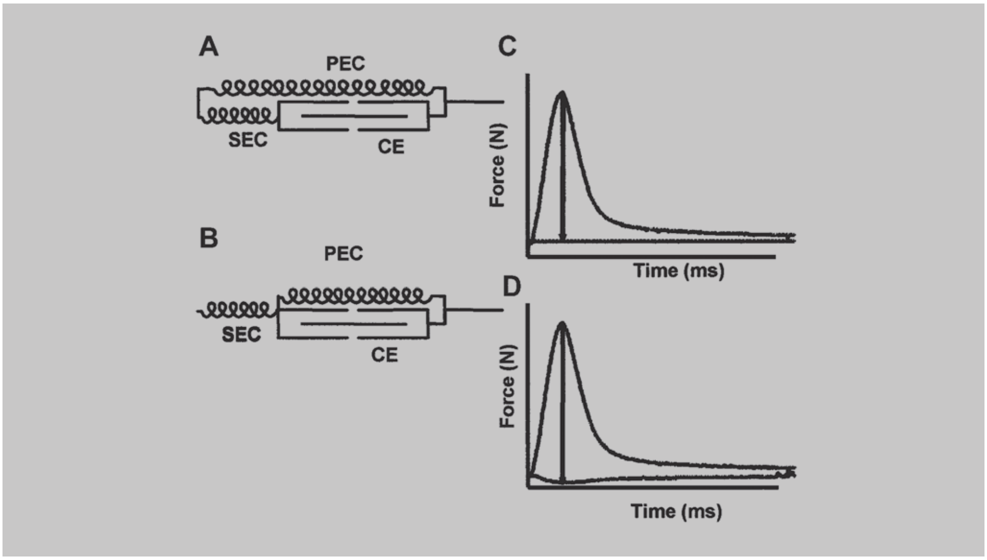

Figure 5. Hill models of muscle contractile element (CE) and passive elastic components. The elastic structures are the parallel elastic component (PEC) and the series elastic component (SEC): A, traditional Hill model with the PEC running from end to end. B, PEC is parallel with the CE, and the length of the PEC will change in correspondence with the length of the CE. C and D, estimation of active force for A and B models, respectively. Source: MacIntosh and MacNaughton, 2005.

muscle itself. It seems obvious, therefore, that higher stiffness levels of the muscle-tendon complex during SSC exercises led to an additional benefit for strength generation in terms of a greater amount of stored and reused energy. Thus, as several structural and mechanical properties of the muscle-tendon complex (i.e., cross-sectional area, collagen fibril characteristics, etc.) influence its functional behavior, tendon properties are also relevant in the physiological mechanisms underlying the total force produced by a muscle. In fact, important results in humans indicate that the percentage gains after PT are mainly attributed to changes in the mechanical properties of the muscle-tendon complex, rather to the muscle activation strategies (see below).

Musculoskeletal adaptations to strength training

Humans

For the specific development of muscle strength, human athletes commonly use heavy resistance training (80-90% of maximal load) and (or) explosive-type training in a form of either explosive (ballistic) resistance training (30-60% of maximal load) or PT which typically consist of SSC exercises defined as eccentric loading followed by a concentric contraction. But although both heavy resistance and explosive-type resistance training methods can improve muscle strength, PT represents the most optimal method for this goal (see Markovic, 2007 for a meta-analytical review). This method consists of several maximal effort SSC exercises performed with or without overweight which include different types of jumps such as static jump, vertical countermovement jump, drop jump, etc. To minimize the risk of injury the number of jumps is progressively increased in sessions of about 20-40 minutes, three times per week for 8-12 weeks (Malisoux *et al.*, 2006a; Kubo *et al.*, 2007).

Together, the results of the extensive human literature describing the effects of PT on the musculoskeletal system suggest that the increase of muscle strength seen with this training method can be attributed to a number of specific physiological adaptations (Table 1). For example, this improvement was initially related with neuromuscular adaptations, that is, the pattern of motor unit recruitment and muscle activities of agonists and antagonists (Chimera *et al.*, 2004). In fact, SSC exercises have been credited with inducing neuromuscular adaptations to the stretch reflex, muscle elasticity and Golgi tendon organs (Wilk *et al.*, 1993). The stretch reflex is initiated during the eccentric loading phase and can facilitate greater motor unit recruitment during the ensuing concentric contraction. As Golgi tendon organs have a protective function against exercise tensile loads in the muscle-tendon complex, it is thought that a Golgi tendon organ desensitization can also occur after PT (Hulton and Atwater, 1992), allowing the elastic components of muscle to undergo greater stretch.

However, the results of other more recent and refined experimental studies have unequivocally demonstrated that the increase in muscle strength seen in human athletes after PT are not mainly caused by

Table 1. Main strength-related musculoskeletal adaptations to plyometric training in human athletes.

Adaptations	Reference
Increase of motor unit recruitment	Chimera *et al.*, 2004
Stretch reflex potentiating	Chimera *et al.*, 2004
Golgi tendon organ desensitizing	Chimera *et al.*, 2004
Single fibre cross-sectional area increment	Malisoux *et al.*, 2006a
Single fibre type maximal shortening velocity increment	Malisoux *et al.*, 2006a
Fibre type peak force improvement	Malisoux *et al.*, 2006a
Increment of absolute peak power of fibre types	Malisoux *et al.*, 2006a
Single-fibre Ca^{2+} sensitivity increased	Malisoux *et al.*, 2006b
Increase of type II muscle fibre stiffness	Malisoux *et al.*, 2006a
Higher joint stiffness	Kubo *et al.*, 2007
Increase of maximal tendon elongation capacity	Kubo *et al.*, 2007
Increase of storage elastic energy by tendon	Kubo *et al.*, 2007
Improvement of running economy and running distance performance	Spurs *et al.*, 2003
Improvement of jumping performance	Markovic, 2007

adaptations of muscle activation strategies, but it must be attributed to relevant changes in both contractile and mechanical properties of the muscle-tendon complex (Malisoux *et al.*, 2006a,b; Kubo *et al.*, 2007). Thus, after 8 weeks of PT single fibre cross-sectional area increased 23% in type I, 22% in type IIA, and 30% in hybrid IIAX fibres; and maximal shortening velocity increased 18, 29 and 22% in these same fibre types (Malisoux *et al.*, 2006a). As a consequence, peak force increased 19% in type I, 15% in IIA; and 16% in IIAX; whereas absolute peak power of single fibre types was enhanced by 25, 34 and 49% in these same fibres types (Malisoux *et al.*, 2006a). It is clear from these data that SSC exercises are an effective training stimulus to improve muscle fibre force, shortening velocity, and therefore power. Plyometric training also increases single-fibre Ca^{2+} sensitivity, but this change cannot be explained by a modified TnT isoform expression pattern (Malisoux *et al.*, 2006b). In general, PT induces very few significant changes in human fibre type composition (Kyröläinen *et al.*, 2005; Malisoux *et al.*, 2006a), but a recent study has shown a significant increase in the percentage of type IIA fibres (+ 9%) and a not significant decrease of IIX fibres (- 5%) (Malisoux *et al.*, 2006b), which resemble those observed after resistance training (Widrick *et al.*, 2002).

Plyometric training can also induce adaptations in the passive elastic properties of skeletal muscle fibre types. Eight weeks of SSC exercise training in humans induce a significant increase of type II fibre stiffness, not affecting type I fibres (Malisoux *et al.*, 2006a). The most likely candidate to confer elastic properties is titin, an about 3 MDa giant structural protein that spans a half sarcomere from the M-line to the Z-disk (Neagoe *et al.*, 2003), but 15 weeks of PT did not change its expression in human skeletal muscle (Kyröläinen *et al.*, 2005). It is possible that the greater capability of muscle fibres to produce force passively after PT can be associated with the enhanced functional joint stability specifically induced by this training tool (Chimera *et al.*, 2004; Kubo *et al.*, 2007). Komi (1992) suggested that higher stiffness levels of lower limb muscles during SSC exercises led to a benefit in terms of the greater amount of stored and reused elastic energy. According to the limited information available, it can be assumed that the human tendon properties would change to be more suitable for SSC exercises after PT. Thus, even when tendon stiffness does not increase after PT, the maximal tendon elongation and storage elastic energy increase significantly after this training tool (Kubo *et al.*, 2007). Because of the unchanged tendon stiffness (Kubo *et al.*, 2007), and the increased passive tension of single muscle fibres (Malisoux *et al.*, 2006a) after PT in humans, it is likely that the increase in joint stiffness seen after this training methods (Kubo *et al.*, 2007) could be related to changes in active muscle properties (i.e., cross-bridge mechanisms of single fibres).

Several studies in humans have shown a positive correlation between leg stiffness and running economy, concluding that economical runners posses a running style that is stiffer during ground contact (Heisse and Martin, 1998). Spurs *et al.* (2003) reported that PT improves running economy and distance running performance, and thus increases the musculotendinous stiffness of the ankle joint using the oscillation technique. Finally, PT provides a statistically significant and practically relevant improvement in the human vertical jump height (range, 4.7 to 8.7%), justifying its application for the purpose of developing explosive strength in healthy individuals (Markovic, 2007).

Horses

Experimental studies demonstrating specific musculoskeletal adaptations associated with enhanced strength are much more limited in horses than in humans. Nevertheless, as SSC exercises represent the most common and natural movement of equine locomotion (Wilson *et al.*, 2001; Bobbert and Santamaria, 2005; Butcher *et al.*, 2009), it can be assumed in practice that similar adaptations also occur in horses with training.

To the author's knowledge, only a recent study in horses has demonstrated that training results in a significant adaptation of quantitative electromyographic activity that potentially resembles synchronization of individual motor unit fibre action potentials (Wijnberg *et al.*, 2008). Although still controversial, single muscle-fibre type hypertrophy can be stimulated in horses with some training tools and well designed training programs (see Rivero and Piercy, 2008 for a recent review). The main consequence of increased muscle mass due to myofibre hypertrophy in response to training is to produce a muscle with a greater peak force capacity, because force output is proportional to the total cross-sectional area of the fibre mass recruited. At low speeds, this adaptation has an impact on gait performance of dressage horses, causing a marked reduction in both stance and stride duration (Rivero *et al.*, 2001). Moreover, such an adaptation has considerable influence on the performance of show jumpers via enhanced power output from the hindquarters (Rivero and Letelier, 2000). Furthermore, because increased power output results in a greater ability to accelerate and may increase stride length (Snow and Valberg, 1994), this strength-training adaptation is also very important for racehorses competing over short distances. But enhanced power through training-induced myofibre hypertrophy comes with the cost of a corresponding 'theoretical' decline in stamina, because the increased mass of larger fibres recruited and concomitant rise in ATP utilization occur simultaneously with a relative inability of oxygen to diffuse into the larger fibres. In practice, this means that excessive training-linked myofibre hypertrophy can be a 'handicap' for endurance horses and running stayers. Due to this important reason, conditioning programs in athletic horses should ideally be focused to the development of musculoskeletal system properties that optimize equilibrium among several physiological traits (i.e., stamina, strength and speed).

Although there are no specific studies in horses describing adaptation to training of passive elastic components of the force, a combination of *in vivo* and *post-mortem* experiments on 'normal' horses have shown new insight into the influence of exercise-training on equine tendon (see Smith and Goddship, 2008 for a recent review). These studies have investigated mechanical, morphological (ultrasonographic and histological), ultrastructural, compositional, and metabolic changes in association with aging and exercise. The effects of exercise have been found to be different between immature (growing) and mature (adult) tendon. The age at which the equine digital flexor tendon matures is estimated at approximately 2 years. Although the effect of exercise-training on mechanical and morphological properties of mature tendon has shown inconsistent results, these adaptations seem to be scarce. In the growing tendon, however, exercise induces stronger tendons and a significant rise in the rate of increase in cross-sectional area of the superficial digital flexor tendon (Smith and Goodship, 2008).

Training stimulus for the development of strength in horses

Perhaps the most underrated aspect of training (conditioning) is the design of the program itself, i.e., length of the training, nature, intensity, duration, and frequency of exercise sessions applied (Davie 2006; Rogers *et al.*, 2007). But it is also important to understand that the responses of equine skeletal muscle to training also largely depend of the basal status of the muscle, determined by breed, age, and level of fitness and injury status of the horse (Rivero and Piercy, 2008). For example, this response is usually more prominent in young inactive horses than in active mature regularly-trained horses (Miyata *et al.*, 1999; Yamano *et al.*, 2002). Also, as Thoroughbreds have more glycolytic type IIX muscle fibres in their propulsive muscles than other breeds, training Thoroughbred racehorses is much more difficult than training endurance and sport horses. Consequently, the training program needs to be soundly based and individualized with consideration to the above variables in an attempt to match the ability of the horse and the type of discipline it is being trained for.

A basic principle of training is that a single exercise session leads to fatigue and mild cellular damage which in turn, results in short-term adaptive responses (Figure 6). When exercise is performed regularly

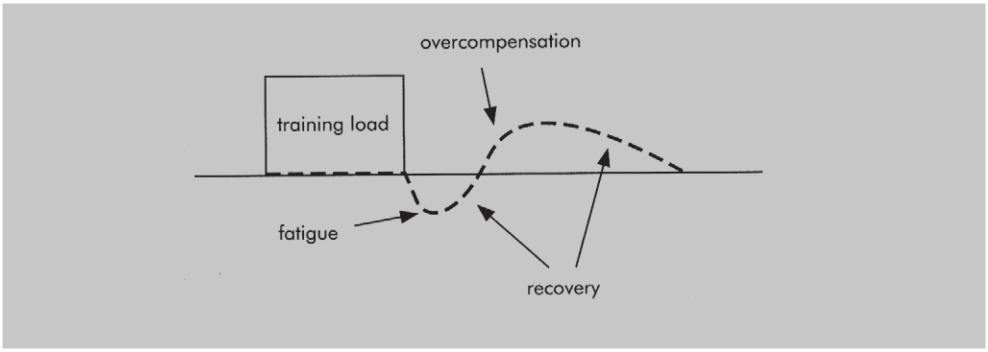

Figure 6. Principles of training: a single session. The performance curve (dashed line) of a single training session is shown in comparison with basic performance level. During training (training load), performance is conserved but declines when the horse starts to get tired (fatigued). When the training session ends and the horse gets rest, the horse recovers from the training load resulting in a performance capacity that at first increases to the baseline level but increases further with rest (overcompensation), If the recovery period is extended further the horse adapts again, and performance capacity is reduced to baseline. Source: Rogers et al.*, 2007.*

and training stimulus is increased gradually, the adaptation that occurs during the recovery period of a single exercise session leads to an overall improvement in performance (Figure 7). Thus, the basis of any training program is to continually provide increased levels of stress to the physiological systems to improve performance and strength. The 'overload principle' states that for continual adaptation this level of stress needs to be continually increased (Rogers *et al.*, 2007). However, it is important to appreciate that there is an upper limit for these adaptations and, much more interesting, that individual horses will differ in relation to how well they can cope with this stress.

When training is too vigorous and (or) rest periods between training sessions too short, performance is reduced due to an imbalance between training stress and recovery (see Figure 7C). The time period before exposure to the next training stimuli should be of sufficient duration to allow time for the training effect (adaptation) from the previous session to occur. If the next training session is applied without sufficient time for recovery, overcompensation and adaptation, then performance decrements occur in the form of earlier onset of fatigue within each session. In applying the 'overload principle', it is important to understand that adaptations take time to occur, since

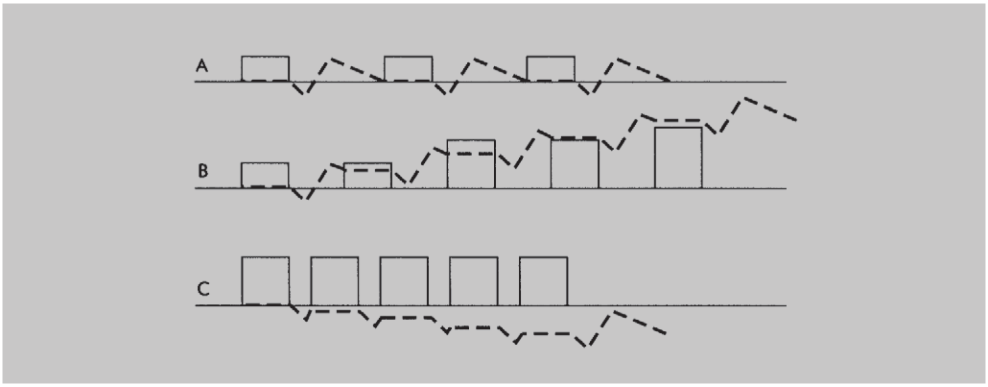

Figure 7. Principles of training: training strategies. The performance curve (dashed line) during different training strategies. A) Regular training sessions with the same load and relatively long rest periods do no increase performance. B) Regular training sessions with increasing training loads with sufficient rest periods increase performance (the overload principle). C) Regular training sessions with increasing training loads without sufficient rest periods decreases performance. Source: Rogers et al.*, 2007.*

they depend of altered rates and regulation of transcription of specific genes, and consequently a change in the amount of specific structural proteins within individual fibres (Rivero and Piercy, 2008). Human studies have shown that strength gains were fast in the early stages of training due to neural adaptations, with later improvements in strength being more a result of muscle hypertrophy (Moritani, 1992). The larger adaptations in strength occur in the first six to eight weeks of training (Davie, 2006).

In bringing about strength training adaptations, the type of exercise (training tools or training facilities), the length of the training program, the intensity, duration, and frequency of exercise sessions, and the timing and length of recovery periods are all of paramount importance. When designing a training program it is important to firstly consider the discipline that the horse is primarily being trained for, in order to individually adjust the stimuli towards the optimal development of physiological traits more directly related with a particular discipline. Recent years have seen the publication of several well-controlled training studies on determining the influence of the structure of the training program on equine skeletal muscle (Table 2). From the comparative analysis of these studies, it seems clear that improved stamina, through enhanced aerobic capacity, is the most common and

Table 2. Physiological implications of muscular adaptations in various training programs scientifically evaluated in horses.

Horses		**Conditioning protocol**			
Breed	**Age**	**Intensity**	**Duration (distance)**	**Frequency**	**Length**
Thoroughbred	4-8 yr	~55% VO_{2max}	60 min (~13-14 km)	daily	10 days
Thoroughbred	2-9 yr	~80% VO_{2max}	3 min (1500 m) x2	6 d/wk	6 wk
Thoroughbred	5-7 yr	~80-100% VO_{2max}	5 min (x2)	5 d/wk	12 wk
Thoroughbred	2-3 yr	~100-165% VO_{2max}	1.6-5.3 min (1600-3200 m)	5 d/wk	16 wk
Thoroughbred	2 yr	~100-165% VO_{2max}	1.6-5.3 min (1600-3200 m)	5 d/wk	16 wk
Standardbred	3-5 yr	~60-100% VO_{2max}	6-12.5 min (~3-9 km)	5 d/wk	16 wk 32 wk
Standardbred	2 yr	~100-140% V_{LA4} ~65% V_{LA4}	15 min 60-90 min	2nd day 2nd day	5 wk
Arab	8.6 yr	~80% V_{LA4}	50-80 min (~10-20 km)	3 d/wk	12 wk
Andalusian	~4 yr	~25-30% V_{LA4} ~50-60% V_{LA4}	45-60 min 75-120 min	5 d/wk	12 wk 32 wk

Intensity is expressed as a fraction of either VO_{2max} (velocity at maximal aerobic capacity) or V_{LA4} (velocity inducing a blood lactate concentration of 4 mmol/l). The symbols + and – indicate that either the muscular adaptations to training had positive or negative effect respectively towards the particular characteristic; the number of symbols is proportional to the magnitude of the adaptation; ne: no effect; ni: not investigated.

easy to obtain muscular adaptation to training, regardless of training parameters. However, simultaneous improvements of both strength (trough a myofibre hypertrophic growth) and speed (via enhanced anaerobic capacity) are much more difficult to obtain. The rest of this paper will try to describe what training stimulus can be used for improving strength in sport and racehorses.

Specific muscle fibre hypertrophic growth can be stimulated in horses with burst of high-resistance muscle activity and by prolonged stretch beyond normal resting length. Weight bearing, as a form of progressive resistance exercise training, has been investigated in ponies for improving strength, muscle power, and muscle size (Heck *et al.*, 1996). In this experiment, ponies underwent 8 weeks of 3 days/

Functional significance			Reference
Stamina	**Strength**	**Speed**	
+	ni	–	Geor *et al.*, 1999
+	ni	–	Sinha *et al.*, 1993
ne	ni	+ +	Eto *et al.*, 2004
+ +	+	–	Miyata *et al.*, 1999
+ +	+	+	Yamano *et al.*, 2002
+ +	+ +	ne	Tyler *et al.*, 1998
+ +	+	–	
+	–	+	Rivero *et al.*, 2002
+	+ + +	–	D'Angelis *et al.*, 2005
+	+	–	Serrano *et al.*, 2000
+ + +	+	–	

week progressive resistance training walking on a level treadmill at a constant speed of 1.9 m/s, and carrying sheets of lead over their saddle region (wither). They performed a series of progressive sets of weight carrying to fatigue. The ponies carried 44.5 kg for the first set with increases of 22.3 kg per set until fatigue, and a rest period (without weights) of 60-90 s between consecutive steps. This training stimulus resulted in an improved strength (peak weight carried during workout increased by 260%), which was accompanied by substantial increase in whole forearm diameter (19%), individual muscle cross-sectional diameters (7-36%) and selective type II muscle fibre hypertrophy (26%). There were not parallel changes in fibre type composition (contractile properties).

Jumping, as natural form of SSC exercises training, also improves muscle strength through a selective type II muscle fibre hypertrophy of the hindquarters (Rivero and Letelier, 2000). In this study, 10 middle-level show jumpers were conventionally trained for 6 months with three types of exercises: (1) 20-30 minutes of riding exercise (walk, trot and canter) at about 40-65% of the maximal aerobic capacity, 6 days/week; (2) 60 minutes of jumping work with the rider over fences of about 60-100 cm high every second day in the morning; and (3) 60 to 90 minutes of free jumping exercises over fences of 80-110 cm high every second day in the afternoon. This training stimulus produced a very significant hypertrophy of type IIA fibres (23%), and a no-significant hypertrophy of type IIX fibres (8%).

Riding school dressage-type exercises on ground with different slopes (30 to 45 minutes in the morning plus 45 to 60 minutes in the afternoon at about 50% of maximal load, 3 days/weeks for 12 weeks, induced in young untrained Spanish horses a moderate, but significant, hypertrophy of type I (15%) and type IIA (10%) fibres (Rivero *et al.*, 2001). These changes were accomplished with modified kinetics of the trot (i.e., decreased stance duration) compatible with improved strength.

Long-term endurance training programs, with exercises of low intensity and long duration, induce discrete muscle hypertrophy (Table 2). Andalusian horses exercising <50% of maximal load, 60 minutes/day, 5 days/week for 12 weeks resulted in moderate hypertrophy of type IIA fibres (16%), but 5 additional months of progressive endurance training did not increase further myofibre hypertrophy (Serrano *et al.*, 2000). A much more extent and generalized muscle fibre hypertrophy (50%) was documented in endurance Arabian horses with exercises of higher intensity (80% of the maximal aerobic capacity) and long duration (50-60 minutes per session), 4 days/weeks for 12 weeks (D'Angelis *et al.*, 2005; Table 2).

Since the pioneering study by Lindholm and Piehl (1974) in Sweden, it is now well known that interval training, consisting of high-intensity and short-duration exercises, is the training method of choice for optimal racing performance of both standardbreds and thoroughbreds. A significant improvement of strength and stamina can be induced in young mature standardbreds with incremental exercise sessions of 6-10 minutes at 60-100% of the maximal aerobic capacity, 5 days/ week for 16 weeks (Tyler *et al.*, 1998; Table 2). Very interestingly, no additional changes in fibre size were seen in this later study afterwards, even when exercises of very high intensity (80-110% of maximal load) were chronically applied for a long period of 18 weeks more (overload training phase). These data confirms that, in horses, also there is an upper limit in the range of muscle hypertrophic growth induced by training which usually will be in the first 3-4 months of training as maximum. Prolonging and (or) intensifying training beyond this period, although improving stamina, reduces speed and has no effect on strength. Furthermore, trainers, riders and practitioners should be able to recognize when signs of overtraining are present, as this situation reduces performance considerably and is pernicious to the horse's health (see Rivero *et al.*, 2008 for a recent review).

In general, practical training of Thoroughbred racehorses would appear to involve short distance and low-to-moderate intensity workouts because of serious concern for muscle and tendon damage (Miyata *et al.*, 1999). Relevant muscular adaptations to training (i.e., fibre hypertrophy and enhanced aerobic and anaerobic capacities) can be obtained in Thoroughbreds with supramaximal intensity exercises (about 100-165% of the maximal aerobic capacity) of short duration (2-5 minutes) over relatively short distances (1,600-3,600 m), 5 days/ week for 16 weeks (Miyata *et al.*, 1999; Yamano *et al.*, 2002; Table 2). Training-induced myofibre hypertrophic response seems to be more dependent on the intensity than on the duration of the exercise. In adolescent 2 year-old Thoroughbreds, pre-trained basically for 8 weeks, 3 additional weeks of training with higher exercise intensities (equivalent to 100% of the maximal aerobic capacity) applied for 5-15 minutes every second day, may be necessary to enhance muscular strength and power (Rivero *et al.*, 2007).

The importance of high-intensity exercises in the Thoroughbred is to recruit the abundant fast-glycolytic type IIX fibres (Yamano *et al.*, 2006). An increase in the oxidative capacity and size of these fibres, without a concomitant decrease in contraction velocity (i.e., IIX-to-IIA fibre type transition) may cause improvement in strength and stamina of these racehorses. Recent studies in Thoroughbreds have involved high-speed running on flat and sloped track. In general, muscle adaptations to training were similar in horses trained on a slopped (3°) track than in horses trained on a flat track (Miyata *et al.*, 1999). Nonetheless, quantitative electromyographic activity of various locomotor muscles in the hindquarters was significantly higher in horses exercised on the slopped track than those exercised on the flat track, suggesting a differential rate of motor units recruitment. Similarly, smaller peak vertical forces were calculated in the forelimbs and increased peak vertical forces were calculated in the hindlimbs in the Thoroughbred racehorse galloping at high speed on an incline (Parsons *et al.*, 2008). These authors concluded that horses provide the power necessary to elevate their centre of mass up an incline surface by increasing muscle recruitment at low speeds and hence work per stride cycle increases. At high speeds, stride frequency increases to provide power for moving up the slope, perhaps indicating that the hindlimb muscles may be fully recruited during gallop.

Finally, only recently the importance of training in the early stage of development has been considered in Thoroughbreds and properly differentiated by using control groups (Eto *et al.*, 2003). Exercise of relatively low intensity during the first year post-partum induces relevant muscular adaptations, including hypertrophy of types I and IIA fibres (Eto *et al.*, 2003), which cause an improvement in later strength performance.

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Expanded abstracts

Preliminary report into the relationship between farm management and the incidence of OCD in young Andalusian (PRE) horses in Spain

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Take home message

Preliminary analysis of the data collected to date from Andalusian breed young stock on farms in Spain suggests that the farms exhibiting the lowest incidence of OCD had the following common factors:

- Good Pasture Management;
- Controlled Hay Quality;
- pregnant mares that were fed correct amounts of specific nutrients in the last trimester of gestation as set forth by the NRC;
- veterinary involvement in the general management of young stock, rather than only being called on to manage problems;
- an average foal condition score of 5 (out of 9);
- a management plan in place for when the foals develop problems;
- a weaning preparation programme that introduce the foals to feed prior to weaning;
- foals kept in the same pasture during the weaning process;
- foals kept in groups during the weaning process.

Farms exhibiting the highest incidence of OCD had the following common factors:

- very lush or large pastures;

- mare nutrition levels below the minimum levels set forth by NRC;
- foal diets that were not balanced according to NRC minimums;
- an average foal condition score of 6 or more (out of 9);
- were a smaller breeding operation (less than 15 mares);
- had a stocking rate of more than 1 mare per hectare;
- maintain their mares with average condition scores of 7 or more.

Introduction

A host of factors have been suggested to be associated with an increased risk of Osteochondrosis Dissecans (OCD) including a genetic predisposition, nutritional excesses as well as deficiencies and imbalances plus the general management of the mare and foal (Donabédian *et al.*, 2006; Knight *et al.*, 1985; Savage, 1992). Although the true cost of OCD *per se* in loss of revenue from foal sales or in horses breaking down before they realize their full athletic potential perhaps cannot be determined, it is commonly accepted that this is a major problem, which needs to be addressed. Whilst total eradication may not be possible, avoiding dietary or management practices that may increase the risk of a genetic predisposition being expressed has been recommended as a practical route forward.

Little work has been undertaken in Spain to evaluate the potential association between management practices and the incidence of OCD. We have initiated a study to look at the possible relationships in a number of breeds commonly found in Spain. The objective being to identify practices that may be associated with an increased or decreased risk, so that recommendations can be made that may result in an overall decrease in the incidence of OCD in Spain.

Materials and methods

Three hundred and nineteen Andalusian (PRE Pura Raza Espaniola) breed foals and yearlings raised on 10 farms in the areas of Andalusia and Extremadura have been evaluated approximately every 4 to 8 weeks throughout 2007, 2008 and 2009 (to date). Typically, the foals were evaluated from two to 18 months of age. We have gathered 1619 data collections points to date regarding height, weight and body condition score (BCS).

On each occasion body weight was measured using a calibrated electronic animal scale (Model – HorseWeigh Novice Plus), and withers height was determined using a hippometer. The BCS was also recorded, using a scale of 1-9 based on the system developed by Henneke *et al*. (1983) by the same person on each occasion. Average daily gain was calculated and the weight plus height growth curves plotted using specialized equine growth tracking software (Gro-Trac, Kentucky Equine Research, USA).

The farm manager, or the veterinarian, responsible for the breeding program completed a detailed questionnaire which covered the general management and other aspects of the farm as well as the procedures undertaken in preparation for foaling and weaning. The questions included the common breeding management to be able to compare the data with breeding and management studies currently published (Ott and Kivipelto, 2005; National Research Coucil NRC, 1997; Sondergaard, 2003; Weeks *et al*., 2005). Wherever possible the answers were crosschecked during the regular evaluations undertaken by the research team on each premise.

Many of the questions asked were used to cross check other questions to see if there were anomalies in the answers. E.g. we ask 'Is there someone on the premises to attend the mares during the births' and 'does someone make sure that the foals receive colostrum'. If the answers to both questions did not coincide, we delved further to better understand the farm practices. Statistically, some of these questions do not look significant, but as we are both checking practices and continuing to include more farms in the study, this may change.

At least 30% and up to 100% of the yearlings on each farm were randomly chosen to undergo a radiographic analysis, which was carried out between 18 and 24 months of age. The percent of OCD was determined by the radiographic diagnosis. The radiography was carried out according to the standard accepted by the Ministry of Agriculture, set forth by the University of Córdoba, and consisted of 10 radiographic views per horse. Only lesions diagnosed as Grade 2 or Grade 3 were considered to confirm the presence of OCD in the foal. The exact views for the radiographs taken and information related to diagnosis and grading is set forth by the University of Cordoba (http://www.uco.es/empresa/hcv/ostecondrosis.php).

The size and type of the breeding operation determined the percent of horses included in the radiological study. Many of the participating farms actually undertake such a radiographic evaluation on 100% of their yearlings as part of their standard management.

Standardized answers from the questionnaire, % of OCD on each farm and growth information have been submitted to a multiple regression statistical evaluation using farm as a random effect with the statistical software STATGRAPHICS Plus at the Universidad Complutense de Madrid.

Results and discussion

The incidence of OCD on the farms evaluated ranged from 0-34.3%. A percentage of 12 or less was considered to be a low incidence with a percentage of 20 or above being a high incidence.

Preliminary investigations suggest that farms with the lowest incidence of OCD share certain management and nutritional practices. Similarly those farms with the highest incidence of OCD also seemed to have practices in common. Further work is needed to confirm these potential relationships in larger number of animals and farms.

The study is still ongoing and it is hoped that with the inclusion of more young horses a better understanding of the potential management and nutrition associated risk factors for OCD in Spain can be determined. Clear advice can then be given to breeders and veterinarians with the aim of reducing the incidence of OCD associated with certain management or nutritional practices. In the future it is hoped that information can be obtained for other breeds and other developmental orthopedic diseases of the horse in Spain.

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The effect of a feed ingredient with endogenous phytase activity on phosphorus availability in equine diets

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Take home message

Inclusion of Allzyme® SSF (Alltech inc.) at 0.4kg/tonne in a concentrate feed, where the inclusion of dicalcium phosphate had been reduced, was associated with decreased daily phosphorus (P) output (P<0.05) and a concomitant increase in apparent P digestibility (P<0.05). There was also a clear trend towards reduced faecal output of phytate-bound phosphorus (Phyt-P) and an increase in its digestibility ($P=0.08$). Whilst there was no significant difference in magnesium (Mg) digestibility, there was again a clear trend for increased apparent Mg digestibility associated with the Test diet, however, this did provide a small but significantly higher daily amount of Magnesium. This preliminary data, suggests that the fortification of horse feed with ingredients such as Allzyme® SSF, produced using solid-state fermentation technology, warrants further investigation as it has the potential to improve endogenous P availability from the diet thus reducing the reliance on inorganic P sources. This is particularly relevant for circumstances where apparent P digestibility may be reduced, such as in aged animals, or those with sub-optimal digestive function due to cumulative worm damage or previous colic resection.

Introduction

Dietary phosphorus (P) is essential given its ubiquitous requirement in cellular metabolism. Endogenous P in many feed ingredients has limited availability in horses, due to its presence in the form of phytate or phytic acid (inositol hexaphosphate). Soya, oats (and bi-products), maize and wheat bran, which are all common ingredients in horse feed, possess relatively high phytate concentrations (Selle *et al.*, 2003). Phytates are salts formed from the interaction between phytic acid and dietary minerals including calcium (Ca), magnesium (Mg), iron or zinc. In addition to the primary effect on P availability in feed, a high dietary phytate concentration can also inhibit Ca and Mg absorption through their chelation within the digestive tract to form insoluble phytate salts.

Phytate can be degraded by phytase enzymes that are naturally present in some feed ingredients, or found to a limited extent within the digestive tract of animals. Phytase can also be produced by bacteria and fungi such as *Aspergillus niger spp* and this has led to the commercial production of feed ingredients using solid-state fermentation technology (Jorquera *et al.*, 2008). Fortification of animal feed with exogenous phytase has been reported to significantly improve phytate-derived P (Phyt-P) digestibility in pigs, poultry and other livestock (Lei and Porres, 2007). In horses, effects have been equivocal. Added phytase did not significantly affect faecal P loss, or P digestibility, at application rates between 300-1000 FTU/kg concentrate feed (Patterson *et al.*, 2002; Van Doorn *et al.*, 2004), although, apparent Ca digestibility increased with phytase added at 1000 FTU/kg (Van Doorn *et al.*, 2004). Conversely, faecal P output was decreased using a different phytase source, at a higher application rate (3,000 FTU/kg), although this effect was dependent on dietary composition and was apparent only when applied to a 'sweet feed' containing a ration where the level of insoluble or Phyt-P was presumed to be high (Hainze *et al.,* 2004).

The interest in the use of ingredients produced using solid-state fermentation technology in horse feeds has increased recently owing to the significant increases in the market price for the main feed P additives, such as dicalcium and tricalcium phosphate. This, coupled with the shift in global economic climate, has led to further pressure on margins for horse feed. Feed ingredients that offer the potential for improved endogenous P availability from the diet are therefore of considerable practical interest.

This study aimed to investigate the effect of a feed ingredient Allzyme® SSF (ASSF) (Alltech inc.), produced using solid-state fermentation technology, on the retention and apparent digestibility of dietary P, Phyt-P, Ca and Mg.

Materials and methods

During a crossover feeding study, 8 equids including 7 Thoroughbred horses (5 geldings, 2 mares) and 1 pony (gelding); mean body weight (BW) 503±101 kg (range 272-585 kg), were randomly assigned to one of two diets. The Control diet, comprised a ration of haylage plus concentrate feed, whilst the Test diet, additionally incorporated ASSF. Each dietary period consisted of a 16-day acclimatisation period, a 4-day collection period and a 7-day washout period, before the dietary groups were switched and the procedure repeated.

Diet composition

Horses were offered haylage on a semi *ad libitum* basis, with a maximum of 8.5 kg dry matter (DM) being fed per day. Whilst the daily intake of haylage varied slightly between individuals, it was very consistent throughout the trial for a given individual. The concentrate feed ± ASSF (0.4 kg/tonne) was fed 3 times per day at a level sufficient to satisfy energy requirements (NRC, 2007), and the total amount offered per individual was fixed for the duration of the study. During the last two days of the acclimatisation period and throughout the collection periods, animals were housed in loose boxes and bedded on wood shavings. Regular and controlled light exercise was undertaken by all animals.

The composition of the Control and Test concentrate feeds (Table 1) was formulated to satisfy daily requirements for energy, protein and vitamins and minerals (NRC, 2007). In combination with haylage the NRC daily requirement for Ca, P and Mg was exceeded. The level of dicalcium phosphate added to the Test concentrate was reduced by 0.8% and ASSF was added to this feed at an inclusion level of 0.4 kg/tonne. Common ingredients were used in both the Control and Test concentrate feeds and minimal changes were made to the Test feed formulation to accommodate the addition of ASSF and removal of dicalcium phosphate. The inclusion of dicalcium phosphate was

Table 1. Ingredient composition (%) of the Control and Test concentrate feeds.

% Inclusion	Control concentrate diet	Test concentrate diet
Soya hulls	30.00	30.00
Oatfeed	19.87	19.87
Molasses derivative	8.53	8.53
Micronized soya flake	8.00	8.00
Micronized peas	6.00	6.00
Micronized maize	5.00	5.00
Wheatfeed	4.45	4.45
Hipro soya meal	4.20	4.20
Sunflower meal	4.20	4.20
Wheatbran	4.00	4.00
Soya oil	2.21	3.02
Dicalcium phosphate	1.59	0.74
Calcium carbonate	0.99	0.99
Vitamin & mineral premix	0.54	0.54
Sodium chloride	0.44	0.44
ASSF	----	0.04

decreased in the test concentrate diet in order to reflect potential practical use of ASSF to reduce the reliance on inorganic P in proprietary feeds. Whilst the level of P calculated for the Test concentrate was 0.15% below the control concentrate, it continued to provide P at a level to easily satisfy the published daily requirement for P for horses at maintenance and in work when fed in conjunction with forage. Removal of the dicalcium phosphorus was accommodated by the addition of soya oil, in order to maintain the same level of inclusion of other endogenous P containing raw materials. Soya oil (refined) was used due to its very low reported P content (<0.02%) compared to the other ingredients present (Sinram, 1986). The calculated and analysed mineral contents of the Control and Test concentrate feeds and haylage are shown in Table 2.

Sample collection and analysis

Faeces and urine were collected using collection harnesses (Stable Maid®, Melbourne, Australia). For the geldings, urine and faeces

Table 2. Analysis of the daily diet (% DM) (calculated where applicable in brackets) and mean daily intake for P, Ca, Mg and Phyt-P for the Control and Test (ASSF) diet.

	% of diet			Daily intake (day)	
	Haylage	Control - ASSF	Test + ASSF	Control - ASSF	Test + ASSF
DM	70.4	87.9	87.9	9.1±2.0kg	9.1±1.8kg
Phosphorus	0.25	0.68 (0.74)	0.64 (0.57)	35.5±7.9g	34.4±7.2g
Calcium	0.36	1.23 (1.38)	1.19 (1.14)	57.7±13.0g	56.5±12.0g
Magnesium	0.11	0.23 (0.25)	0.30 (0.25)	13.4±3.0g	15.6±3.3g[1]
Phytate-P	0.24	0.52	0.56	29.9±6.6	31.1±6.4

[1] (P<0.05).

were collected separately, whereas in the mares they were collected combined. For each collection period, representative samples of urine, faeces (or urine + faeces) were taken using commonly accepted methods.

Feed, haylage and faecal samples were oven dried (100 °C, 24 or 48 hours), milled and DM calculated. Feed samples underwent a nitric acid microwave digestion, whereas faecal samples were digested with hydrochloric-nitric acid (Aqua Regia digestion). Feed and faecal digests were analysed for P, Ca and Mg content by inductively coupled plasma optical emission spectroscopy or ICP-OES (Perkin Elmer OES Optima 4300DV).

Urine sub-samples were thoroughly mixed and acidified (~ pH 2 using 6 M hydrochloric acid) to solubilize sedimented minerals. Ca and Mg concentration, and urinary creatinine concentration were determined using standardised and commonly employed clinical chemistry methodologies on automated analyzers, whilst urine P was analysed using ICP-OES.

Statistical analysis

Statistical analysis was carried out using Sigma Stat® (SPSS Science Inc). Where the data was normally distributed, a paired T test was carried out to establish significance. Where the data was not normally distributed, the non-parametric test (Wilcoxon signed rank test) was applied. The significance level was set at a confidence interval of 95% and reported at (P<0.05).

The data relating to faecal P for the two mares was included in the statistical analysis as faeces are the main route for P loss (Schryver *et al.*, 1972). In addition, whilst the level of P lost in urine can increase with increased dietary P intake, the excretion of P in urine remains negligible in comparison to faecal losses for both low and high P containing diets (Van Doorn *et al.*, 2004). However, as Ca and Mg are excreted extensively in both faeces and urine the data from the two mares was excluded from the statistical analysis with respect to Ca and Mg.

Results

Whilst the removal of a proportion of dicalcium phosphate was calculated to decrease % P by 0.15% in the Test concentrate, the subsequent analysis, which was carried out on a composite sample of 13 grab samples, revealed that % P was only decreased by 0.04% compared to the Control concentrate. This is likely to have been due to a combination of manufacturing, sampling and analytical variation. The coefficient of variation for the analysis of P by ICP-OES was stated to be 3.7%.

Intake

There was no statistically significant difference in the intake of haylage, concentrate feed, or water between Control and Test diets (P>0.05). Mean daily intake of haylage during the collection periods was 1.23±0.23 kg DM/100 kg BW and 1.24±0.18 kg DM/100 kg BW for the Control and Test diets respectively, whilst mean intake of the concentrate feed was 0.58±0.10 kg DM/100 kg BW for both the Control and Test diets, respectively. Mean daily water intake was 6.9±1.3 l/100 kg BW and 7.2±2.0 l/100 kg BW for horses receiving the Control and Test diets, respectively.

There was also no significant difference in the intakes of Ca and P (g/day) between the Control and Test diets (P>0.05). However, there was a small but significant difference in Mg intake, 13.4±3.0 and 15.6±3.3 g/day, between the Control and Test diets (P<0.05).

There was no significant difference in Phyt-P intake between the Control and Test diets respectively (P>0.05). However, as was expected due to the removal of 0.8% of dicalcium phosphate, Phyt-P accounted for a greater percentage of the total P content in the Test (86.0%) compared with the Control concentrate feed (75.3%).

Output

Mean daily faecal output (DM) was 4.4±1.3 kg and 4.2±1.1 kg for the Control and Test diets, respectively, and there was no significant difference in faecal output between diets (P>0.05).

Addition of ASSF to the Test diet was associated with a significant decrease in mean total faecal P output (g/day) (P<0.05) (Table 3, Figure 1) and a significant increase in mean apparent P digestibility (%) (P<0.05) (Table 4, Figure 2).

Table 3. Daily faecal output in g/day of P, Ca, Mg and Phyt-P (mean ± SD) and effect of dietary supplementation with ASSF.

	Control diet		**Test diet**		**Statistical difference**
	Mean	**SD**	**Mean**	**SD**	
Phosphorus	29.1	6.2	25.9	6.0	(P = 0.023)
Calcium[1]	27.2	7.0	26.7	9.4	NS
Magnesium[1]	5.8	1.5	5.8	1.5	NS
Phytate-P	16.6	5.0	15.4	3.9	NS

[1] n=6.

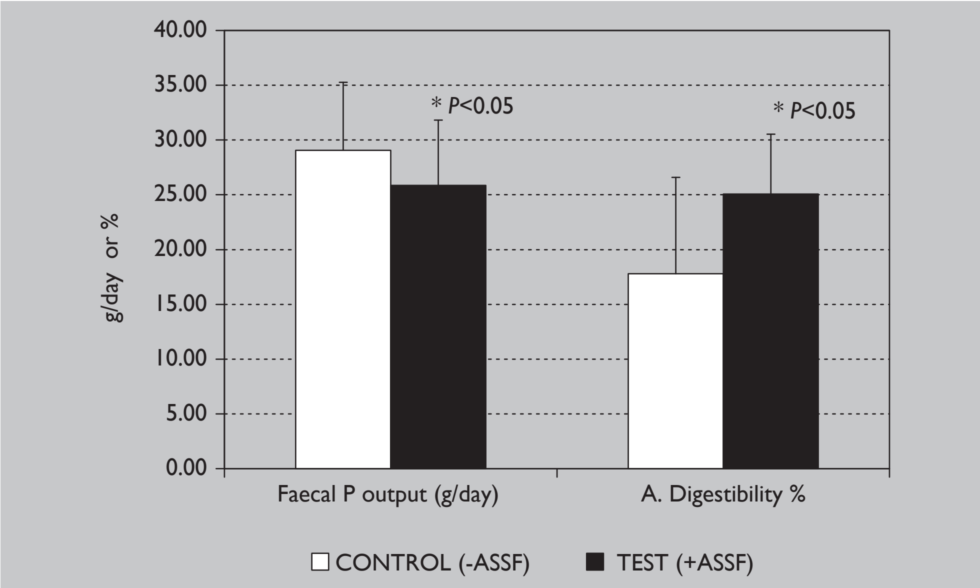

Figure 1. Effect of the addition of ASSF to the diet on mean faecal P output (g/day, mean ± SD) and on mean apparent P digestibility (%, mean ± SD).

Table 4. Apparent digestibility (%) of P, Ca, Mg and Phyt-P (mean ± SD) and effect of dietary supplementation with ASSF.

	Control diet		**Test diet**		**Statistical difference**
	Mean	**SD**	**Mean**	**SD**	
Phosphorus	17.8	8.8	25.1	5.5	(P = 0.039)
Calcium[1]	50.6	7.7	52.9	7.0	NS
Magnesium[1]	54.4	7.5	61.7	4.1	NS (P= 0.11)
Phytate-P	45.3	6.49	50.38	7.67	NS (P = 0.08)

[1] n=6.

Mean daily faecal output of Phyt-P was decreased in 6 out of 8 of the horses when fed the Test diet, but this was not statistically significant (P>0.05, Table 3). Likewise, Phyt-P digestibility (%) was increased in 6

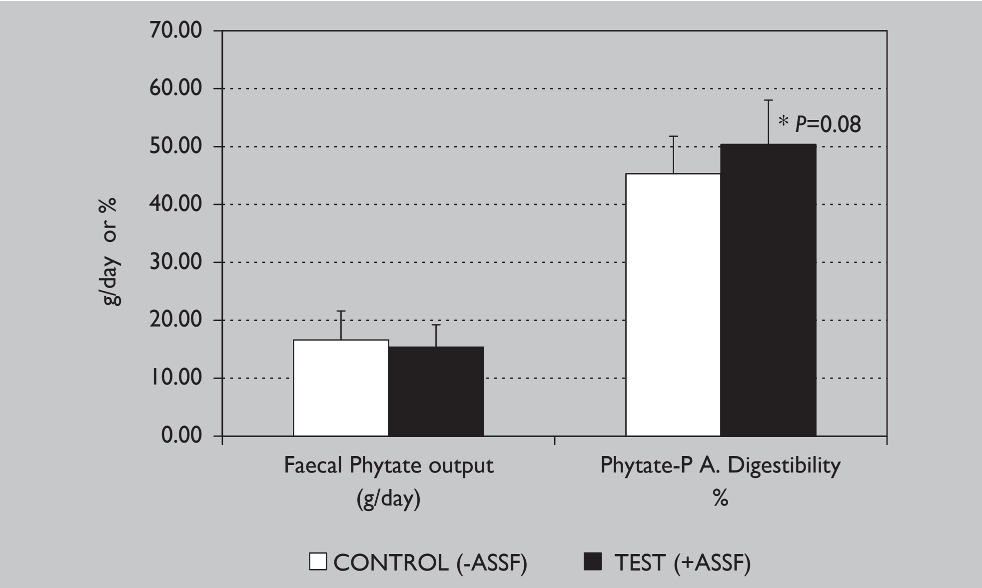

Figure 2. Effect of the addition of ASSF to the diet on mean faecal Phyt-P output (g/day, mean ± SD) and on mean apparent phytate-p digestibility (%, mean ± SD).

out of 8 horses when fed the ASSF supplemented diet, although again this result failed to attain statistical significance ($P = 0.081$, Table 4).

There was no significant difference in faecal output or apparent digestibility of Ca between the Test and Control diets ($P > 0.05$). There was, however, a clear trend towards an increase in apparent Mg digestibility (%), with the Test diet ($P = 0.1$) that was apparent in 5 out of the 6 geldings, although the faecal output of Mg remained unchanged ($P > 0.05$) (Tables 2 and 3). Additionally, there were no significant effects of diet on the mineral creatinine ratio in urine with respect to Ca, P or Mg ($P > 0.05$).

Discussion and conclusions

In agreement with Hainze *et al.* (2004), this trial reported a reduction in daily faecal output of P, in response to the addition of an ingredient, produced by solid-state fermentation, to a coarse mix type feed. In both the current and earlier studies the feed ingredient, produced using solid-state fermentation technology, was ASSF. In contrast to Hainze *et al*. (2004), the present study additionally reported an increased apparent P digestibility (%) and a trend towards an increased apparent Phyt-P digestibility (%). These results were observed despite a significantly lower estimated delivery of phytase by the Test diet (~ × 6) compared to that of Hainze *et al*. (2004). Whilst the total P content of the concentrate feeds in the current study and that of Hainze *et al*. (2004) was comparable, the Phyt-P:total P ratio may have differed. The ASSF was also added to the concentrate feed during manufacture in the current study, in contrast to being fed directly to the horse in the earlier trial of Hainze *et al*. (2004). Additionally, horses were fed 3 small meals per day in this study, which may have increased the opportunity for, or duration of phytase activity within the stomach.

The removal of 8.5 kg/tonne of inorganic P as dicalcium phosphate would have been expected to decrease faecal output of P of inorganic origin and may also have contributed to the observed change in apparent total P digestibility. However, the actual analysis of the Control and Test concentrates indicated close equivalence in % P, with only 0.04% difference between them. This would be expected to lessen the potential effect of differential P intake on faecal P output and digestibility. However, it should be noted that if the true level of P (%) in the concentrate feeds was as expected and the results were calculated on this basis, then the apparent P digestibility no longer shows a significant difference.

Phyt-P output and digestibility would seemingly be unaffected by inorganic P intake. There was a clear trend towards reduced Phyt-P output and increased apparent Phyt-P digestibility (%) suggesting an increased liberation and absorption of Phyt-P. This would also have contributed to the changes reported in faecal output and digestibility of total P. The apparent digestibility (%) of Phyt-P reported in this study is numerically much higher than that of total P, which has been previously observed in rabbits (Marounek *et al*., 2003). The dorsal large colon and small colon are both major sites of P absorption in horses (Schryver *et al*., 1972). The main site of action of exogenous phytase is reported to be the stomach in other species (Lei and Porres, 2007). Supplementation with exogenous phytase may therefore increase the pre-caecal breakdown of phytate, increasing the opportunity for additional P absorption in the small intestine. Whilst Phyt-P may be degraded efficiently by bacterial phytases in the stomach and hindgut, only a proportion of the P liberated will be absorbed, either in the small intestine or colon, with the remainder contributing to daily faecal P output. This can potentially contribute to an overestimation of Phyt-P digestibility.

Supplementation of the diet with oil has previously been suggested to adversely affect caecal fermentation in horses thus reducing digestibility of ADF, NDF and crude fibre (Jansen *et al*., 2002, 2007), although these findings are not unequivocal (Kronfeld *et al*., 2004). However, several studies have failed to show any adverse effect of fat supplementation on the digestibility of Ca, P and Mg (NRC, 2007). Additionally, as the inclusion of soya oil to the test concentrate was only 0.8% above that of the control in this trial, it is unlikely to have had any physiologically relevant effect.

The increase in Mg digestibility (%) that was observed in this study in association with ASSF suggests improved absorption of Mg from Mg-phytate salts present in the diet. However, there was a small but nevertheless a statistically significant increase in Mg intake per day with the Test diet. Whilst this may have contributed to the suggested increase in digestibility, the effect of the ASSF cannot be discounted. In contrast, the absence of any effect of ASSF on dietary Ca apparent digestibility (%) may have reflected the relatively high Ca content of the diet relative to requirements in these animals.

The use of the data from the two mares and pony could be viewed as a limitation of this study and so for interest some further statistical analysis was undertaken with the mare and pony data being discounted. In this instance, there was no longer a statistically significant effect of the ASSF supplementation with respect to Faecal P output (P>0.05), P digestibility (P>0.05) or Mg digestibility (P>0.05). It is however, relevant to point out that removal of the mare and pony data considerably reduced the power of the statistical analysis applied. In contrast the power of the statistical test applied with respect to Phytate-P output g/day and Phytate-P digestibility was considerably increased by removal of the mare and pony data. The previously described trend towards a decrease in Phytate-P output and increased Phytate-P digestibility with ASSF supplementation became significant for both these parameters (P<0.05, P<0.01), respectively by removal of the mare and pony data. The anomaly in these results suggests that any future studies carried to further investigate the effect of ASSF supplementation should evaluate a larger numbers of horses in order to improve the power of the statistical analysis that can be applied to the data giving more rigour to the results.

Studies to date on the effect of exogenous phytase on mineral digestibility in horses have been contradictory. This may in part be due to the source and activity of added phytase, as well as the nature of the diet, making interpretation and practical application more difficult. For clarity, this study sought to investigate the effect of an additive, produced using solid-state fermentation technology, on mineral digestibility using a ration formulated to include ingredients commonly used in proprietary concentrate feeds, with comparable levels of Ca, P and Mg. The level of ASSF applied to the concentrate feed was also added at a commercially viable inclusion rate of 0.4 kg/tonne.

In conclusion, the results from this preliminary trial suggest that the use of ingredients produced using solid-state fermentation technology, such as Allzyme® SSF to horse feed, warrants further investigation and may offer some potential to improve P availability from endogenous sources. Some further investigation using diets with equal provision of Ca, P and Mg would be beneficial, especially where greater statistical power could be applied through the use of a larger group of horses. Increasing the digestibility of Phyt-P would allow P status to be improved in circumstances where apparent P digestibility may be reduced, such as in older horses, those with cumulative worm damage (Ralston, 2007) or following colic surgery involving significant hindgut resection (Ralston *et al.*, 1986). In addition, this may also be an economically viable alternative to simply increasing the inclusion of inorganic P. This is especially relevant at this current time with the price of inorganic P being high worldwide.

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Non-invasive bone assessment by quantitative ultrasonography in Lusitano suckling foals: a preliminary study

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Take home message

The present work provides data on cortical bone assessment by quantitative ultrasonography in the Lusitano suckling foal. Speed of sound (SOS) measurements at the lateral aspect of MC III increased with age, but age had no significant effect on the dorsal measurements. Quantitative ultrasonography could be a practical and non-invasive method to assess superficial cortical bone properties in the growing horse.

Introduction

Nowadays, skeletal development and soundness of bone tissue is one of the major concerns for the sport horse industry. Quantitative ultrasonography (QUS) is a quick non-invasive technique, already applied in the horse as a means of assessing superficial cortical bone properties (Carstanjen *et al.*, 2002, 2003a,b). With this method, the sound waves' shortest propagation time through the bone in axial transmission mode is calculated to determine the ultrasound's velocity or speed of sound (SOS m/s) which depends on both Young's modulus (or elasticity modulus) and bone mineral density (Carstanjen *et al.*, 2002). QUS therefore provides an indication of both the elasticity and bone mineral density of the superficial cortical bone. Previous work has suggested that elasticity and other mechanical properties such as the bending strength of the cortical cannon-bone are positively correlated with its mineral content, in growing horses (Bigot *et al.*, 1996). The main objective of this study was to evaluate the variation of superficial cortical bone properties in Lusitano foals, from birth until weaning, by the assessment of ultrasound speed in the third metacarpal bone.

Materials and methods

Eighteen Lusitano foals from 3 stud farms (group A: n = 6, B: n = 6, and C: n = 6) were monitored at approximately 1.5 months of age and then every two months until weaning (weaning average age: 228±6 d). All foals were kept on pasture with their mothers. Mares of groups A and B were supplemented once a day with compound feeds plus grass hay or cereal straw, according to pasture availability and farm practices. Mares of group C were rarely supplemented. Every two months, foals were subjected to measurements of speed of sound (SOS) in axial transmission mode on the mid section of the right third metacarpal bone (MC III) (dorsal and lateral aspects) with a QUS device (Sunlight EQUS, Sunlight Medical, Israel). The measurements were taken with the foals in a standing position. They did not require sedation. SOS data were grouped in 4 age classes (average±SD): 1.6 (48±16.8 days), 3.5 (106±18.6 days), 5.4 (162±17.0 days), and 7.3 (219±17.6 days) months old. Data were analysed using the MIXED procedure of SAS (SAS Institute Inc., Cary, NC, USA). The model considered the group, age and interaction group × age as fixed factors and foals as random effect. Measurements at different ages in the same foal were treated as repeated measures on animal within group as a subject. Averages and standard error of means of the dorsal and lateral aspects of MC III were used to adjust quadratic models by non-linear regression (Graphpad Prisma 4.0, Graphpad Software, S. Diego, CA, USA). The effect of the measurement aspect of MC III (dorsal vs. lateral) was analysed independently of each age class by a mixed model considering the animal as random factor and the aspect as fixed factor, and treated as repeated measures on animal as a subject.

Results

The interaction between group and age was not significant. Therefore, the results regarding the effect of group and the effect of age are presented separately. SOS values for the dorsal aspect measurements of group C tended to be lower ($P = 0.052$) than the other two groups. There were no significant differences between groups concerning SOS values at the lateral aspect of MC III (Table 1).

Values of SOS measurements at the lateral aspect of MC III increased significantly with age ($P<0.001$). The effect of age was not significant on dorsal aspect measurements (Table 2).

However, a good adjustment was obtained when quadratic models were fitted to describe the variation of SOS values obtained at the dorsal and lateral aspects of the third metacarpal bone during the considered period (Figure 1).

Table 1. Speed of sound (SOS, m/s) means and standard errors of the dorsal and lateral aspects measurements on the MC III in the three groups of foals.

Aspect	**Group**			**P value**
	A	**B**	**C**	
Dorsal	3,937±23.9	3,933±23.7	3,858±24.9	0.052
Lateral	4,183±31.6	4,207±31.3	4,147±33.2	0.426

Table 2. Speed of sound (SOS, m/s) means and standard errors of the dorsal and lateral aspects measurements on the MC III in the four considered group ages.

Aspect	**Age (months)**				**P value**
	1.6	**3.5**	**5.4**	**7.3**	
Dorsal	3,878±19.1	3,903±18.2	3,928±20.0	3,928±18.6	0.167
Lateral	4,107±26.3^{a}	4,168±24.7^{b}	4,200±28.3^{bc}	4,240±25.4^{c}	0.0007

^{a,b} Means in rows with different superscripts are significantly different ($P<0.001$).

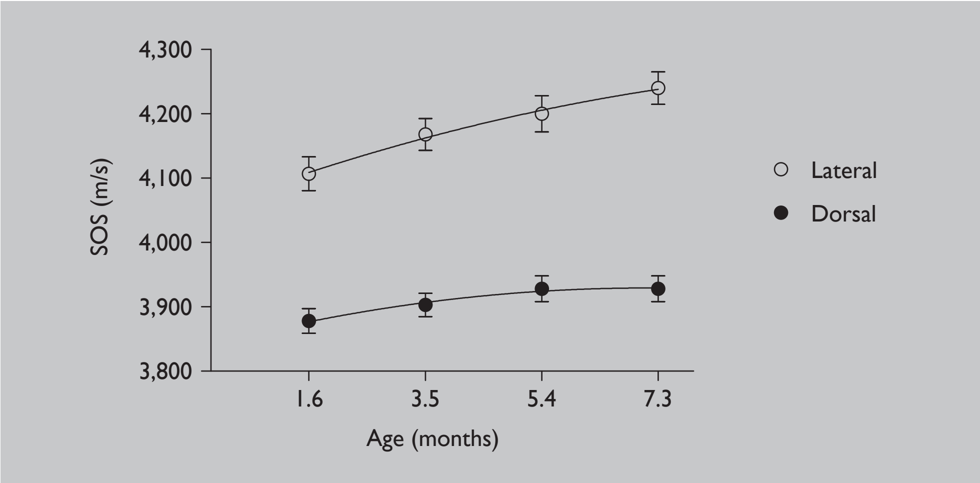

Figure 1. Models of variation of SOS values obtained at dorsal (D) and lateral (L) aspects of the MC III. Regression equations: $D = 3842+24.62t-1.73t^2$ ($R^2 = 0.98$); $L = 4056+35.63t-1.45t^2$ ($R^2 = 0.99$) t = age (1.6, 3.5, 5.4 and 7.3 months).

SOS values for lateral aspect measurements were significantly higher than SOS values for dorsal measurements at all considered ages ($P<0.0001$).

Discussion and conclusions

A previous study in young foals suggested that diet and time can influence bone mineral content of the third metacarpal bone (Hoffman *et al.*, 1999) as monitored using radiographic absorptiometry. As SOS values could also reflect bone mineral density, the lower values for SOS recorded at the dorsal aspect for group C (basically an unsupplemented group) could potentially be a management effect, probably related with the mares' feeding regimen, which was highly dependent on the quality and quantity of natural pastures.

The significant increase of SOS values with age at the lateral aspect of the MC III is in agreement with the observations for young Thoroughbreds (Carstanjen *et al.*, 2003b). Although other SOS data in suckling foals are lacking, consistently higher SOS values at the lateral aspect than SOS values at the dorsal aspect, have also been reported for older horses (Carstanjen *et al.*, 2003b). These differences could be linked with the geometry, structure and biomechanical properties of MC III in the horse. When compared to the lateral region, the dorsal region of MC III has been shown to contain smaller osteons (Martin *et al*., 1996a). Furthermore, in the MC III collagen fibres are regionally oriented, with the highest longitudinal fibre values in the lateral region (Martin *et al*., 1996b).

The present work provides preliminary data on cortical bone assessment by QUS in the young Lusitano horse. This non invasive technique could be important in the evaluation of bone quality and potentially for the detection of an increased risk of fracture in horses, because it may provide an indication of the bone mineral density as well as its mechanical properties, which are linked to the ability to withstand physical stress. However, further research should be pursued.

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Effect of long-term conditioning period on digesta total tract mean retention time and biochemical faecal profile in endurance horses

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Take home message

We recently demonstrated that the total tract apparent digestibility of organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF) and hemicelluloses was significantly higher after a long-term conditioning period in endurance horses (Goachet *et al.*, 2009a). This does not seem to be due to changes in digestive microflora concentrations or activities.

Introduction

In human athletes, the physiological effects of exertion on digestion have been reported to differ whether it is a punctual exercise or long-term conditioning. Intense and acute exercise can lead to gastro-intestinal disturbances, although regular physical activity leads to an improvement of the digestive physiology and health on the long term (Peters *et al.*, 2001). In horses, digestion may also be affected by work or exercise in such a way that it is improved by light exercise and inhibited by heavy work (Olsson and Ruudvere, 1955; Orton *et al.*, 1985; Pagan *et al.*, 1998; Katsuki *et al.*, 1998; Bergero *et al.*, 2002).

In endurance horses, that face nutritional challenges, the investigation of the impact on digestion of exercise is essential, particularly in terms of energy, a key point in performance. We recently demonstrated that a long-term conditioning period had a positive effect on the digestive efficiency in endurance horses (Goachet *et al.*, 2009a): total tract apparent digestibility of organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF) and hemicelluloses was significantly higher after 11 and 22 weeks of conditioning than before conditioning. This study aimed at determining whether the increase in OM and fiber total tract apparent digestibility observed with endurance conditioning was due to changes in mean retention time (MRT) of digesta and/or in digestive microflora concentrations or activities.

Materials and methods

Eight endurance horses (pure-bred and cross-bred Arabians, aged from 7 to 13 years, 418±22 kg) underwent a regular endurance training programme for 8 consecutive months. Basal training consisted in 20 km outdoor-rides at low speeds (walk and trot mainly) every two days, and was completed with specific training sessions on a racetrack (2 hours gallop at 18 km/h) performed three weeks before each race. Each horse competed in two 120 km-endurance events, after 11 and 22 weeks of conditioning. In order to isolate the effect of a bout of exercise from the effect of long-term conditioning, horses were not exercised during the measurement periods.

Horses were fed meadow hay (29% cellulose, 62% NDF, 34% ADF) and pelleted feed (commercial horse feed containing 25% alfalfa, 20% barley, 20% wheat bran, providing 12% CP, 3.7% fat, 23% starch, 13% cellulose, 32% NDF and 17% ADF). During the four weeks preceding each race, they also received dietary oil (commercial oil containing sunflower, rapeseed and grapeseed oils) to provide 5% fat in the whole ration. With the exception of the race days, hay and concentrate were offered in two equal meals, at 10:00 and 16:00 and at 8:00 and 17:30 respectively. Dietary oil was offered top-dress with each concentrate meal. The amounts of feed offered were determined to maintain stable body weight and condition scores. During the measurement periods, the diet was fixed at 1.9% BW and was composed of 75% meadow hay/25% pelleted feed. The average quantity of dietary oil was 232 ml/day.

MRT of liquid and solid phases, fecal microbial counts and biochemical parameters measurements were done after a 3 months 'winter rest' before the beginning of the conditioning period (P0), and after each of the two endurance races (P1 and P2 respectively), during the second week of the resting period allowed to horses (Goachet *et al.*, 2009b).

For MRT measurements, a partial collection of faeces was performed during three consecutive days (Goachet *et al.*, 2009c). A single meal of Europium-labelled hay (60 g) and Ytterbium-labelled pellets (60 g) mixed with sugar (sucrose) to increase palatability was offered to each horse at 16:30 on the first day of the collection period. A Cr-EDTA solution (125 mL) was administered via a naso-oesophagal tube at 17:00 the same day. Faeces collection began at 20:00 and continued every 2 hours during the three following days. The faecal collection consisted in taking faecal material from the horse bedding.

For microbial enumeration and activity measurements, an additional sample of faeces was taken from the rectum of each horse, at 13:00 on day 3 of the sampling period. A sub-sample for microbial counts (total viable anaerobic bacteria, lactic acid-utilising bacteria, *Lactobacilli spp, Streptococci spp* and cellulolytic bacteria) was immediately placed in a sterile CO_2-saturated flask, which was held at 38 °C in a water bath between collection and inoculation. A second sub-sample of faeces was filtered (Blutex 100 µm) and pH of filtrate was measured with an electronic pH-meter (MP 120, Mettler, Toledo, Spain) immediately after collection. The filtered faeces were then immediately frozen (-20 °C) for determination of D- and L-lactate (1 ml) and volatile fatty acids (VFA).

Statistical analysis of the data was performed using the GLM procedure of SASv8. Means were calculated for all variables and compared using linear contrasts (pdiff option of SAS).

Results

Five horses were able to perform the entire conditioning program and the three measurement periods (P0, P1, P2). Three others developed lameness during the training period (before the first or the second race). The chemical composition of the whole diet was constant within the three digestion trials (Table 1).

At P0, total anaerobes, lactic acid utilizing bacteria and cellulolytic bacteria concentrations were 7.3, 6.7 and 6.3 log_{10} CFU/g respectively. No difference was detected between P0, P1 and P2 (Table 2). At P0, *Lactobacilli* concentration was 5.8 log_{10} CFU/g, and was significantly lower at P1 but not at P2 (P<0.05). Due to technical problems, *Streptococci* concentrations were not determined at P0 and P1.

At P0, total VFA, acetate, propionate and butyrate concentrations were 48.8, 34.3, 9.5 and 2.8 mmol/l respectively. Acetate, propionate and butyrate represented 69.6, 20.6 and 5.6% of the total VFA. The (acetate + butyrate/propionate) (C2+C4/C3) ratio was 3.8. The faecal pH was 7.0 (Table 2). For these parameters, no differences were detected between P0, P1 and P2. At P0, D- and L-lactate concentrations were 3.6 and 2.1 mmol/l respectively. D- and L-lactate concentrations were lower at P2 than at P0 and P1 (Table 2).

Table 1. Chemical composition (g/kg DM) of the whole diet (75% hay/25% concentrate) for each digestion trial (P0, P1 and P2).

	P0	P1	P2
DM	892	901	886
OM	923	917	934
NDF	554	552	565
ADF	299	298	292
ADL	42.4	42.4	42.3
Hemicellulosis (= NDF - ADF)	259	254	273

Table 2. Mean microbial concentrations, pH and VFA concentrations in faeces before endurance conditioning (P0), after 11 weeks (P1) and 22 weeks (P2) of endurance conditioning.

n=5	P0	P1	P2	SEM	p
Total anaerobic bacteria (log_{10} CFU/g)	7.3	7.1	7.2	0.17	ns
Lactate utilizing bacteria (log_{10} CFU/g)	6.7	6.3	6.1	0.21	ns
Lactobacilli (log_{10} CFU/g)	5.9[a]	5.4[b]	5.6[b]	0.06	0.01
Streptococci (log_{10} CFU/g)	nd[1]	nd	5.1		
Cellulolytic bacteria (log_{10} MNP/g)	6.3	5.8	5.8	0.22	ns
pH	7.0	6.6	6.8	0.11	ns
D-lactate (mmol/l)	3.6[a]	5.4[a]	0.4[b]	0.94	0.02
L-lactate (mmol/l)	2.1[a]	2.2[a]	0.1[b]	0.55	0.008
Total VFA (mmol/l)	48.8	57.4	56.8	6.24	ns
Acetate (mmol/l)	34.3	39.2	40.1	4.88	ns
Acetate (%)	69.6	67.4	70.5	1.71	ns
Propionate (mmol/l)	9.5	11.5	10.5	0.97	ns
Propionate (%)	20.6	21.3	18.4	1.44	ns
Butyrate (mmol/l)	2.8	3.7	3.6	0.36	ns
Butyrate (%)	5.6	6.5	6.4	0.43	ns
(C2+C4)/C3	3.8	3.8	4.2	0.35	ns

[a,b] Means in rows with different superscripts are significantly different ($P<0.05$).
[1] Not determined.

Discussion

Even if the MRT results are not available yet, we can expect that they will not explain the higher total tract apparent digestibility observed in endurance horses. According to previous studies, it seems that exercise leads to shorter MRT in horses (Orton *et al.*, 1995; Pagan *et al.*, 1998). Moreover, the higher total tract apparent digestibility of fibre observed in horses trained for endurance racing can not be explained by an increase in microbial concentrations, nor in their fermentative activities. An effect of the individual was observed on several parameters (*Lactobacilli* concentration, pH, percentage of propionate, total VFA, acetate and butyrate concentration and C2+C4/C3 ratio) and may have hidden the effect of conditioning. For three horses, total VFA concentration was higher after the two conditioning periods than before conditioning. This suggests an enhanced fibrolytic activity in trained horses. The use of biomolecular techniques recently developed in our lab may be helpful to give more accurate details about the impact of endurance conditioning on the digestive ecosystem of horses.

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Is there an impact of feeding concentrate before or after forage on colonic pH and redox potential in horses?

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Take home message

Feeding a concentrate meal before or after hay provision does not interfere with the redox potential, but affected pH markedly when the concentrate was ground barley.

Introduction

In order to prevent problems associated with consumption of high amount of concentrate, horse owners commonly offer forage before concentrate. However, the effects of this recommendation on digestive health are unknown. According to Brüssow *et al*. (2005), addition of alfalfa hay before or after a meal of oat did not influence the rate of intake and affected little the chewing frequency. Moreover, the order in which these two feeds were supplied may not affect pre-caecal starch digestibility as reflected by glycaemic and insulinaemic responses (Vervuert *et al*., 2008). Also, the mean retention time of dietary components in the total digestive tract did not seem to be modified by feeding concentrate before or after forage (haylage or oat straw; Hyslop, 2005).

Based on these literature data, we supposed that feeding concentrate either before or after a meadow hay does not influence the intestinal ecosystem. However, the interference with the intestinal ecosystem could depend on the type of concentrate, starchy or fibrous. For starchy concentrates, it could be more important when starch intake

(>2 g starch/kg bodyweight (BW)) exceeded the ability of the small intestine to digest this constituent (Julliand *et al.*, 2006).

The aim of the present study was to determine the effect of feeding concentrate meal before or after forage provision on two main physicochemical parameters of colonic content, pH and redox potential, when two types of concentrate are offered to horses.

Materials and methods

Four crossbred geldings (449±44 kg), fitted with a cannulae in the right ventral colon, were used in a 4 × 4 Latin square design. They received a diet consisting of a meadow hay to concentrate ratio of 72:28 on DM basis. Concentrate was either ground barley (starchy, S) or commercial pelleted concentrate (fibrous, F; Table 1). They had free access to water.

Mean daily DM intake was 1.2% BW and was divided in two equal meals of hay and two meals of concentrate of different size. The amount of ground barley offered at the morning meal provided 1.75 g starch/kg BW. The same quantity of pelleted concentrate was fed (i.e. 0.57 g starch/kg BW). The morning meal of concentrate was either offered 2 hours before or after the forage morning meal. Meals of either concentrate or forage were offered at either 08:30 or 10:30 hours and at either 16:30 or 18:30 hours. Each 15 days period consisted of a 13 days adaptation and a 2 days recording phase.

Colonic content was collected by gravity through the cannula just before the first meal in the morning (concentrate or forage) and every

Table 1. Chemical composition (% DM) of the hay, commercial pelleted concentrate (F) and barley (S).

Item	Hay	F	S
OM	94.3	91.2	97.7
NDF	67.2	32.5	16.8
ADF	37.8	16.3	5.1
Starch	-	19.3	60
Non starch polysaccharide	19.2	41.1	67.2

hour for 10 hours. Immediately after collection, pH and redox potential were recorded, respectively, with a pH electrode and a platinum electrode on colonic content maintained under strict anaerobic conditions in a water bath at 37.5 °C.

A repeated-measures ANOVA was performed to compare differences in redox potential and pH values within the 10 hours post-feeding (SAS Inst Inc, Cary, NC). The model used included the effect of horse, period, type of concentrate, order of feeding and the feeding practice (interaction between the two latest factors). Least square means were calculated for each variable and separated using pairwise t-tests (PDIFF option of SAS). Statistical significance was set at $P<0.05$.

Results and discussion

This study reported data concerning redox potential and pH post-prandial changes in the colonic content within 10 hours post-feeding. Just before the morning meal, the redox potential averaged -228 (±50.4) mV and did not vary with feeding practice. During the 10 hours post-feeding, mean potential redox ranged from -335 to -213 mV (Figure 1). Feeding concentrate either before or after hay did not affect the mean redox potential whereas the type of concentrate had a significant effect: S-diets led to a lower mean potential redox than F-diets, -288 (±38.4)

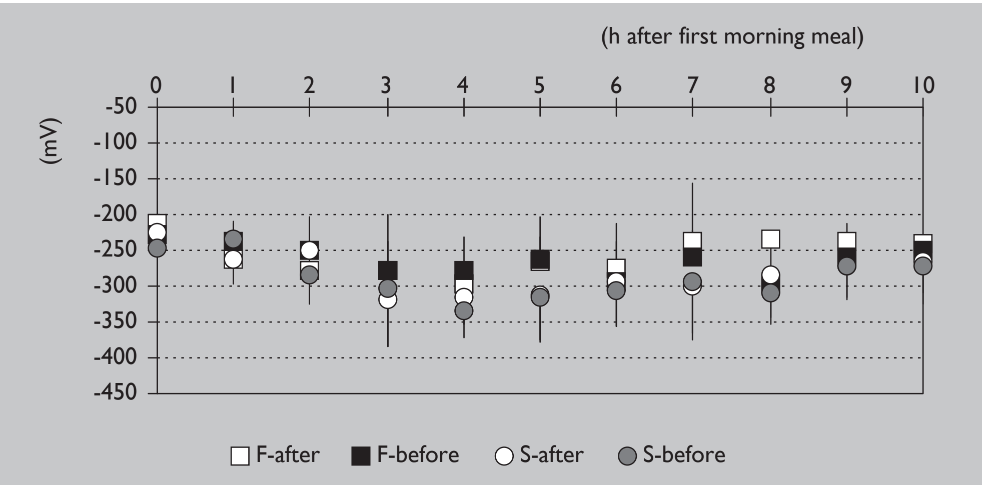

Figure 1. Mean post-prandial changes in colonic redox potential.

mV and -260 (±21.3) mV, respectively. The mean potential redox varied also with time collection, but there was not found an interaction with feeding order and type of concentrate. It slowly decreased for 4 h post-feeding and then increased until the end of the measurement period. In literature there is few data reported concerning redox potential in the hindgut of horses. In ruminal content, large variations in potential redox are reported. It varies between -260 to -145 mV (Marden *et al.*, 2004). And, it increased within the 3 and 6 hours post-feeding, respectively (Marden *et al.*, 2005; Andrade *et al.*, 2002). In horses, we observed an increase in the redox potential occurring later than in ruminants. This difference could be related to the delay of arrival of non digestible organic matter in the hindgut, 4 or 5 h after the meal.

Before the morning meal, the pH in the colonic content averaged 7.2 (±0.3). The mean pH measured within 10 hours after the morning meal ranged from 5.8 to 7.3 (Figure 2). It decreased after the morning meal and this variation depended on the feeding practice (interaction between the distribution order and the type of concentrate). The magnitude of the pH drop was the greatest with the 'S-before' diet. For this feeding practice, the pH was the lowest at 6 and 7 h after the morning meal (P<0.05). Our minimal data was lower than minimal data reported in the colonic content of horses receiving a high starch

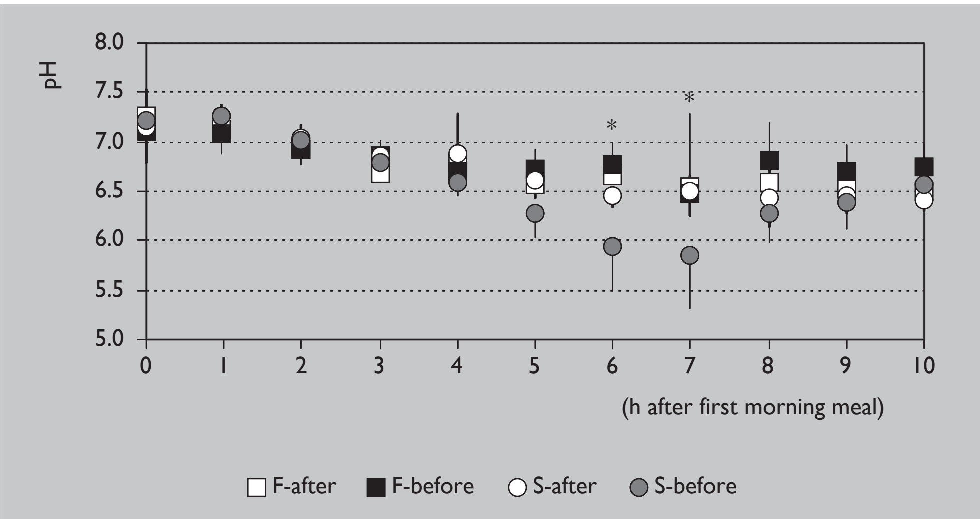

Figure 2. Mean post-prandial changes in colonic pH.

diet (6.4) (Medina *et al.*, 2002). In our trial, the colonic pH was below 6.0 for 1.3 hours. In ruminal content, the cellulolysis was diminished below this threshold value (Archimede *et al.*, 1997). However, we could question if, in our study, the time during which pH remained below 6 is enough to limit the cellulolysis. In contrast to ruminal content (Andrade *et al.*, 2002), there was no clear negative relationship between redox potential and pH. Surprisingly, the 'S-before' diet led to a decrease in both parameters, potential redox and pH.

In conclusion, this study provides new data concerning post-feeding changes of potential redox in colonic content of horses. Feeding a concentrate meal before or after hay provision did not interfere with the redox potential, but the pH was markedly affected when the concentrate fed was ground barley. More investigations should be done to determine if offering pelleted starch instead of native starch could lead to the same post-prandial changes in pH. Further studies should be carried out to examine if the variation in pH noticed in this trial could cause microbial dysfunction in the hindgut.

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In vitro and *in situ* cecal digestion of legumes in horses

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Take home message

Lucerne hay presents better *in vitro* digestibility, and *in situ* cecal dry matter degradation values than the tropical legumes stylo hay Campo Grande and stylo hay Mineirão. Nevertheless, the tropical legumes present potential to be used in horse diets.

Introduction

There are few studies estimating digestibility values of tropical roughages in horses. Feedstuffs chemical composition and its nutrients degradation in the hindgut of horses generate information that allows understanding digestion dynamics, besides characterizing feedstuffs quality. This study aimed to evaluate *in vitro* digestion and *in situ* cecal digestion of legume hays in horses.

Material and methods

The study was carried out at Equine Health Laboratory at Universidade Federal Rural do Rio de Janeiro. Treatments were: lucerne hay *(Medicago sativa)*, stylo hay Campo Grande[1], which is a seed mix with 20% *Stylosanthes macrocephala* and 80% of *Stylosanthes capitata*, and stylo hay Mineirão *(Stylosanthes guianensis cv. Mineirão)*. The stylo hays were harvested from three different areas in the field and taken as replications. Lucerne hay was bought in a local commerce

[1] Commercial name.

and samples were picked from three different bales. A randomized block design with three treatments and three replicates were used to evaluate the *in vitro* digestion of hays. All samples were milled through 2 mm mesh. Dry matter and crude protein mean values of lucerne, stylo hay Campo Grande and stylo hay Mineirão are shown in Table 1. The *in vitro* procedures consisted in two steps adapted from Abdouli and Ben Attia (2007).

A pepsin digestive solution was prepared and consisted of 1.25 g of pepsin[2]/l in 0.1 M HCl solution with pH adjusted to 2.0±0.05, and maintained at 39 °C. A pancreatin digestive solution was prepared and consisted of 10 g pancreatin[3]/l in 0.1 M phosphate buffer solution with pH adjusted to 6.8±0.02, and maintained at 39 °C. Step 1: each 0.5 g of dry matter (DM) of samples was added to 40 ml of pepsin solution and given into Erlenmeyers. Samples were maintained in a water bath for two hours at 39 °C, and shaken every hour. After acid digestion, pH was evaluated (pH_1) to know solution stability. Step 2: pepsin digested samples were added to 10 ml of 0.1 M phosphate buffer solution/0.5 g DM sample. The pH was adjusted to 6.8, and after that, 2 ml of pancreatin solution/0.5g DM sample was given into Erlenmeyers. The digestion took place during 4 hours and the Erlenmeyers were shaken every hour. At the end of the *in vitro* digestion the pH was evaluated again (pH_2). Then, the substrate was filtered through nylon cloth (45 µ)[4]; washed with distilled water, ethanol (95%) and acetone. Residues were dried during 72 h at 55 °C in a ventilated oven to estimate

Table 1. Dry matter and crude protein means values of three samples of lucerne, stylo hay Campo Grande and stylo hay Mineirão.

	Lucerne hay			Stylo hay Campo grande			Stylo hay Mineirão		
	1	2	3	1	2	3	1	2	3
DM %	88.6	89.3	88.1	88.9	89.7	91.1	88.9	89.8	89.9
CP %	17.9	17.3	20.6	12.2	11.8	12.7	10.8	11.3	11.2

[2] 1:10000, Vetec® Pure Chemistry Ltda, Rio de Janeiro, Brazil.
[3] Vetec® Pure Chemistry Ltda, Rio de Janeiro, Brazil.
[4] Tenyl®, São Paulo, Brazil.

the coefficients of digestibility. In the *in situ* assay, a factorial design 3 × 7 was used, with three treatments and three replicates and seven durations of incubation. A *caecum* fistulated 230 kg mare was used as incubation site, strategy adapted from Tomich and Sampaio (2004). The diet fed to the mare was composed by concentrate and *coastcross* hay (20:80, w/w). The mare was fed at 7:00; 12; 17 and 22 hours. The bags measured 6.5 × 20 cm with 45 μ/pore (Hyslop *et al.*, 1999). Each bag was filled with 5 g of DM sample of *in vitro* digested feedstuff, sealed, and suspended by nylon thread in the *caecum*. Incubation was done at 2; 4; 6; 8; 12; 24; 48 hours, as described by Hyslop *et al.* (1999). The study lasted 29 days, 14 days for *in vitro* procedures and 15 days for *in situ* digestion. The bags were taken from *caecum*, and frozen at -20 °C until the end of incubation procedures, when all were defrosted at the same time. Thereafter, bags were gently hand washed, until water ran clear. The soluble content at zero time were obtained by washing three bags of each feedstuff with the same procedure. Finally, the bags were dried for 72 h at 55 °C in a ventilated oven. The dry matter and crude protein analyses were made at the Animal Nutrition Laboratory of Universidade Federal de Minas Gerais. Values of pH_1, pH_2, DM and CP digestibility were submitted to ANOVA and means compared by Student-Newman-Keuls (SNK) test at 5%. *In situ* degradation parameters of DM and CP were estimated according to Orskov and McDonald (1979) model: $d = a + b(1-e^{-c*t})$; d = degradation at time 't'; a = soluble fraction; b = insoluble fraction, but potentially degradable; c = degradation rate of '*b*' fraction; t = incubation time. Statistical analyses were carried out by Statistical and Genetics Analyses System – SAEG. This project was registered in the Institutional Animal Care Committee of UFMG (no.124/2008).

Results

Means values of acid (pH_1) and basic (pH_2) steps of *in vitro* digestion and means of dry matter (DM) and crude protein (CP) of *in vitro* digestion of hays are shown in Table 2.

A pH difference was observed between hays. Lucerne hay presented higher pH in both stages of *in vitro* digestion (P<0.05). Stylo hays did not present differences between them. Lucerne DM and CP were more digestible than those of stylo hays (P<0.05). Estimates of *caecum* degradation parameters of DM and CP are shown in Table 3. The DM

Table 2. Mean values of acid (pH_1) and basic (pH_2) steps of in vitro *digestion and mean of dry matter (DM) and crude protein (CP) of* in vitro *digestion of hays.*

Hays	**pH**		***In vitro* digestion**	
	1	**2**	**DM %**	**CP %**
Lucerne *(Medicago sativa)*	3.52[a]	7.02[a]	38.45[a]	39.79[a]
Stylo Campo Grande *(Stylo capitata+Stylo macrocephala)*	2.87[b]	6.97[b]	27.09[b]	36.36[b]
Stylo Mineirão *(Stylosanthes guianensis)*	2.81[b]	6.96[b]	26.09[b]	34.54[b]
CV %	6.6	1.5	11.7	14.2

Means followed by the same letter in column do not differ by SNK test (5%).

Table 3. Estimatives of in situ *cecal degradation parameters of DM and CP of hays.*

Nutrient	**Parameters**	**Lucerne hay**	**Stylo hay Campo Grande**	**Stylo hay Mineirão**
DM	a (%)	3.1[NS]	2.64 [NS]	4.5 [NS]
	b (%)	53.1***	45.67***	43.22***
	c (%/h)	15***	14.2***	13.3***
	R^2	93.5	95.8	93.8
CP	a (%)	7.9 *	10.9**	0.0 [NS]
	b (%)	73.9***	69.9***	78.4***
	c (%/h)	13.2***	18.6***	13.3***
	R^2	92.9	94.0	84.0

a=soluble fraction; b=insoluble fraction, but potentially degradable; c=degradation rate of '*b*' fraction
Significance of models parameters: *** (P<0.001); ** (P<0.01); * (P<0.05); [NS] (P>0.05).

degradability parameters of lucerne were higher than those of the other hays, with potentially degradable fraction *b* values of 53.1%, and degradation rate *c* of 15%/h. However, the potentially degradable fraction of CP from Mineirão hay was higher, while degradation rate of CP was higher in stylo hay Campo Grande.

Discussion

The pH evaluation at the end of acid and basic steps of *in vitro* digestion aimed to observe if the solution maintained the pH for enzymatic action. The lucerne hay presented higher pH in both stages of digestion. However, the higher pH did not inhibit enzymatic action and CP of lucerne hay was more digestible than those of the others hays, of 39.8%. The amount of protein digested from stylo hay Campo Grande and stylo hay Mineirão were 36.4 and 34.5%, respectively. Moore-Colyer *et al*. (2002), inserted nylon bags into horse's stomach, and then removed the bags through *caecum* fistula. The chemical composition of hay cubes was 93% of DM, 8.3% of CP, 62% of neutral detergent fiber. The authors observed a DM prececal digestion of hay cubes of 32.0% similar to the *in vitro* DM digestion of hays used in the present study. Silva (2007) evaluated lucerne and stylo hay Mineirão in a cecum fistulated horse and DM and CP *a* parameter of lucerne were 23.6 and 33% respectively. These results were higher than those observed in the present study. Regarding stylo hay Mineirão, Silva (2007) observed larger values for *a* fraction of DM and CP, 30.3 and 32.4%, respectively. The differences can be explained by the previously removed soluble fraction *a* by *in vitro* digestion. The values observed for *b* fraction of DM and CP for stylo hay in Silva (2007) were 46.5 and 62.7%, respectively. Those values are similar to the results observed in the present study, indicating that the potential fraction was maintained even after *in vitro* procedures. The degradation rate (*c*) of stylo hay Mineirão observed in the present study was smaller than that of the same hay observed by Silva (2007), with degradation rate (*c*) values for DM and CP of 14.8 and 19.1%/h, respectively. Although lucerne is more digestible than stylo hays, important aspects of Stylosantes sp. legumes are that these species grow on acid soils with low fertility, and need low precipitation index to keep growing (Skerman *et al*., 1991) and therefore must be considered for horse production in tropical regions. Finally, the results should be considered carefully because particle loss may occur using the bag technique through nylon cloth during incubation and filtration procedures.

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NMR-based metabonomic analysis of serum from Standardbred yearlings with or without hock osteochondrosis dissecans lesions: metabolic markers for OCD

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Take home message

NMR based metabonomic analyses were able to detect significant differences in amino acid, lipoprotein, lipid and perhaps glucose profiles of yearling horses that developed OCD lesions versus half or full siblings that did not develop lesions, despite having had the same nutritional and environmental management since birth.

Introduction

Metabonomic investigations use multivariate statistical analyses of Nuclear Magnetic Resonance (NMR) spectra from a statistically relevant pool of biological samples to find metabolic markers associated with diseases. The detection of significant differences in large numbers of metabolites from diseased and normal individuals can lead to the development of novel diagnostic and therapeutic modalities based on deeper understanding of the metabolic processes involved (Hodavance *et al.*, 2007; Lindon and Nicholson, 2008; Nicholson *et al.*, 2002; Pelczer, 2003). Osteochondrosis dissecans (OCD) is a developmental orthopedic disease that has been well documented to have a genetic basis in Standardbreds and other breeds but the actual metabolic defects causing the lesions have not been well defined (Lillich *et al.*, 1997). The presence of OCD lesions has been correlated with hyperinsulinemia in Standardbred weanlings given a standardized glucose challenge (Ralston, 1996) and Thoroughbred yearlings fed high starch/sugar grain rations (Pagan *et al*., 2001) but the presence of hyperglycemia/hyperinsulinemia was not predictive of the presence or absence of lesions (Pagan *et al*., 2001). A preliminary metabonomic study (Ralston *et al*., 2009) of serum from matched pairs of yearling Standardbreds in which one member of each pair had had OCD lesions surgically corrected (OCD, n = 20) and the other had no hock lesions (Control, n = 20) revealed distinct clustering of OCD versus Control horses. Principal component partial least squares analysis (PCA-PLS, SIMCA-P + ver 12.0, Umetrics, Sweden) of the serum NMR spectra revealed differences (P<0.05) in the serum amino acid, lipid, lipoprotein and glucose profiles of yearlings that had had hock OCD lesions surgically corrected versus those that did not have hock lesions.

Each pair had the same sire, similarly bred dams and had been raised on the same farm with the same nutritional and environmental management.

Our overall objective was to replicate the preliminary study and to determine if metabonomic analyses of NMR spectra of serum samples of yearling Standardbred horses would detect consistent differences between yearlings that had had hock OCD lesions versus closely related yearlings raised in the same environment that did not have lesions.

Materials and methods

Twenty matched pairs of Standardbred yearlings and 50 other yearlings related to those used in the preliminary study were used. Each matched pair had the same sire and similarly bred dams. The only consistent difference between the two members of each pair was that one had had surgical correction of hock OCD lesions 1 to 6 months prior to sampling (OCD, n = 20), the other had no physical or radiographic evidence of lesions (Control, n = 20). The unmatched horses were siblings to those used in the previous study. Of the 50 unmatched yearlings, 41 were Controls and 9 were OCD. All of the subjects had been maintained on pastures in Hanover, PA from birth to being brought into the sales preparation barns 2 to 3 weeks prior to sampling. On pastures all horses (pregnant and lactating mares and weanlings/yearlings) were group fed 3 to 4 kg Strategy© (Purina Mills, St. Louis, MO) per head per day. Once in the barns for sales prep (1 to 2 weeks before sampling dates) they received free choice alfalfa hay and 3 to 4 kg Strategy© divided into two feedings at 05:00 and 16:00 h. Blood samples were taken by venipuncture between 12:00 and 15:00 h on 8/14/08 (n = 46, Control = 26, OCD = 20), and 9/26/08 (n = 44; Control = 35, OCD = 9), centrifuged within 30 min of collection and immediately placed on ice. Serum and plasma were separated from the red blood cells within 6 hours of collection and stored frozen (-80 °C) pending analyses.

Plasma samples were analyzed for insulin (Mercordian Insulin ELISA Kit 10-1113-10, American Laboratory Products Company (Salem, NH) and glucose (colorimetric assay glucose-SL reagent, Diagnostic Chemicals Limited, Oxford, CN) with absorbance being read at 340 nm.

Serum samples were subjected to 1H NMR analysis by a Varian (Palo Alto, CA, USA) Unity/Inova 600 MHz spectrometer at 25 °C controlled temperature using excitation sculpting (ES) water suppression (Ralston *et al*., 2009). The spectra were converted to the same scale of total integrated intensity for comparative purposes (normalized) and had peaks aligned to the methyl doublet of valine at 1.04 ppm using HiRes (Columbia University, NY, USA) (Hwang and Shaka, 2005) to compensate for the small differences between spectra due to technical variations. The NMR spectra were then compared using Principal Component Analysis (PCA) scores and loading plots (SIMCA-P + 12, Umetrics, Sweden), comparing horses with and without OCD on both sampling dates and between sampling dates. Spectral peaks that had variable importance scores (VIP) >2.0 were compared between groups by Students T-Test (Statistix 8, Ocala, FL). Metabolites associated with the peaks were identified using previously published data (Pelczer, 2005). Significance was set at $P<0.05$.

Results

Plasma glucose concentrations of the horses that were sampled in September were not different between the OCD (85.4±2.6 mg/dl, n = 9) and Control horses (82.4±3.9 mg/dl, n = 36, $P>0.5$). Due to an error in handling, the glucose samples from August were inadvertently lost.

The horses sampled in August with OCD (n = 21) had a mean plasma insulin concentration of 13.0 ± 1.2 μIU/dl while the control horses (n = 26) had a mean of 14.0±1.1 μIU/dl, (P>0.5). In September insulin concentrations also did not differ between OCD and control horses (OCD: 11.1±0.95 μIU/dl, n = 9; Control: 17.0±1.6 μIU/dl, n = 36, P>0.05). The insulin concentrations did not differ between the horses when grouped by date either (August: 13.71±0.7 μIU/dl and September: 15.6±1.4 μIU/dl, P>0.2).

There was distinct and highly significant clustering of OCD vs. Control serum spectra, with 3 principal components explaining 26% of the total sums of squares variation when the data sets from only the matched pairs from both sampling dates were combined. Even tighter clusters were seen when just the pairs from the individual sampling dates were used, with 34 and 50% of the total variation explained by 3 principal components in August and September, respectively. As seen in the preliminary trial, metabolites which differed (P<0.05) consistently between OCD and Control horses included alpha- and beta-glucose, phenylalanine, lysine, arginine, histidine, glutamine, threonine, valine, isoleucine, proline, choline, hydroxybutyrate, cholesterol, and various lipids.

Discussion

NMR based metabonomic analyses were able to detect significant differences in amino acid, lipoprotein, lipid and perhaps glucose profiles of yearling horses that developed OCD lesions versus half or full siblings that did not develop lesions, despite having had the same nutritional and environmental management since birth. It appears that alterations in protein and perhaps fat metabolic pathways in addition to the previously identified correlations with insulin sensitivity (Pagan *et al.*, 2001; Ralston, 1996) may be involved in the development of lesions. Based on these results, a characteristic metabolic profile may be able to be established for young Standardbred horses that develop OCD due to genetically based alterations in metabolism. The profile hopefully could be used to detect foals at risk before lesions appear and perhaps identify the defective metabolic pathways in genetically predisposed foals. Once this is accomplished therapeutic modalities may be explored to prevent lesions in foals at risk. Further statistical analyses of the data, comparing the horses used in this study to those from the preliminary group and determining the specific profile that is characteristic of the affected horses, are in progress. Also studies are in progress to determine if the apparent metabolic profile observed is also present in foals before lesions appear.

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A survey of feeding practice for show jumping horses in Northern Italy: are scientific new findings being applied?

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Take home message

Comparing field practices applied with the INRA requirements, the present study underlines that there is still a distance between theoretical and practical issues to ensure better performance and health in horses.

Introduction

Diet is a key factor in the management for athletic performance, but estimating requirements still remain a matter of debate. Probably this is also due to a lack of relation between theory and practice management. To our knowledge few data are reported on how owners really feed their horses. Few studies report the common feeding and management practices for performance horses and they are generally focused on racing horses, namely Thoroughbreds and Standardbreds (Blanchard *et al.*, 2008; Harris, 1999; Martin *et al.*, 2008).

There are still many challenges facing equine nutritionists to understand the real nutritional needs of sport horses. The athletic demands we put on the horse for certain competitions are very high: the diets of these athletes need to be balanced. If these criteria are not considered properly the metabolism to support exercise is compromised and horses do not consume sufficient nutrients to support the performance (Martin *et al.*, 2008). The objective of this present paper is to analyze and describe the dietary practice applied in the field and to compare them with INRA requirement tables.

Materials and methods

Horses

54 horses, aged 5 to 15 years, were included in the study because their owners asked the authors for nutritional advice. The horses were divided into two groups with horses being classified either as an elite show jumper (ESJ) competing on fences higher than 125 cm (n=38) or a medium level show jumper (MSJ) competing on fences between 115 and 125 cm (n=16). The horses were kept in 22 different stables located in the north of Italy. All the subjects included in this study were visited in the their own place to exactly evaluate the general health and their feeding schedule. The information was collected after a discussion with the owner, the trainer and the grooms.

Body weight and conditions

Body weight (kg) was estimated using a tape and applying the formula $0.00184c^2$-129 (c=hearth girth in cm) according to Bergero (1996). The body condition score (BCS) on a 9-point scale, was determined by the same operator with visual appraisal and palpation of the six areas suggested by Henneke *et al*. (1983).

Feeding plan analysis and stable practice

All the diets given to the horses were accurately analyzed to study meal composition. The feed used was weighed with a scale to estimate the quantities of food given to the horses. Roughage (R) was evaluated by visual inspection to assess the stage of maturity, grass families and species (grass or the possible presence of weeds), moulds or dust, color and odor. On this basis the nutritional values ware calculated according to the INRA suggestions (1990). Concentrate feeds (C) included sweet feed such as commercial coarse mixture or pellets, other feedstuffs given as simple ingredients such as beet pulp and bran, cereals and vegetable oil. All the concentrate feed was evaluated to estimate the quantities and the qualities using the manufacturer indications: dry matter content (DM), raw fat (RF), raw protein (RW), crude fiber (CF) and ash. They were also evaluated to determine the brand of the company (International or Italian) and type (fat and fiber or balanced feedstuff expressly formulated for horses or cereal-legumes mix with no vitamin mineral supplementation). The analysis of nutritional content of the diet was performed for each horse according to INRA using a commercial software (Bergero *et al.*, 1996). The workload was determined on the owner perception basis (light<40 min/day, intense 60 min/day) and the number of practices performed in the week. A direct determination of the duration of the effort in the single practice and the number of practices in the week was also performed, when possible.

Statistics

All the data obtained from the feeding plan analysis were investigated using SPSS 11.1® software. In particular the differences from INRA maintenance levels for energy and protein were calculated. For both groups the mean daily energy (UFC ± Standard Deviation) and protein (MADC ± SD) intake and mean dry matter intake (kg/day) were also calculated. In both groups the Roughage/Concentrate ratio (R:C), mean roughage intake (kg/day), mean concentrate intake (kg/day), the % of hay quality, the % brand and type of C were calculated. The % of the problems reported by the owner and the work intensity perception of the owner were also assessed.

Results

Horses and management

The mean BW was 572±50 kg for ESJ and 579±36 kg for MSJ. 66% of the ESJ had a BCS score between ≥ 4 and ≤ 5 and 34% a BCS between > 5 and ≤ 6. 44% of MSJ had a BCS score between ≥ 4 and ≤ 5 and 56% between > 5 and ≤ 6.5.

All horses were housed in individual stalls, with 74% of ESJ and 69% of MSJ having no access to a paddock for grazing during the week. No straw was used as bedding.

66% of the owners of ESJ and 56% of the owners of MSJ perceived that the work of their horse was hard (60 min/day vs. 40 min/day of work including jumping).

The reasons that prompted the owners to ask for nutritional advice are reported in Table 1.

Feed evaluation

The mean R:C (as fed basis) was 60:40% for ESJ and 66:34% for MSJ. First cut meadow hay was the only forage used for all horses. R was found to be poor quality hay (PQH): Large presence of weeds, moulds and dust, late stage of maturity with the presence of lignification of the stems and alteration of color and odor; high quality hay (HQH): free of mould, dust, weeds and undesired materials, leafy with fine stems, soft and pliable to the touch, pleasant aroma, bright green, no sign of flowering or seed; (MQH) medium: with intermediate characteristic such as moderate content of weeds, moulds and dust, no sign of color and odor alterations, but cut after the flowering stage.

63% of MSJ were offered HQH and only 6% received PQH. For the ESJ 42% were offered HQH hay and 11% received PQH.

63% of ESJ horses were fed with a C made by a multinational company whereas 93% of MSJ horses received a C made by an Italian (mainly local) company.

Table 1. Reasons that prompted the owners to ask for nutritional advice (Chronic anemia= diminution of the number of red blood cells with no correlation with other pathologies; Nervous temperament = hyper excitability; Low BCS: decreasing of the body condition perceived by the owner; For the good of the horse: general welfare of the subject; Colic: recurrent gut troubles; General poor performance: decrease of the desired physical performance (sometimes light lameness).

	ESJ (%)	MSJ (%)
Chronic anemia	13.2	12.5
Nervous temperament	23.7	12.5
Low BCS	7.9	12.5
For the good of the horse	39.5	31.3
Colic	13.2	12.5
General poor performance	2.6	18.8

Considering the type of concentrate 11% of the ESJ horses were fed only simple cereals, whereas 100% of MSJ received a C mix claimed as expressly formulated for horses such as coarse mixture or pellets. However 56% of these products were cereal-legumes mixes without vitamins and minerals supplementation. None of the concentrates used was a fat and fiber type. Only 5% of the ESJ horses and 6% of the MSJ horses became soaked not molassed beet pulp in their diet; 19% of MSJ horses had oil added to the concentrate, but none of the ESJ horses. 6% of the horses only received wafer hay but not as hay substitute.

Diet evaluation

The estimated UFC amount above maintenance was 57% in both groups and for MADC 119% for ESJ horses and 102% for MSJ horses.

Daily UFC energy intake was 7.3±0.9 for ESJ and 7.1±0.4 for MSJ horses, and the daily protein intake was 723±153 g of MADC for ESJ and 663±74 g for MSJ. The daily dry matter intake was 10.5±1.16 kg for ESJ and 10.4±0.8 for MSJ (Table 2).

Table 2. Daily dry matter (DM), roughage (R), concentrate (C) intake and daily estimated energy (Unité Fourragère Cheval or Horse Feed Unit) and protein intake (Matières Azotées Digestibles Cheval or Horse Digestible Protein).

	ESJ	**MSJ**
Mean daily DM intake (kg/day)	10.5±1.2	10.4±0.8
Mean roughage intake (kg/day)	7.3±1.5	7.9±1.2
Mean concentrate intake (kg/day)	5±1.5	4.1±0.9
R:C ratio	1.7	1.9
Mean daily energy intake UFC[1]	7.3±0.9	7.1±0.4
Mean protein daily intake (MADC[2])	723±153	663±74

[1] UFC: Unité Fourragère Cheval or Horse Feed Unit: Net energy (1 UFC=1700 kcal net energy).
[2] MADC: Matières Azotées Digestibles Cheval or Horse Digestible Protein: g/day.

Discussion and conclusions

Details of the management, feeding practice and health care of horses are essential to resolve possible troubles. Diet evaluation is an essential part of this. From this report, it appears that the management of sport horses in Italy is still tied to tradition. The feeding management had only the purpose of achieving a good body condition score, without considering other important factors for the welfare of the horse like providing adequate fiber for good hindgut function and avoiding carbohydrate overload or protein excess (Martin *et al.*, 2008). Nutrition is an essential part of the daily management of the horse and it affects its welfare and performance (Ellis and Hill, 2005).

Little attention was paid to the nutrition of the sport horses, even if they were top athletes. They received a diet based only on meadow hay and simple cereals according to Italian tradition; horse owners often think that grandfathers' way of feeding the horses is still good because they do not really feel the feeding impact on performance. It is surprising that this kind of diet is still present in ESJ, where the horses are required to compete always on top level. These horses are believed to be more like 'production animals', so the feeling of owners is mainly based on results. Possibly for this reason the owners are centered on technical problem solving, considered pivotal, and pay less attention to the food selection using poor or only medium quality hay and simple cereal diet. No owner used for example additional vegetal oil. In this report 58% of the horses were not feed with HQH and 11% were fed with simple cereals like oat or barley. 74% of the horses had no access to a grazing paddock or dry-lot. They were kept in the stable for almost 23 hours of a day because the paddock is considered a dangerous environment. This picture shows that the horses may not be very comfortable, especially when we consider a concentrate based diet with a R:C 60:40 and a mean concentrate intake of 5 kg/day.

The tendency to have a low BCS score on these subjects is probably linked to the difficulties of keeping the horses in a comfortable environment besides the higher demands due to the sportive activity. This kind of management can influence also the temperament of the subject. 23% of the requests for checking the nutritional management of the horses were due to the nervous temperament of the horses and 13% due to recurrent colic.

Another interesting point was that 63% of the horses received a concentrate feed made from a multinational company. This is probably related to the great possibility of the owner to find the same product while participating in international competitions and the general tendency for individuals to prefer foreign products. None of these products were a fat and fiber type (Williams *et al.*, 2001), but all of them were sweet feed specially formulated for horses.

The body condition scores were in an adequate range for the physical activity of the horses in both groups. However there was a general tendency to have a higher BCS in the MSJ group (56% >5 ≤ 6.5 MSJ and no horses over 6 for ESJ). This is related to an overestimation of the energy requirement and to a certain attention in choosing feeds and management. 13% of the MSJ owners asked for a revision of the nutritional plan because they felt that their horses had a too low BCS. In horses, as in cats and dogs (Kienzle and Berglery, 2006) closer relationship and over-humanization can influence the feeding management supplying more than necessary. Even if the MSJ horses received a concentrate claimed for sport horses, 56% of these products were cereal-legumes based products, without vitamins and minerals added. The lack of adequate supplementation could explain the incidence of chronic anemia in both groups. In particular the use of non supplemented mixtures for MSJ horses or the low quality forage for ESJ horses could influence the availability of vitamins and minerals essential for the production of red blood cells (Lewis, 1994).

66% of the owners of ESJ horses and 56% of MSJ horses felt that the work of their horse was hard (60 min/day 6 days a week vs. 40 min/day 6 days a week of flat work with jumping exercise) but there was little possibility to really assess the quantity and quality of the exercise.

This could explain the higher BCS of the MSJ horses and the overestimation of the general energy requirements of this group of horses.

For both groups the UFC intake was 57% above the maintenance requirement. According to INRA tables this is sufficient for light work. From this analysis the ESJ horses are really involved in a light work and the selection of medium quality forages and the hard management forced the owner to use large quantities of concentrate (5±1.5 kg/day) with a mean protein intake of 119% above maintenance (723±153 g MADC). These quantities of concentrates and protein can be related to many pathological conditions such as colic (Durham, 2007) and behavioral problems (Ellis and Hill, 2004). Also the low percentage of DM ingested (1.8% of BW in both groups) can contribute to the increase of behavioral problems and gut troubles. This value is very close to the mean DM intake estimated in show jumping horses in France, by Martin *et al*. (2008).

From this report it is clear that feeding practice in both ESJ and MSJ horses are still related to tradition and little attention is paid to the selection of concentrate to balance protein and mineral/vitamin needs. Generally the owner selects the C based on label claim and with no attention to the real composition of the feedstuffs. Moreover the selection of national products could be also explained by lowering the cost of feeding horses.

From this report, the general picture is as follows: horse nutrition is still based on unchallenged dogmas that have been passed through many generations of horsemen. The biggest obstacle for an improvement is probably the attitude of on average very conservative equine community (Van Weeren, 2008).

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A method of measuring the influence of rider interaction and training on the temporal relationships between limb cycles of horses using inertial motion sensors

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Take home message

The affects of rider interaction and training are potentially measurable using this system to provide information with regards to progress, whereby providing a way of monitoring aspects such as how a horse is adapting to training or during recovery from an orthopedic problem.

Introduction

The detailed study of locomotion patterns is essential for assessing locomotor performance in the sports horse. It is of particular importance in evaluating training responses (Gaughan, 1996; Miyata *et al*., 1999; Parsons *et al*., 2008), in exploring subtle variations and fluctuations in gait, in diagnosing lameness (Cheney *et al*., 1973), in assessing horse rider interactions (Lagarde *et al*., 2005; Schollhorn *et al*., 2006), and investigating the effects of different training and competition surfaces (Burn and Usmar, 2007; Cheney *et al*., 1973; Gustas *et al*., 2007; Thomason and Peterson, 2008). All of which are critical factors that ultimately determine the locomotor performance of the horse. Hence there is a need to take locomotion analysis into the working/competition environment of the sports horse. The advancement of inertial based sensor technologies including accelerometers and gyroscopes present us with an opportunity to further investigate locomotion patterns within this working (field) environment.

Inertial motion sensor (IMS) studies on the temporal relationships between horses' limbs have been reported in literature (Parsons *et al.*, 2008; Robilliard *et al.*, 2007). Defining these temporal relationships has traditionally been based on the measurement of specific events within the limb cycle, for example initial ground contact of the respective limbs, with the gait being defined by the relative time lag between the limbs, expressed by percentage of the stride duration relative to a given reference limb.

This characterization is based on an underlying assumption that the reference event (for example initial ground contact) which is used to determine these temporal lag relationships between the limbs occurs at the same stage within the respective limb cycles, and this assumption has yet to be demonstrated. However these techniques offer little information regarding the defining characteristics of the complete locomotor cycle, and the temporal inter-relationships between the respective limbs.

Furthermore previous work has demonstrated that the rider can influence the horses gait (Lagarde *et al.*, 2005; Peham *et al.*, 2004; Schollhorn *et al.*, 2006) with a skilled rider having a stabilizing effect. There is little known about whether the rider can alter the cyclical relationship between the limbs and the respective events occurring, particularly between the fore and hind limbs. Indeed anatomical differences between the respective fore and hind limb support an assertion that the respective limb cycles are likely to be different.

It is therefore our aim to use a measurement technique which defines the entire limb cycle to determine the temporal inter-relationships between the limbs for each gait of ridden horses.

Material and methods

Trials were carried out with three horses training at Advanced British Dressage level or above. One rider, experienced at training up to Grand Prix level, was used with all horses to reduce variability. All trials were completed on the same day on sand and rubber surfaced 20 × 60 m arena.

In order to measure the rotations of each limb the horses were equipped with an IMS, containing a tri-axial 5 g accelerometer and three single axis 1,200 deg/s gyroscopes, housed in adapted brushing boots. The rider wore a global positioning satellite (GPS) device fitted onto their riding hat to enable calculation of speed and stride length during working trials. The GPS device and the IMS were synchronized and time and date stamped by a computer previous to recording. To represent a real training situation each horse was warmed up (WU) in walk, trot and canter to the rider's discretion. This was followed by approximately 20 minutes of work (WK), consisting of collecting, working and extending gaits. The training sessions were filmed using a miniDV camcorder. The camcorder and the IMS designated as the reference limb were switched on simultaneously to enable timed identification of transitions within and between gaits.

Data from the z-axis gyroscope, recording the rotations of the metacarpal/tarsal (MC/MT) region perpendicular to the direction of forward motion of the horse, was used for analysis. A cross correlation approach was used to calculate the temporal phase-lag (TPL) between the cyclical wave forms of each limb with regards to a reference limb. The results were presented as a percentage of the stride duration of a reference limb.

Results and discussion

Temporal stride data collected during symmetrical gaits from IMS attached to the MC/MT region of the horse differs from data collected at the level of the hoof when comparing ipsilateral paired limbs. The fore limb (FL) rotation occurs at a different rate than the hind limb (HL), particularly in the preparation for hoof landing. This is not surprising given the anatomical and biomechanical differences between the FL and HL. However the 50% phase lag measured between contralateral paired limbs in symmetrical gaits was in agreement with previous studies (Abourachid, 2003).

The median TPL between limbs in walk, trot and canter is shown in Figure 1. All horses showed changes in TPL between limb cycles from

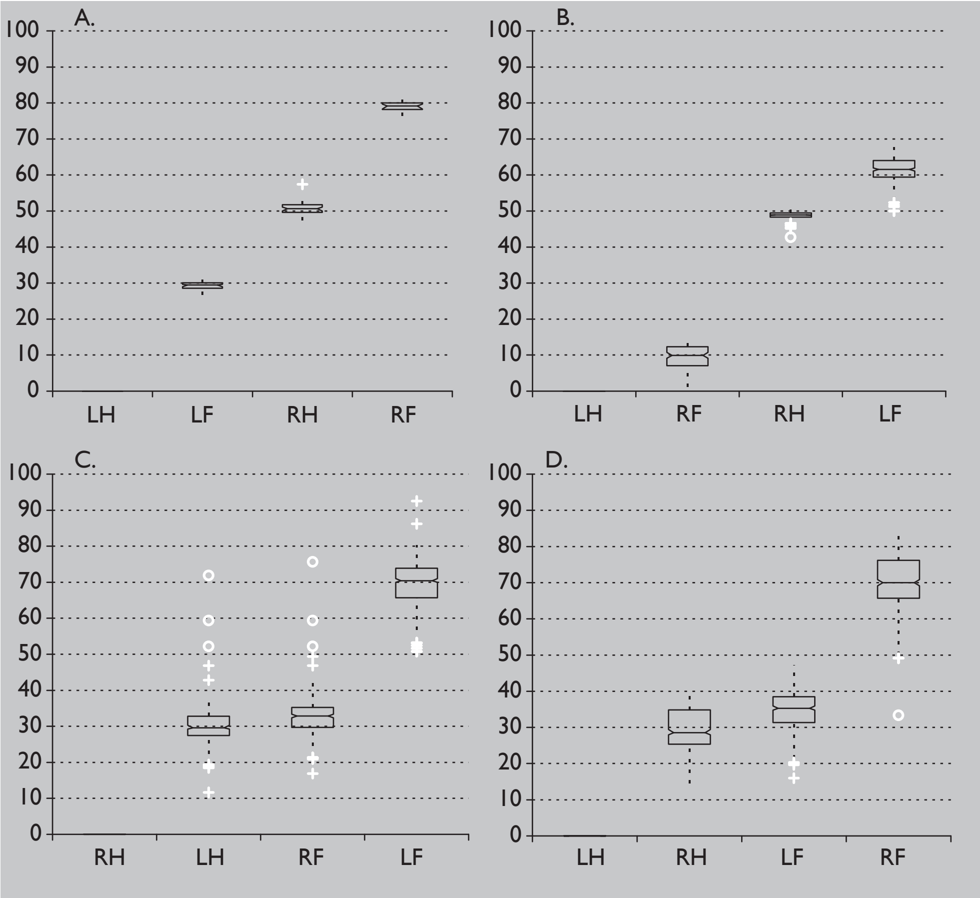

Figure 1. The median and interquartile range (IOR) temporal phase-lag of the limb cycle all horses in A. walk, B. trot, C. canter left and D, canter right for the left fore (LF), right hind (RH) and right fore (RF), as a percent of the left hind (LH) stride cycle. The mid line of the boxes show the median value, the outer lines shows the confidence interval of interquartile range, '+' shows observations between 1.5 and 3.0 IQRs away from the quartiles, 'o' show far outlier observations over 3.0 IQRs away from quartiles.

WU to WK that were individual; however comparing the full WK trial gait to the WU gait is difficult as there is more variation in the former. The contralaterally paired limbs demonstrated a close coupling in TPL during the symmetrical gaits regardless of work type. This was not surprising as it is argued that characteristic time-lag reflects the underlying action of the central pattern generator (CPG) (Grillner *et* *al*., 2000; Williams *et al*., 1989) and that the contralateral pairs are activated as an antagonistic pair (Griffin *et al*., 2004).

The TPL between ipsilateral/diagonal pairs in the symmetrical gaits reduced from WU to WK trials, most obviously for H1 and H2 (Figure 2). During WU a horse is asked to progressively work with more engagement from the HLs and 'lighten' its forehand (FEI) this may have the effect of causing the rotation of the FL in preparation for hoof landing to occur later in its cycle and become more closely in-time with the HL rotation.

Intra horse asymmetries in TPL between ipsilateral and diagonal pairs in trot were recorded for H1 and H2 during WU that reduced for H1 during WK while H2 became almost symmetrical. H3 demonstrated symmetry in left and right diagonal pairs for both WU and WK. This may be an indication of a stronger side in the individual horse and may demonstrate the usefulness of monitoring symmetry of gait as an early indication of compensatory movement (Keegan, 2007).

During the working element of the trials all horses altered the TPL particularly between the ipsilateral or diagonal FL and HL in a pattern that appeared to be dependant on stride length rather than stride duration or speed. The FLs had a greater range of TPL variation than the HLs in symmetrical gaits and the amount of variation appeared to increase with training level.

Conclusions

This method provides continuous information regarding the rhythm and regularity of the limb cycles of horses in a training environment enabling easy comparison for both inter and intra horse variations of gait and the effect of different surfaces. The system has the potential to be used for lameness assessment, training or locomotory rehabilitation.

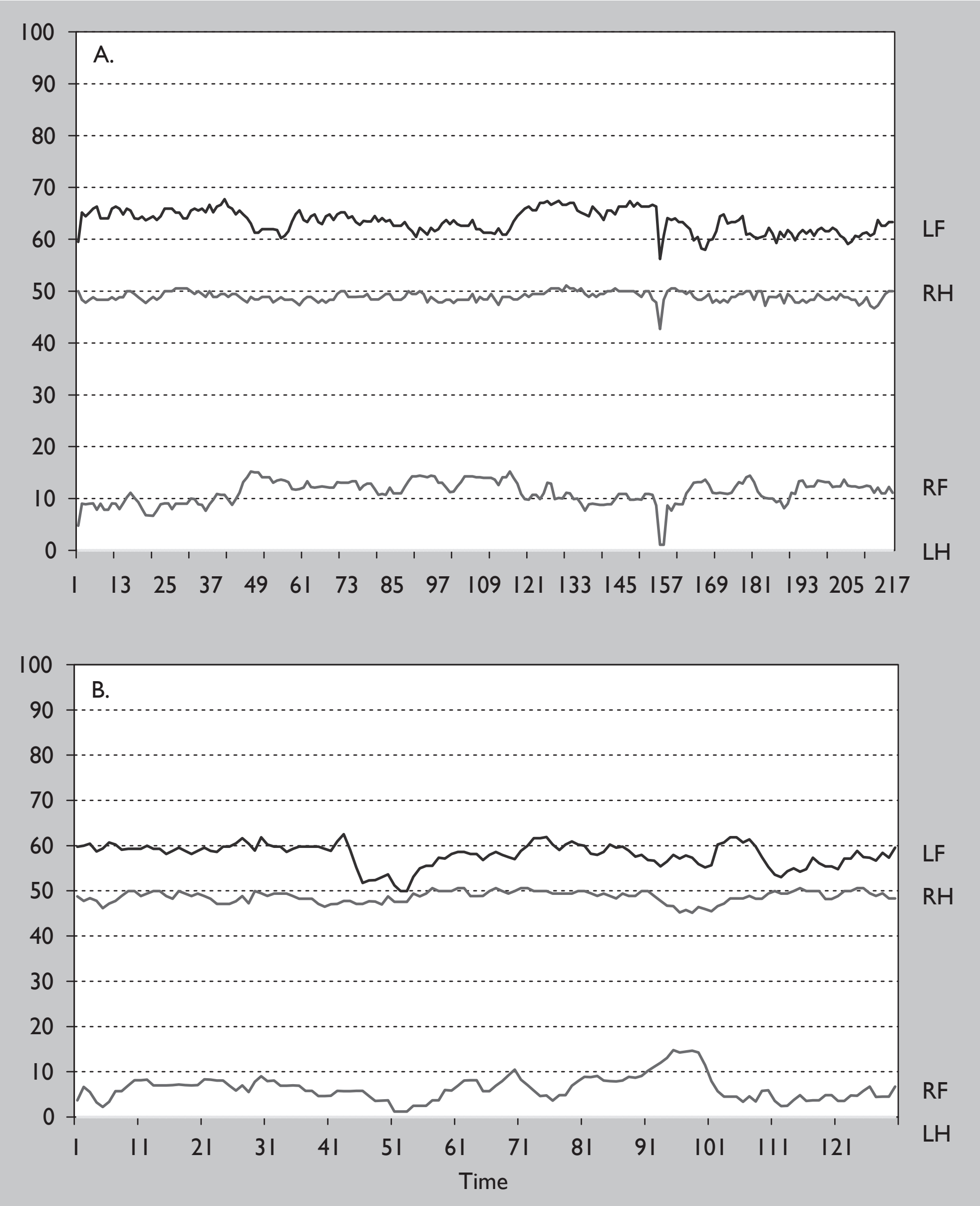

Figure 2. Temporal limb phase lag of the right fore (RF), right hind (RH) and left fore (LF) in relation to the left hind (LH) for horse 2 showing the difference in trot between A. warm up and B. working. Notice the reduction lag between the fore limb and diagonally paired hind limb.

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