



Sveriges lantbruksuniversitet
Swedish University of Agricultural Sciences

Faculty of Veterinary medicine and Animal science
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Nutrition-related risk factors for colic in horses

Nutritionrelaterade riskfaktorer för kolik hos häst

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Table of contents

1. Abstract	1
2. Sammanfattning	2
3. Introduction	3
4. The impact on gastrointestinal tract of various feed components.....	4
4.1. Fibers.....	4
4.2. Starch	5
4.3. Water	6
4.4. Salt	6
4.5. Feeding recommendations.....	7
5. Colic	7
5. 1. The epidemiology behind colic	8
6. Nutrition-related risk factors for colic.....	8
6.1. Type and amount of feed	9
6.1.1. <i>Concentrate and grains</i>	9
6.1.2. <i>Specific type of concentrate- Oats and corn</i>	10
6.1.3. <i>Specific types of concentrate-Processed feeds and feed additives</i>	10
6.1.4. <i>Forage</i>	11
6.2. Feeding strategies for concentrates and forage.....	11
6.3. Dietary changes in concentrate and roughage.....	12
6.4. Watering practices.....	14
7. Aim and objective	15
8. Materials and methods	15
8.1. Experimental design	15
8.2. Design of questionnaire	15
8.3. Transformation of data.....	16
8.3.1 <i>Reclassification of variables and additional variables created</i>	16
8.4. Data analysis	17
9. Results	17
9.1. Descriptive statistics	17
9.1.1. <i>Colic</i>	17
9.1.2. <i>Type and amount of roughage</i>	18
9.1.3. <i>Type and amount of concentrate</i>	18
9.1.4. <i>Type and amount of feed supplements</i>	18

9.1.5. Feeding routines for roughage.....	19
9.1.6. Feeding routines for concentrates	19
9.1.7. Watering strategies.....	19
9.1.8. Change in type and amount of roughage.....	19
9.1.9. Change in type and amount of concentrate	20
9.1.10. Changed access to water	20
9.1.11. Changed type of water source.....	20
9.1.12. Number of trainings per week.....	20
9.1.13. Individual factors.....	21
9.2. Results from univariate analysis	30
9.2.1. Type and amount of roughage.....	30
9.2.2. Type and amount of concentrate	30
9.2.3. Type and amount of feed supplements.....	30
9.2.4. Feeding routines for roughage.....	31
9.2.5. Feeding routines for concentrates	31
9.2.6. Watering strategies.....	31
9.2.7. Change in type and amount of roughage.....	31
9.2.8. Change in type and amount of concentrate	31
9.2.9. Changed access to water	32
9.2.10. Changed type of water source.....	32
9.2.11. Number of trainings per week.....	32
9.2.12. Individual factors.....	32
9.3. Results from multivariate analysis.....	35
9.3.1. Change in type of water source in the paddock	35
9.3.2. Number of concentrate meals fed daily.....	35
9.3.3. Daily intake of salt	35
9.3.4. Change in type and amount of roughage.....	36
9.3.5. Change in type and amount of concentrate	36
9.3.6. Amount feed/ 100 kg BW fed daily.....	36
10. Discussion.....	38
10.1. Change in type and amount of feed	38
10.2. Amount of feed per 100 kg BW fed daily	39
10.3. Feeding routines for concentrates and roughage	40
10.4. Change in type of water source in the paddock.....	41
10.5. Daily intake of salt.....	42
10.6. Other nutritional-related management factors	42

10.7. Experimental design and execution.....	43
10.8. Future research.....	44
11. Conclusion	44
12. Acknowledgments	44
13. References.....	45
14. Appendix 1	

1. Abstract

Among several factors including both intrinsic- and management factors, several nutrition related factors has been shown to be associated with an increased risk of colic in previous studies. The aim with this study was to further investigate this association by describing feeding routines and feed rations in horses with a history of colic episodes during the last six months (colic horses) and compare these with horses without colic episodes the last six months (non-colic horses) under Swedish conditions. The study was designed as a questionnaire study which was available on the Internet for respondents from January 18, 2016 to February 29, 2016. The questionnaire contained 17 questions regarding; general horse information (age, sex and breed, BCS, weight, exercise information); type and amount of roughage, concentrate and supplements; and the feeding- and watering practices performed. In total, 3264 complete answers were received of which 523 questionnaires represented colic horses and 2742 questionnaires represented non-colic horses. Each question and sub-questions from the questionnaire were translated to a variable. Also, some new variables were created, resulting in 75 variables in total that were entered into SAS. The variables were analyzed in a univariate analysis using logistic regression in SAS.

All variables with a P value of <0.05 in the univariable analysis were considered for possible inclusion in the multivariable model. The final model for the class variables identified as having a significant association with colic were: horses having a change of water source in the paddock were more than two times likely to be colic horses ($p=0.002$) relative to horses not having a change of water source in the paddock; horses fed concentrate three times ($p=0.005$) daily were more than two times more likely to suffer from colic relative to horses not receiving any concentrate; horses having a changed type of roughage were more than two times more likely to be colic horses ($p<.0001$) relative to horses not having a change in type of roughage; horses having an increased amount of roughage were less likely to be colic horses ($p<.0001$) relative to horses not having a change in the amount of roughage; horses having a changed type of concentrate were more than three times more likely to be colic horses ($p<.0001$) relative to horses not having a change in type of concentrate; horses having an changed amount, increased or decreased, of concentrate were less likely to be colic horses ($p<.0001$) relative to horses not having a changed amount of concentrates. The results from the final model (for the continuous variables significantly associated with presence of colic) showed that for each 1 kg increase in the daily amount of müsli per 100 kg BW consumed, the presence of colic increased over three-fold ($p=0.005$); for each ml of liquid vitamins per 100 kg BW consumed daily, the presence of colic increased by 3 % ($p=0.03$) and for each 1 kg increase in the amount of roughage per 100 kg BW consumed daily, the presence of colic decreased over three-fold ($p=0.007$).

The result from this study indicates that the amounts of feeds and supplements, as well as change in feeding and watering practices are the main nutrition-related factors affecting the presence of colic in horses. In order to reduce the risk of colic from a nutritional point of view it is therefore crucial to ensure that the main source of nutrition in the diet is roughage, to try to reduce the use of concentrates as much as possible, and to carry out all kinds of changes, in both feeding and watering, successively.

2. Sammanfattning

Bland flera faktorer, inklusive både individuella och skötselfaktorer har flera visat sig vara associerade med en ökad risk för kolik i tidigare studier. Syftet med denna studie var att ytterligare undersöka denna association genom att beskriva utfodringsrutiner och fodermedel för hästar som har haft kolikepisoder de senaste sex månaderna (kolikhästar) och jämföra dessa med hästar utan kolikepisoder de senaste sex månaderna (icke-kolik hästar) under svenska förhållanden. Studien var utformad som en enkätstudie riktad till hästägare, och var tillgänglig online under perioden 18 januari 2016 till 29 februari 2016. Enkäten innehöll 17 frågor omfattande allmän information om; hästen (ålder, kön och ras, BCS, vikt, motion); typ och mängd av grovfoder, kraftfoder och fodertillskott; vattenkälla i hage och stall; samt förändringar i utfodring och vattning. Totalt inkom 3264 fullständiga svar där 523 representerade kolikhästar och 2742 enkäter representerade icke-kolik hästar. Varje fråga och delfråga från frågeformuläret transformerades sedan till olika variabler. Även nya variabler skapades vilket resulterade i totalt 75 variabler som importerades till SAS. Alla variablerna analyserades i en univariat analys med hjälp av logistisk regression.

Alla variabler som fick ett P-värde $<0,05$ i en univariat analys ingick i en efterföljande multivariat analys. Resultatet visade att; hästar vars vattenkälla i hagen förändrats hade mer än två gånger så hög förekomst av kolik ($p = 0,002$) jämfört med hästar vars vattenkälla i hagen inte förändrats; hästar som utfodrades med kraftfoder tre gånger ($p = 0,005$) dagligen hade mer än två gånger så hög förekomst av kolik än hästar som inte utfodrades med något kraftfoder; hästar vars grovfodertyp förändrats hade mer än två gånger så hög förekomst av kolik ($p < 0,0001$) jämfört med hästar vars grovfodertyp inte förändrats; hästar som fått förändrad (ökad eller minskad) mängd grovfoder hade lägre förekomst av kolik ($p < 0,001$) jämfört med hästar som hade oförändrad mängd grovfoder; hästar vars kraftfodertyp förändrats hade mer än tre gånger så hög förekomst av kolik ($p < 0,0001$) jämfört med hästar vars kraftfodertyp inte förändrats; och att hästar som fått förändrad (ökad eller minskad) mängd kraftfoder hade lägre förekomst av kolik ($p < 0,0001$) jämfört med hästar som inte fått förändrad mängd kraftfoder. Resultatet från den slutliga modellen för kontinuerliga variabler visade att för varje 1 kg ökning i mängden müsli per 100 kg kroppsvikt och dag så ökade förekomsten av kolik trefaldigt ($p = 0,005$); för varje ml ökning av mängden flytande vitaminer per 100 kg kroppsvikt och dag ökade förekomsten av kolik med 3% ($p = 0,03$), och för varje 1 kg ökning i mängden grovfoder per 100 kg kroppsvikt och dag minskade förekomsten av kolik med 20% ($p = 0,007$).

Resultaten från denna studie tyder på att mängden av olika foderslag, och förändringar i utfodring och vattning är de viktigaste utfodringsrelaterade faktorerna som påverkar förekomsten av kolik hos hästar. För att minska risken för utfodringsrelaterad kolik är det därför viktigt att i största möjliga utsträckning se till att den huvudsakliga näringskällan är grovfoder, att minska användningen av kraftfoder så långt det är möjligt, och att utföra alla ändringar i foderstaten, både för foder och för vatten, successivt.

3. Introduction

The horse has evolved, and is adapted, to a grazing and browsing existence in which it selects forages containing large amounts of e.g. water, soluble proteins and structural carbohydrates (Frape, 2010). In comparison, the dietary demands of modern equine activities has resulted in quantitative and qualitative dietary changes and feeding routines (Durham, 2009). Nowadays, horses are subjected to restricted feeding time and quantitative and qualitative diet changes at infrequent intervals. Also, they are introduced to unfamiliar materials like starchy cereals, fats, protein concentrates, and dried forages (Gonçalves *et al.*, 2002; Frape, 2010). When evaluating feeding practices among different horse populations all over the world it has been shown that cereal-based diets represent the main energy source of the modern performance horse. This increase in use of concentrates has resulted in a decreased amount of forage in the diet (Hill, 2007). Studies has shown that there is a lack of knowledge about feeding practices among horse owners in USA (Hoffman *et al.*, 2009) but also among Swedish horse owners (Henricson, 2007). Inadequate dietary management strategies, for which the equine gastrointestinal tract is not evolutionarily adapted, may lead to digestive disturbances such as colic (Hill, 2007; Durham, 2009).

Colic has been reported to be one of the most common and painful conditions in modern equine practice, and it is one of the single most common causes of death in some horse populations (Tinker *et al.*, 1997; Archer & Proudman, 2001; Durham, 2009). The incidence of colic has been shown to vary from 3.5 to 10.6 colic episodes per 100 horses per year in some studies, and in the same studies Overall mortality rate for colic has been reported to be 0.45-0.7 deaths/100 horses per year in USA and UK (Kaneene *et al.*, 1997; Tinker *et al.*, 1997; Hillyer *et al.*, 2002; Traub-Dargatz *et al.*, 2001). The frequency of colic in Sweden is high; 22 percent of 3100 horses were reported to be suffering from colic in 2008. Last year 428 horses were admitted to the equine clinic at the University animal hospital in Sweden (UDS), which is one of the largest equine hospitals in Sweden, for treatment of colic (personal message, Cecilia Eriksson, UDS). Besides having a negative effect on the health and welfare of the horse, colic also have a negative impact on the overall economy of the horse industry (Traub-Dargatz *et al.*, 2001), as well as for the individual horse owners, as an median gross cost for veterinary treatment of colic, in Sweden, has been reported to be 4 729 SEK/ horse/ treatment (Egenvall *et al.*, 2008).

Previous studies have illustrated colic to be a condition that is complex and multifactorial by nature, as it is caused by both nutritional- and non-nutritional factors including management and individual factors of the horse. However, nutrition-related factors (including amount, type and change in diets as well as feeding strategies and watering practices) have been found and concluded to be main causes of colic in several studies. Therefore, it is important to further investigate associations between colic and nutritional factors in different horse populations.

The aim with this study was to investigate associations between presence of colic and nutrition- and management factors by describing feeding routines and feed rations in horses that have had colic episodes the last 6 months (colic horses) and compare these with the same routines for horses without any colic episodes the last 6 months (non-colic horses) under Swedish conditions.

4. The impact on gastrointestinal tract of various feed components

The type and amount of feed and water, as well as the combination of feeds included in the diet have different impacts on the gastrointestinal tract (GIT) of the horse. The variables affected in the hindgut of the horse are primarily pH, bacterial profile and the amount and ratio of volatile fatty acids (VFA's) produced (Shirazi-Beechey, 2008). When talking about horse feed rations from a nutritional point of view, in terms of health and welfare, the ratio and amount of roughage and concentrate included in the diet is often discussed. When analyzing feeds and investigating how these feeds effect the GIT, the components fiber and starch respectively, are more commonly used (Frape, 2010).

4.1. Fibers

On a forage based diet the horse receives energy mainly from the structural carbohydrates (fibers). The structural carbohydrates are primarily digested in the cecum and colon. Insufficient amounts of fiber in the diet can result in digestive disturbances such as colic, as fiber also prevents the emergence of harmful bacteria as the environment in the GIT is to their disadvantage. Fibers are degraded more slowly than non-structural carbohydrates and provide a greater intestinal filling, and are therefore more suitable for the horse as horses are adapted for fiber rich diets (Frape, 2010). One factor that may affect the GIT health is the transit time of the digesta, which is the time the feed resides in the GIT. The transit time varies with the type of feed consumed and has been shown to decrease when the horse digest rough, energy-poor feed. A shorter transit time means that microorganisms have shorter time to breakdown and become less efficient, which the horse compensates by eating more (Pearson *et al.*, 2001).

The diet affects the composition of the micro flora in the GIT, and also the type of VFA's produced in the hindgut. Horses having a diet high in forage or that are fed only forage has a higher proportion of acetic acid in the hindgut, while propionic acid is present in smaller proportions. As acetic acid is a weaker acid than propionic acid, the pH in the hindgut lumen won't decrease as much as when horses are fed a higher proportion of concentrates in the diet (Varlout *et al.*, 2004). Also, a high proportion of forage in the diet stimulates the secretion of gastric fluid to a higher extent than high proportion of concentrates. Forages stimulate a more intense and prolonged secretion of gastric fluids due to the longer intake time compared to concentrates. One way to stimulate the production of gastric fluid is to feed forage before feeding concentrates. When the horse is fed roughage feed passes from the stomach at the same rate as it enters the stomach. This means that pH does not decrease as much when the horse is fed mainly concentrates, which may result in survival of foreign bacteria (Frape, 2010). Apart from providing the horse with primarily energy from microbial fiber fermentation, the large intestine also functions as a fluid reservoir. This is possible as fiber binds water to itself. Fibers have a high water holding capacity and horses having a roughage based ration have a greater fluid reservoir relative to horses on a high concentrate diet. When the horse cannot maintain its hydration status in body tissues, fluid is taken from the colon to compensate and protect the liquid volume in tissues. This means that an insufficient amount of fiber in the diet may cause constipation colic in the horse, as the water volume in the intestine may be too small (Nyman, 2001).

4.2. Starch

Starch rich feeds have the best documented influence on colic risk in horses (Shirazi-Beechey, 2008). Diet composition, particularly carbohydrate composition, affects the microbial population of the large intestine as well as the proportions of VFA's that are produced (NRC, 2007). Diets with a high inclusion of concentrate and grains, with a high proportion of starch (Table 1), gives a high production of propionic acid and enable lactic acid fermentation, and can lead to a drastic decrease in hindgut pH which increases the risk of intestinal disorders and colic (Varlound *et al.*, 2004). When the horse is fed large amounts of grains, starch is not digested in the small intestine and therefore continues into the colon where the starch is fermented by microorganisms. This may lead to increased formation of lactate and propionate, and a decreased proportion of acetate as well as decreased pH. These changes in the environment of the gastrointestinal tract may cause digestive problems and result in conditions like colic (Julliand *et al.*, 2001).

Table 1. Starch content of commonly used feeds for horses. Modified from Jansson *et al.*, (2011)

Feed	Amount of starch (g/kg dry matter)
Molassed sugar beet pulp	0
Grass	0
Lucerne	15
Oats	340
Barley	520
Wheat	645
Corn	710

The concentration of amylase in the pancreatic fluid in horses is very low compared to other animal species, causing a relatively poor prececal starch digestibility in horses. The combination of the limited prececal starch digestibility and the fermentative effects of starch suggest that equids are poorly suited to high-starch diets (Shirazi-Beechey, 2008). Incomplete prececal digestion of starch is one of the most important factors to consider in the association between diet and colic (Gonçalves *et al.*, 2002). High-grain diets, with high amounts of starch, have also been shown to result in a rapid transit time of digesta through the stomach and small intestine. This reduces the time for digestion and absorption of available starch in the small intestine. Pelleted and ground feeds move faster than forage and even if processed feeds are used (where prececal starch digestibility is increased), there is always a risk, that undigested starch will reach the large intestine, which has been shown to have an association with digestive disturbances like colic (Bentz, 2004). Also, starch from different cereal sources has different proportions of straight-chain amylose and branched-chain amylopectin and therefore differing relative digestibility. Oat and sorghum starch is more highly digestible than starch in barley, wheat or maize, which can explain the different impacts of these feeds on the digestive tract. The capacity to digest starch can be equivalent to 0.4 kg oats per 100 kg body weight (BW), and grains containing larger amounts of starch than oats, *e.g.* barley and maize, should therefore be given in smaller amounts in the diet (Frape, 2010).

High-starch diets has been shown to have a negative effect on the digestion, gastrointestinal environment (bacteria) and VFA's produced, and therefore as having a negative effect on the horse health. Diets high in concentrates can cause disruption of the normal colon function (Clarke *et al.*, 1990).

Feeding high-starch diets has been shown to increase the concentration of total anaerobic and lactic acid-utilizing bacteria ($p < 0.001$) and decrease the numbers of cellulolytic bacteria ($p < 0.05$) in the cecum and also been shown to increase the numbers of Lactobacilli and Streptococci ($p < 0.001$) in the cecal and the colonic contents, which is associated with a decrease ($P < 0.001$) of pH, (acetate + butyrate)/ propionate ratio and with an increase ($p < 0.001$) in lactic acid concentration (Medina *et al.*, 2002). Lactic acid concentration has been reported to be higher in horses fed starch-rich grain, followed by a marked lowering of the colon pH from 7.5 to 6.2, and has a major effect on microorganisms and fermentation in the colon. Also, in the same study bacteria that can easily ferment starch rather than structural carbohydrates, will multiply rapidly in starchy surroundings and produce not only an excess of lactic acid but also large amounts of carbon dioxide that causes gastric distension and pain (Shirazi-Beechey, 2008).

4.3. Water

Horses are supplied with water through direct consumption, indirect consumption from the feed and through metabolic water formed in the body (Frape, 2010). The loss of water in horses is mainly through faeces, urine, evaporation, breath and sweating (Nyman & Dahlborn, 2001). During exercise, when sweat losses increase, horses can lose up to 15 liter of fluid per hour (Jansson *et al.*, 2011). In order to meet the maintenance requirement of water, approximately 5 liters of water per 100 kg of body weight per day is required (Jansson *et al.*, 2004). The amount of water that has to be provided can be altered or decreased dependent on sweat losses, ambient temperature and dry matter (DM) content of the feed. If the horse is fed feeds with a high DM content like hay, which has a DM of approximately 85%, the water consumption will increase and when provided feed with a lower DM content like pasture or wrapped forages, water consumption will decrease (McDonnell *et al.*, 1999; Williams *et al.*, 2014). Stabled horses, consuming roughages with high DM, has been shown to have a three times larger water consumption than horses kept on pasture grass (Williams *et al.*, 2014). The water consumption is also dependent on the type of water source provided for the horse due to amount of water and water flow in automatic waterers. Studies have shown that a water flow of 8 liter per minute in automatic waterers is equivalent to if the horse is provided with water from a bucket with open surface. Also, it has been shown that a water flow of 6 liter per minute was also concluded to provide a sufficient water intake, but a water flow of 3 liter per minute could not provide the amount of water needed to meet the horse requirements (Nyman & Dahlborn, 2001). A limited water intake can result in a reduced appetite followed by a reduced feed intake as a result and may also contribute to impaired performance and metabolic disturbances such as colic (Frape, 2010).

4.4. Salt

Salt, or more specific sodium (Na), is important for the horse to maintain the fluid volume in the body. Losses of sodium occur mainly through faeces and urine (about 1.5 g/100 kg body weight and day) and by sweating. Sodium is not available in sufficient quantity in the natural diet of the horse and must therefore be added. Some horses are able to regulate their sodium requirement themselves if they are offered a salt block *ad libitum*, but those who make large salt losses through sweat do not. All horses should have access to a salt block (except for foals) but performing horses must also be offered extra salt. Racing-, eventing- and endurance horses that make greater sweat losses several times a week can be offered extra sodium by adding table salt (NaCl) in the diet, or as a physiological salt solution (9 g NaCl or less per liter of water) (Jansson *et al.*, 2004). When horses have a decreased intake of salt it might lead to a decreased ability to maintain fluid balance in the body and a decreased water intake. The

concentration of sodium in the body fluid is one of the controlling factors for the total fluid volume in the body. When sodium concentration in the blood is high, the horse will feel thirst and drink water to regulate sodium concentration and body fluid volume. If the concentration of salt is low in the blood, the horse will get an increased appetite for salt (Sjaastad *et al.*, 2010) and consume more salt, if it is readily available.

4.5. Feeding recommendations

Starch boluses fed at greater concentrations than 300 g/100 kg bodyweight (BW) have been shown to have a marked effect on microbial populations in the hindgut with perhaps half of the ingested starch reaching the cecum. Also, the same studies have shown that starch boluses fed at 100 to 200 g/100 kg BW per meal still frequently provide 40 g starch/ 100 kg BW to the cecum (Harris and Arkel, 2005; Gonçalves *et al.*, 2002). In order to avoid strain on the horse's digestive tract and the development of digestive disturbances, feeding recommendations have been developed. These recommendations include restraints for the amount of starch-rich feeds; a maximum of 0.4 kg of grain-based concentrates per 100 kg BW and meal, and a maximum of 150 g starch/100 kg BW and meal. It is important that the horse is provided with enough roughage in the diet and the recommendations are at least 1 kg DM/ 100 kg BW and day as a minimum, but preferably around 1.5-2 kg DM per 100 kg BW and day. Also, horses should be offered roughage before concentrates (if any are needed) at any meal to avoid digestive disturbances and welfare issues (Jansson *et al.*, 2011).

5. Colic

Colic is defined as abdominal pain, often of digestive origin, which causes a disruption of intestinal physiology and function of the digestive tract (Pilliner & Davis, 2004). It is commonly described as a symptom and/or a condition rather than a disease (Archer & Proudman, 2006). Horses are hindgut fermenters and the modern practice has led to a diet including more starch rich- and dry feeds. This type of diet, and subjecting the horse to changes in feeding- and watering practices causes alterations in the environment of the gastrointestinal tract which might lead to colic (Bentz, 2004).

Horses can suffer from different types of colic and most colic cases do not require surgery to resolve. These types of colic are often referred to as medical colic and include gas- and impaction colic. The other type, referred to as surgical colic, includes rotation and/or position changes of parts of the gastrointestinal tract which requires surgery to resolve (Bentz, 2004). Impactions are a common cause of colic and occur as a result of accumulated ingested feed, causing an obstruction within the intestinal tract that inhibits passage of intestinal contents (Reeves, 1997). Impactions can occur throughout the gastrointestinal tract, but most frequently affect the large colon as a result of a too low water intake and/or water content in the feed ration (Bentz, 2004). Sand colic, due to ingested sand that remains in the intestine and end up in cecum- and colon, is another form of impaction which may occur when horses graze on sandy soils and/or have limited access to grazing. Another type of colic is gas colic which is when there is a too large formation of gaseous products of microbial digestion in the large intestine. This can be caused by a too large ingestion of easily fermented carbohydrates (*e.g.* soluble starch) which then causes an increased fermentation rate. As a result, the gaseous swelling in the hindgut may lead to colic (Cunha, 2012).

Enteroliths, “stones” formed within the intestinal tract, is another nutrition-related cause of colic. They are most commonly formed in the large intestine as a result of excessive levels of dietary magnesium, nitrogen, and phosphorus, which forms struvite “stones” or enteroliths.

On the basis of reported feeding and management practices, horses with enteroliths were fed a higher proportion of lucerne in their diet and were less likely to have daily access to pasture grass than horses without enteroliths (Hassel *et al.*, 2004).

5. 1. The epidemiology behind colic

Previous epidemiological studies have illustrated the complex and multifactorial nature of colic (Reeves, 1997) by illustrating that colic is affected by both non-nutritional and nutritional-related risk factors.

One of the non-nutritional factors include weather, where associations have been made between an increased risk of colic and recent weather changes (Cohen & Gibbs, 1999); increased ambient temperature (Kaya *et al.*, 2009); spring- and autumn season (Proudman, 1991); and summer season (Hillyer *et al.*, 2002; Traub-Dargatz *et al.*, 2001). Other non-nutritional factors reported to increase the risk of colic are use of deworming products (Reeves *et al.*, 1996; Cohen & Gibbs, 1990; Kaya *et al.*, 2009) and recent anthelmintic administration (Hudson *et al.*, 2001; Cohen & Gibbs, 1999; Love, 1997; Kaya *et al.*, 2009). Previous history of colic (Tinker *et al.*, 1997; Cohen *et al.*, 1999; Cohen & Peloso, 1996; Hillyer *et al.*, 2002; Fielding and Dechant, 2012) and previous abdominal surgery (Cohen *et al.*, 1995; Cohen & Peloso, 1996) are two other risk factors associated with increased risk of colic. Individual factors, including age, gender and breed, has also shown to be associated with an increased risk for colic; age groups found to be associated with an increased risk of colic are horses < 6 months old (Traub-Dargatz *et al.*, 2001; Reeves *et al.*, 1989; Cohen *et al.*, 1997; Southwood *et al.*, 2002), 2-10 years old (Tinker *et al.*, 1997; Mehdi & Mohammad, 2006), >8 years old (Cohen & Peloso, 1996), 5-15 years old (Penell, 2009) and > 10 years old (Cohen & Gibbs, 1999; Reeves *et al.*, 1996; Kaneene *et al.*, 1997). Geldings have also shown to be at an increased risk of colic (Kaya *et al.*, 2009; Cohen & Peloso, 1996; Abutarbush *et al.*, 2005) relative to mares and stallions. Among horse breeds, Arabian horses (Reeves *et al.*, 1996; Cohen & Gibbs, 1999), Warm-blooded horses (Reeves *et al.*, 1996), Thoroughbreds (Tinker *et al.*, 1997; Traub-Dargatz *et al.*, 2001; Sawesi *et al.*, 2015), Crossbred horses (Mehdi & Mohammad, 2006) and Swedish Warm blood horses (Penell, 2009) have been shown to be associated with an increased risk of colic. Working horses, horses used for hobby riding (Kaya *et al.*, 2009), horses participating in showing activities (Kaneene *et al.*, 1997) and racing (Sawesi *et al.*, 2015) has also been reported to be at an increased risk of colic. Also, horses having a change in activity level, due to injuries or breaks from competitions (Hillyer *et al.*, 2002; Cohen *et al.*, 1997; Cohen *et al.*, 1999) has been showed to be at an increased risk of colic. Many of the studies investigating associations between non-nutritional factors and colic risk has also investigated the association of nutritional risk-factors, and in many studies nutrition-related risk factors have emerged as the main factors for increased risk of colic.

6. Nutrition-related risk factors for colic

Although there are some contradictory results, several studies have found associations between increased risk of colic and several nutritional factors. These factors include type and amount of feed, feeding practices (frequency and number of feedings per day), watering practices and also, changes in feeding- and watering practices.

6.1. Type and amount of feed

The type and amount of feed is one of the most investigated nutritional risk factors for colic. The findings from previous studies have illustrated an association between colic and the total amount of concentrate in the diet, as well as the amount of specific types of feeds. Even though forage has long been known to promote a healthy GIT environment, in comparison to concentrates, studies have also found that the amount of forage and specific types of forage may be associated with an increased risk of colic (Gonçalves *et al.*, 2002).

6.1.1. Concentrate and grains

The amount of concentrate fed daily has been shown to be associated with an increased risk of colic, although some studies have not found any associations between the amounts of concentrates fed daily and increased risk of colic (Traub-Dargatz *et al.*, 2001).

An association between the amount of concentrate consumed daily and presence of colic was also found in another study, but this result showed that horses having a higher daily intake of concentrates were more likely to be colic-horses relative to horses receiving lower amounts of concentrates in the diet (Tinker *et al.*, 1997). The study was designed as a prospective cohort study from a random population of horses on 31 different farms in Maryland and Virginia, USA. The amount of concentrates given to the horses were classified as 0 kg/day DM, <2.5 kg/ day DM, 2.5-5 kg/day DM and > 5 kg/day DM. The results showed that horses receiving 2.5-5 kg/ day were almost five times ($p=0.01$; OR= 4.8; 95% CI= 1.4-6.6) more likely to be colic horses relative to horses not fed any concentrates. The strongest association with colic was shown for the horses fed >5 kg/ day, as these horses were over than six times ($p=0.004$; OR=6.3; 95% CI= 1.8-22.0) more likely to be colic cases relative to horses not fed any concentrates. The author also investigated whether the amount of whole grain (oats, barley or other unprocessed grain) were associated with presence of colic. The results showed that horses consuming whole grains were less likely ($p=0.01$; OR= 0.4; 95% CI= 0.2-0.8) to be colic cases relative to horses not fed whole grain in the diet. In addition, odds ratios for concentrate type variables were analyzed in an alternate model with type replacing DM intake. The results from this analysis showed that horses fed whole grain only were shown to be almost six times ($p=0.09$; OR=5.8, 95%; CI= 0.8-4.5) more likely to be colic cases relative to horses fed a forage only diet. Also, the results showed that horses fed a mixed feed including whole grain were shown to be almost three times ($p=0.12$; OR=2.9; 95% CI= 0.8-11.2) more likely to be colic cases relative to horses fed a forage only diet, and that horses fed a diet including whole grain were less ($p=0.29$; OR=0.6; 95% CI= 0.2-1.5) likely to be colic cases relative to horses not fed whole grain (Tinker *et al.*, 1997).

An association between an increased likelihood for colic and a higher amount of concentrate was also found by Kaya *et al.* (2009). This study was designed as hospital-based case study where the data was based on 366 colic horses and 2377 non-colic horses admitted to a veterinary hospital in Vienna (Austria) during a 1-year period. Some information regarding feeding and watering practices was not available from the medical records, and was therefore provided through a questionnaire handed out to the horse owners. The average daily amount of concentrate (classified as oats, squeezed oats, corn, cereals and pellets) fed to the horses was shown to be higher for the colic-horses (2.74 ± 2.09 kg per day) than for the non-colic horses (1.64 ± 1.16 kg per day), and was associated with an increased risk for colic ($p= 0.04$) (Kaya *et al.*, 2009). Another study also found that an increased amount of concentrate was associated with an increased risk of colic, and in this study the risk for specific types of colic (simple colonic obstruction and distension) was studied (Hillyer *et al.*, 2002). The study was

designed as a case-control study, including 76 case and 76 control horses, where horses were recruited from veterinary school clinics in Bristol and Liverpool, UK. The amount of concentrates was classified as; >1.8 kg and <1.8 kg concentrate per day. The result showed that horses that were fed > 1.8 kg concentrate per day were over two times (OR= 2.76; 95%CI= 1.42-5.35) more likely to have simple colonic obstruction and distention compared to when horses were fed (<1.8 kg) concentrate/ day (Hillyer *et al.*, 2002).

An association between colic and the amount of non-roughage feed (concentrates) was found in a study by Reeves *et al.* (1996). In this study, 406 colic and 406 non-colic horses that were admitted to a veterinary clinic in Ohio, USA, were included. When investigating the amount of feed consumed, the non-colic horses were reported to have a higher average intake of concentrate (3.9 ± 0.11 kg per day) relative to colic horses (3.4 ± 0.10 kg per day). The results showed that a one kg increase in the amount of concentrate consumed daily were associated with a 12 % reduced (OR=0.8, 95% CI= 0.8-0.95) risk of colic (Reeves *et al.*, 1996).

6.1.2. Specific type of concentrate- Oats and corn

The amount of specific types of feed has also been investigated and the results have been contradictory. The daily consumption of oats has not been reported to be associated with colic in some studies (Reeves *et al.*, 1996; Tinker *et al.*, 1997; Cohen *et al.*, 1999; Hudson *et al.*, 2001; Traub-Dargatz *et al.*, 2001; Kaya *et al.*, 2009;). In the study performed by Hudson *et al.* (2001) the average intake of oats was 1.4 kg (0.1-12.7 kg) per day, however no association was found. The amount of corn consumed daily has been shown to be associated with an increased risk of colic. This association was detected by Reeves *et al.* (1996), who reported horses with colic consuming more whole-grain corn (1.3 ± 0.18 kg/ day) compared to non-colic control horses (0.7 ± 0.13 kg/ day). The results showed that for every 1 kg increase of corn per day, the presence of colic increased over three-fold ($p=0.03$; OR=3.4, 95% CI=1.45-7.83); however when all non-roughage concentrate feeds were combined, colic risk decreased with increased intake of concentrates. Other studies have also found that there is no association between the amounts of corn fed daily and presence of colic (Kaya *et al.*, 2009).

6.1.3. Specific types of concentrate-Processed feeds and feed additives

The amount of processed feeds, such as pelleted concentrates and sweet feeds has been reported to be associated with presence of colic. An association was found by Tinker *et al.* (1997), where results showed that horses that were fed pelleted concentrate were over six times ($p=0.007$; OR= 6.1; 95% CI= 1.6-22.6) and the horses fed sweet feeds as concentrate over four times ($p=0.01$; OR= 4.6; 95% CI= 1.4-15.6) more likely to suffer from colic, relative to horses not fed these feeds. However, in other studies no association was found between colic processed feeds (Cohen *et al.*, 1999; Hudson *et al.*, 2001; Kaya *et al.*, 2009).

Whether or not horses are fed vitamins and mineral supplements (including salt-licks) has also been shown to be associated with colic. In a study conducted with the aim to investigate the risk factors for developing enterolithiasis it was found that horses supplemented with vitamins and minerals (including mineral block) had a reduced risk of colic ($p=0.03$; OR=0.3; 95% CI, 0.1–0.9) relative to horses not supplemented with minerals and vitamins (Archer *et al.*, 2008). The feeding strategies for salt and the type of salt supplemented in the diet were investigated by Tinker *et al.* (1997) as possible risk factors for colic in horses. However, neither the strategies for supplementing salt in the diet nor type of salt fed were shown to be associated

with colic. However, access to a salt-block has been shown to be associated with colic in other studies; Archer *et al.* (2008) found that horses that had access to a mineral block were less likely to be colic horses (Archer *et al.*, 2008). The study was designed as a prospective unmatched, multicenter case-control study including 77 colic horses and 216 control horses admitted to clinics in the UK. The aim was to investigate the association between epiploic foramen entrapment (EFE), which is one of the most common causes of small intestinal strangulation, and overall management practices including feeding practices. The results showed that horses having access to a mineral block were less likely ($p=0.03$; OR, 0.3; 95% CI= 0.1–0.9) to be colic horses relative to horses not having access to a mineral-salt lick. The author concluded that this might be due to that the salt-lick salt lick stimulated salivation and therefore acting as a gastrointestinal buffer that offered some protection against EFE (Archer *et al.*, 2008).

6.1.4. Forage

Studies investigating the association between colic and type and amount of forage have shown that consumption of certain types of forage can be associated with an increased colic-risk. A study was performed by Cohen & Peloso (1996) with the aim to identify risk factors for colic in horses in a case-control study at the university equine hospital in Texas, USA. A total of 232 colic horses and 536 non-colic horses were compared and the results showed that feeding of coastal grass hay was associated with an increased risk ($P = 0.045$; OR = 1.6), of colic relative to horses fed other types of hay (wheat hay, Bahia grass hay, oat hay, timothy hay and Alicia hay). These results were contradictory to a prospective matched case-control study performed by Cohen *et al.* (1999) in Texas, USA. In this study 1030 colic and 1030 non-colic horses were examined by veterinarians who also provided data regarding the horse management. The results showed that horses fed other types of hay (wheat hay, Bahia grass hay, oat hay, timothy hay, hay grazer, cane hay and Alicia hay) were two times ($p=0.014$; OR=2.0; 95%CI=1.2-3.5) more likely to be colic horses relative to horses fed Coastal-, Bermuda- or Alfalfa hay (Cohen *et al.*, 1999). The results from this study is not supported by Hudson *et al.* (2001), who did not find an association between type of other types of hay (wheat hay, Bahia grass hay, oat hay, timothy hay, hay grazer, cane hay and Alicia hay) and colic.

6.2. Feeding strategies for concentrates and forage

Whilst horses naturally feeds ‘little and often’ the modern practice of fewer, larger meals, means that feed can enter the small intestine having been subjected to little or no gastric digestion. The modern way of keeping and feeding of horses has led to a reduced feeding time as the feeding is normally divided into two to four feedings per day in modern feeding practices (Warren, 2015). Although it is well accepted that abrupt dietary changes may lead to colic in horses, the common practice of feeding a few daily cereal-based meals interspersed by long periods of high-fiber forage intake (or no intake) of markedly differing nutritional quality represents marked intraday dietary change and probably diurnal variation in hindgut pH and bacterial populations, and perhaps promotion of colic. Therefore, for horses where a high inclusion of concentrate (especially starch-rich feeds) is present in the feed ration, the ration should be divided into three to five smaller portions over the day in order to not cause a drastic decrease in hindgut pH (Harris & Arkel, 2005). The association between number of concentrate feedings and presence of colic was investigated by Tinker *et al.* (1997), where 0, or >1 concentrate feedings per day were compared. The results showed that horses that were fed concentrate one time per day were more than three times ($p<0.001$; OR= 3.6; 95% CI=

1.6-5.4) more likely to be colic horses relative to horses not fed any concentrate. Also, the results showed that horses fed concentrates >1 time per day were more than two (p=0.02; OR= 2.2, 95% CI= 1.2-4.1) times more likely to be colic horses compared to horses not receiving any concentrate. However, dividing the feeding of >5 kg of concentrates in more than three times per day did not reduce the risk for colic associated with high levels of concentrates in the ration (Tinker *et al.*, 1997). The association between the number of daily feedings and colic has not been found in other studies (Cohen *et al.*, 1999; Hudson *et al.*, 2001; Kaya *et al.*, 2009).

Associations between colic and the number of feedings of roughage per day has not been investigated in the same extent as for concentrate meals, however Tinker *et al.* (1997) investigated if colic was associated with numbers of roughage feedings per day and compared feeding hay 0 , 1 or > 1 time/day. The results that when horses were fed hay >1 times per day they were more than two (p=0.01; OR= 2.1; 95% CI= 1.2-3.8) times more likely to be colic horses than horses not receiving any hay (Tinker *et al.*, 1997).

6.3. Dietary changes in concentrate and roughage

Diet changes, in both type and amount of feed, have been shown to be one of the main factors associated with presence of colic (Cohen & Peloso, 1996; Tinker *et al.*, 1997; Cohen & Gibbs, 1999; Hudson *et al.*, 2001; Hillyer *et al.*, 2001). High-starch diets and abrupt dietary changes are probably the most important risk factors for diet-associated colic in the horse, due to their disruption of the stability of microbial populations' resident within the equine hindgut (Durham, 2009). Changes in the diet, especially introduction of high-starch cereal grains to the feed ration, result in changes in the microbial population inhabiting the GI tract (Bailey *et al.*, 2003; de Fombelle *et al.*, 2003; Berg *et al.*, 2005; Varloud *et al.* 2007). One study investigated the effect of an abrupt incorporation of barley in a hay diet on the microbial profile and activity of the horse digestive tract. The diet changes investigated were (1) changing the diet from 100 % hay (100:0) to a diet of 70 % hay and 30 % barley (70: 30), and to a diet of 50% hay and 50% barley (50:50). The results showed an increased amount of total anaerobic bacteria, lactobacilli and streptococci in the cecum and colon, regardless the amount of barley in the diet change and also a decrease in the (acetate + butyrate/ propionate) ratio. The colon appeared to be the main site affected by the abrupt incorporation of starch in the diet. Changes in the diet should therefore be done gradually over a longer periods for the microbes in the gut to adapt to the new environment of the gastrointestinal tract (de Fombelle *et al.*, 2001), and the feeding recommendation is that a change in the diet should be done over a 14-day period (Jansson *et al.*, 2011).

The risk for colic due to a recent change in diet was investigated by Cohen and Peloso (1996) who found horses having a recent change in diet (2 weeks prior to examination) were two times (P=0.005; OR=2.1) more likely to be colic horses relative to horses not having a recent change in diet. This result were supported by the results from a study conducted by Cohen and Gibbs (1999), as they found that horses that had had a recent diet change were five times (p=0.0001; OR=5.0; 95% CI=2.6-9.7) more likely to be colic cases relative to horses not having a recent change in diet. An association between colic and a change in diet was also detected in the study by Tinker *et al.* (1997), who investigated if there was any association between the number of feed changes horses had been subjected to throughout the year and presence of colic. The number of changes investigated were; 0, 1 and >1 times, and the result showed that horses subjected to one change were more than three times (p<0.001; OR=3.6; 95%CI=1.6-5.4) more likely to be colic cases compared to horses not subjected to any change. Also, the authors concluded that horses that had >1 change/ year were more than two times

($p < 0.02$; OR=2.2; 95%CI=1.2-4.1) more likely to be colic cases, compared to horses not subjected to a diet change (Tinker *et al.*, 1997). An increased risk of colic was also detected when investigating recent changes in concentrate and grain feedings when Hudson *et al.* (2001) showed that horses subjected to a recent change (2-week period prior to examination) in the type of grain or concentrate tended to be two times ($p=0.064$; OR=2.6; 95%CI=0.9-7.2) more likely to be colic horses relative to horses not subjected to a change. These findings were supported by the results presented by Hillyer *et al.* (2002) which showed that horses having a recent change in concentrate feeding were more than seven times (OR=7.9, 95% CI 2.37-26.36) more likely to be colic horses relative to horses not receiving any concentrate or having a change in concentrate feeding. Also, it was shown that horses having this change in concentrate feeding 1-7 days prior to the examination were 12 times ($p < 0.001$; OR=12.0; 95%CI=2.7-54.4) and horses having this change 8-14 days prior to examination were three times ($p=0.05$; OR= 3.0; 95% CI=1.0-8.9) more likely to be colic horses relative to horses not receiving concentrate or not being subjected to a change in concentrate feeding (Hillyer *et al.*, 2002). Studies have also investigated if the time of the change in roughage feeding were associated with an increased risk of colic. Results from one study showed that horses subjected to a recent change in type of hay, 2 weeks prior to examination, were more than nine times ($p=0.0346$; OR= 9.8; 95%CI= 1.2-81.5) more likely to be colic horses relative to horses not having a recent change of hay (Cohen *et al.*, 1999).

Whether a change of hay was associated with an increased risk of colic or not have also been studied by Cohen & Gibbs (1999), and the results showed that horses having a recent change in batch and/or type of hay (2-week period before examination) were almost 30 times ($p < 0.001$; OR=29.5; 95% CI= 7.2-120.7) more likely to be colic horses relative to horses not having a change in batch or type of hay. A recent change in batch or source of hay as a risk factor for colic was also investigated by Hudson *et al.* (2001), showing that horses having a change of hay (type and/or batch) during a 2-week period before examination were more than four times likely ($p < 0.01$; OR=4.4; 95% CI=2.0-9.4) to be colic horses relative to horses not having a change in type or batch of hay during the same period. The author also investigated whether a recent change in amount of hay fed and if a recent addition of hay in the diet were associated with an increased risk of colic, however addition of hay in the diet was not associated with colic. The results however showed that horses having a recent change in the amount of hay fed were more than two times ($p=0.06$; OR=2.5; 95% CI=1.0-6.4) more likely to be colic horses relative to horses not having a recent change in amount of hay.

In another study (Hillyer *et al.*, 2002), a recent change in forage feeding was found to be associated with colic. When investigating effects of a recent changes in forage feeding, the results showed that a change within 4 weeks prior to examination increased the risk of colic over five times (OR=5.02; 95%CI=1.99-12.67) relative to horses not having a recent change in forage feeding. The specific time for the change in forage feeding was also investigated and the results showed that horses having a change in forage feeding 1-7 days prior to examination were 22 times (OR= 22.0; 95%CI= 2.84-170.41) more likely to be colic horses relative to horses not fed forage and/or not being subjected to a change in forage feeding during 4 weeks prior to examination. Also, the risk was still increased, but not as high, for horses subjected to a change in forage feeding 8-28 days before examination as these horses were almost five times (OR= 4.9; 95% CI= 1.28-18.60) more likely to be colic horses relative to horses not experiencing a change in forage feeding (Hillyer *et al.*, 2002). However, these changes were not shown to be associated with colic presence in a following multivariate analysis. When investigating the effect of a recent change in dry hay feeding specifically, the results showed that a recent change, 4-weeks prior to examination, in dry hay feeding

increased the risk of colic over six times (OR=6.73; 95% CI=1.72-26.38) relative no change in dry hay feeding. The specific time for the change in dry hay feeding was also investigated and the results showed that horses having a change in dry hay feeding 1-7 days prior to examination were 14 times (OR= 14.0; 95%CI= 1.72-113.79) more likely to be colic horses relative to horses not fed dry hay and/or not being subjected to a change in dry hay feeding during 4 weeks prior to examination. Also, the risk of colic was still increased, but not as high, for horses subjected to a change in dry hay feeding 8-28 days before examination as these horses were three times (OR 3.0, 95%CI 0.5-17.95) more likely to be colic horses relative to horses not experiencing a change in dry hay feeding (Hillyer *et al.*, 2002). However, these changes were not shown to be associated with colic presence in the multivariate analysis.

6.4. Watering practices

The most common type of water source used for horses is automatic waterers, but this type of water source can be problematic as water flow may be low compared to other sources (Dahlborn, 2010). Previous studies have shown that horses prefer water buckets prior to automatic waterers when flow rate in automatic waterers was three liters per minute (Nyman & Dahlborn, 2001). A flow of six to eight liters per minute in automatic waters has been found to give similar water intake as buckets (Nyman, 2001; Jansson *et al.*, 2011). Changed water access and water consumption has been shown to be associated with colic in several studies (Reeves *et al.*, 1996; Hudson *et al.*, 2001; Kaya *et al.*, 2009). Whether access or no access to water on outside enclosures were associated with increased risk of colic or not was investigated by Reeves *et al.* (1996), and the results showed that horses in outdoor enclosures without a continuous supply of water were more than twice as likely (OR=2.2; 95% CI= 1.2-4.3) to be colic cases relative to horses that had an adequate supply of water in outside enclosures. Changed access to water has been found to be associated with presence of colic in one study (Kaya *et al.*, 2009) and the results showed that horses having a changed access to water were over five times ($p < 0.001$; OR= 5.02; 95% CI= 2.47-10.21) more likely to be colic horses relative to horses not having changed water access. Also, the results showed that horses having a decreased access to water were over five times ($p < 0.001$; OR= 5.29; 95% CI= 2.23-12.56) more likely to be colic horses relative to horses not having a decreased access to water. The type of water source provided to horses has also been investigated and the results from one study showed that horses provided drinking water in groups from sources other than buckets and automatic waterers had reduced risk ($p=0.017$; O.R. = 0.16, 95% CI = 0.03 - 0.72) of colic relative to horses provided water from buckets and automatic waterers (Kaneene *et al.*, 1997). However, in studies performed by Hudson *et al.* (2001) and Kaya *et al.* (2009) neither type of water source provided in the paddock (trough, bucket or pond) or type of water source provided in stable (waterer or bucket) were associated with presence of colic.

7. Aim and objective

The aim of this study was to further investigate the association between nutrition-related factors and colic by describing feeding and watering routines in horses that have had colic episodes the last 6 months (colic horses) and compare these with horses without colic episodes the last 6 months (non-colic horses).

8. Materials and methods

8.1. Experimental design

The study was designed as a questionnaire study where the aim was to collect information about feeding and watering practices from owners of both colic and non-colic horses. The collection of data was made by a questionnaire “Feed and feeding strategies as risk factors for colic in horses” (Appendix 1) which was designed using the survey software Netigate (Netigate, 2016) and was available at: <https://www.netigate.se/a/s.aspx?s=258862X49585858X24911> during the period from January 18, 2016 to February 29, 2016. The questionnaire was advertised through the website Hästsverige (<http://www.hästsverige.se/nyhetslista1.html?-news=36001>) and also through the connected Facebook page (<https://www.facebook.com/Hast-Sverige/?fref=ts>). The survey was also advertised at the website Hippson (<http://.hippson.se/artikelarkivet/forskning/examensarbete-om-kolikhastar-hoppas-komma-fram.htm>). A link to the survey was present at the website of the Department of Animal Nutrition and Management (<http://www.slu.se/sv/institutioner/husdju-rens-utfodring-vard/nyheter/2016/enkat-om-utfodring-av-hast-i-relation-till-kolik/>) during the same time period.

8.2. Design of questionnaire

The questionnaire contained 17 questions about the horse, roughage and concentrate feeding practices, watering practices, colic incidents and changes in feeding- and watering practices. The general information about the horse and the feeding- and watering practices as well as a question whether the horse had had colic or not during the last six months were answered for all horses (colic and non-colic), whereas the more detailed questions about colic was only answered by respondents with horses that had had colic during the last six months. Questions about changes in feeding- and watering practices were divided for colic and non-colic horses, where respondents with a colic horse answered questions about changes made 0-4 weeks prior to colic and respondents with non-colic horses answered the same questions, but about changes made the last six months.

The following data was obtained from each horse (colic and non-colic) in the study: age; gender; breed; withers height (cm); body condition score (BCS); body weight (kg); time owned by present owner (months/years); which disciplines performed; training intensity; number of trainings per week; amount of hay fed daily (kg); amount of haylage fed daily (kg); amount of silage fed daily (kg); amount of straw fed daily (kg); amount of pelleted lucerne fed daily (kg); amount of chopped lucerne fed daily (kg); number of feedings of roughage per day; number of feedings with only straw per day; time between two feedings of roughage; how the roughage were fed in the stable; how the roughage were fed in the paddock. Amount of oats fed daily (kg); amount of barley fed daily (kg); amount of molassed sugar beet pulp fed daily (kg); amount of pelleted concentrate fed daily (kg); amount of müsli fed daily (kg);

amount of vegetable oil fed daily (ml); amount of other concentrate fed daily (kg); number of feedings of concentrate per day; number of feedings with only concentrate per day; the time fed concentrate in relation to when fed roughage; source of salt; amount of salt consumed daily (g); amounts of pelleted minerals fed daily (g); amounts of granulate minerals fed daily (g); amount of liquid minerals fed daily (ml); amount of pelleted vitamins fed daily (g); amount of powder vitamins fed daily (g); amount of liquid vitamins fed daily (ml); amount of other vitamins or minerals fed daily (g/ml); type of water source in stable; type of water source in paddock; type of waterer used in stable and paddock, and whether the horse had had colic or not during the past six months. If there was any change in type or amount of roughage; type of change in roughage (specified); if there was any change in type or amount of concentrate fed; type of change in concentrate (specified); if there was any change in access to water in the paddock (specified); if there was any change in access to water in the stable; if there was any change in type of water source in stable (specified); if there was any change in type of water source in paddock (specified).

The following data were only obtained from respondents with colic horses: which type of colic; time for colic episode; whether the horse was on pasture or not at the time for colic episode; if there were any changes in the horse surroundings 0-4 weeks prior to colic. In order to get as accurate answers as possible from the respondents, a help section were created for some of the questions, including pictures to illustrate different classes of body condition scores (BSC); definition of roughage; how to convert different volumes (liter and deciliter) of concentrate feed to kilograms; how to convert different volumes (deciliter) of feed additives to grams, and pictures to illustrate the different types of waterer used (Appendix 1).

8.3. Transformation of data

Data from the obtained questionnaires was exported into Excel (Microsoft Excel, 2010) to be processed and transformed before statistical analyses. This was mainly done to make sure that all data for each variable was expressed in the same way and that no redundant information was included other than the information asked for. If obvious mistakes were present, such as data entered in the wrong way or in the wrong place, it was changed or moved to the right place. Data that could not be interpreted, and therefore not possible to include in the statistical analyses, was changed to missing value. It was also possible for respondents to select the alternative “other” if the answer alternatives provided in the questionnaire did not correspond to the characteristics or management of the horse. If selecting “other”, the owners were asked to specify it by writing their own answer. These specified answers were then added as alternative answers to the questions in order to include them in the same variable. The transformation of data from the questionnaire resulted in 72 variables.

8.3.1 Reclassification of variables and additional variables created

In order to perform a statistical analysis for some of the variables, they had to be reclassified. These reclassifications were made for the variables breed and disciplines performed. In addition to the 23 breeds included in the questionnaire (Appendix 1), 17 additional breeds were specified by the owner under the options “other”, which resulted in a total of 40 different breeds. Therefore, a reclassification was made in which the horses were classified in six new classes as followed; Cold-blooded horses, warm-blooded horses, ponies, Thoroughbreds, Arabians, and other breeds, which were not specified in the received questionnaires. From the answers received about how the horse was fed and watered in the paddock and in the stable new variables were created. Each of these questions contained an alternative which said that the horse was not fed or watered in the paddock or stable and therefore these alternatives were

used to create new variables to be able to investigate if horses not fed roughage or watered in the stable or paddock were at higher risk of colic relative to those who were fed or watered in stable and/or paddock.

Also, some new variables were created, including existing variables or based on existing variables, for calculations of the amount of feed per 100 kg BW fed daily. Information about specific changes in roughage feeding, concentrate feeding, water source in stable and in paddock were all divided into two separate parts to enable the respondents to specify the specific type of change (Appendix 1). However, the two variables (changed from and changed to) were combined to illustrate the change by creating just one variable to specify the type of change. New variables were created by using information about the amount of specific feeds fed daily and the body weight of the horses. The first variable was created by using the given weight and the amounts of feed fed daily for all the feeds specified in the questionnaire (Appendix 1) including roughage, concentrate and additional feeds. This new variable was named amount of feed per 100 kg BW/ day and was calculated as follows;

Amount of feed fed daily/ 100 kg BW = ((amount of feed fed daily)/ (body weight/ 100))

The same procedure was used for calculating the total amount of roughage fed daily per 100 kg BW including the amount of hay, haylage, silage, straw and lucerne. Also, the same calculation was done for the total amount of starch rich feeds fed daily per 100 kg BW including oats, barley, pelleted concentrate and müsli.

8.4. Data analysis

All statistical analyses were performed using SAS (Statistical Analysis System) version 9.4 for Windows. Descriptive analysis and univariate logistic regression analysis were performed for all variables using the PROC LOGISTIC, PROC FREQ and PROC GLIMMIX procedure. Variables which showed a univariable significance of $p < 0.05$ was considered in the multivariable analysis. Multivariable logistic regression models were developed using a stepwise selection technique in which variables are added to the model step by step and in each step, the procedure also examines whether variables already in the model can be deleted (Olsson, 2002).

9. Results

In total 4100 questionnaires, representing 4100 horses, was received from horse owners and out of these 3319 were completed which resulted in a response rate of 80.9 percent. However, 1.7 percent (n=55) of the completed questionnaires were excluded from the study because of missing information about if the horse had had a colic episode or not during the last six months, which resulted in a total of 3264 horses being included in the statistical analysis, and a final response rate of 79.6 percent.

9.1. Descriptive statistics

9.1.1. Colic

Sixteen percent (n=523) of all horses were reported to have had at least one colic episodes during the last six months (colic horses), while 84 percent (n=2742) did not report any colic episodes during the last six months (non-colic horses). Almost all (99 percent, n=517) respondents with colic horses reported when the colic episode occurred during the last six

months, and 31 percent (n=158) of these reported that colic occurred in their horse 1-2 months ago. The distribution of when colic occurred is shown in Figure 1. Type of colic was reported for 98.3 percent (n=514) of the colic horses and 34 percent of these (n=175) had suffered from impaction colic. Distribution for other types of colic is shown in Figure 2.

9.1.2. Type and amount of roughage

Haylage was the most commonly fed roughage for both colic (60 percent, n=316) and non-colic horses (63 percent, n=1717) respectively, whereas silage was the least commonly fed type of roughage for both colic (5 percent, n=27) and non-colic horses (4 percent, n=122). Distribution of roughage type fed is shown in Figure 3.

Information about whether a feed analysis was available or not for the roughage fed were received from 95 percent (n=3091) of the respondents representing 93 percent (n=488) of the colic horses and 95 percent (n=2603) of the non-colic horses. The majority of the respondents of colic and non-colic horses, 60 percent (n=291) and 58 percent (n=1506), respectively, had access to a feed analysis report for on the roughage fed. Distribution of feed analysis performed is shown in Figure 4.

9.1.3. Type and amount of concentrate

Thirty-four percent (n=180) of the colic horses and 29 percent (n=795) of the non-colic horses were fed molassed sugar beet pulp, making it the single most commonly fed concentrate feed for colic horses and the second most commonly fed concentrate feed for non-colic horses. Pelleted concentrate was fed to 32 percent (n=168) of the colic horses and 33 percent (n=897) of the non-colic horses, making pelleted concentrate the single most commonly fed concentrate among non-colic horses. Distribution of the type of concentrates fed is shown in Figure 5.

9.1.4. Type and amount of feed supplements

Sixteen percent (n=82) of the colic horses and 19 percent (n=506) of the non-colic horses, were reported to be fed pelleted minerals and 9 percent (n=46) of the colic horses and 8 percent (n=218) of the non-colic horses were reported to be fed liquid vitamins. Other types of minerals and vitamin feeds, in addition to the ones specified above, was fed to 3 percent (n=17) of the colic horses and 4 percent (n=116) of the non-colic horses, but due to missing or poorly given information about the specific type of other vitamin or mineral feed used, it was excluded from the statistical analysis. The distribution of which types of feed supplements that was fed is shown in Figure 6.

Information about the daily salt intake, as a class variable (i.e. no salt, <10 g, 11-24 g and >25 g), were received from 88 percent (n=2873) of the respondents representing 89 percent (n=463) of the colic horses and 88 percent (n=2410) of the non-colic horses. The majority of colic (79 percent, n=368) and non-colic horses (85 percent, n=2051) respectively, had a daily salt intake >25 g. Distribution of daily salt intake is shown in Figure 7.

Information regarding if the horses had free access to a salt block were received from 99.2 percent (n=3237) of the respondents representing 99.6 percent (n=521) of the colic horses and 99.1 percent (n=2716) of the non-colic horses. The majority of the colic and non-colic horses, 77 percent (n=402) and 83 percent (n=2242) respectively, had access to a salt block. The distribution of in which way horses had access to salt is shown in Figure 8.

9.1.5. Feeding routines for roughage

The number of roughage meals per day were reported for 98.7 percent (n=3224) of the respondents which represented 98.5 percent (n= 515) of the colic horses and 98.8 percent (n=2709) of the non-colic horses. Forty-two percent (n=218) of the colic and 34 percent (n=922) of the non-colic horses were fed roughage four times daily. The distribution of the number of roughage meals per day is shown in Figure 9.

The number of meals per day with straw as the only roughage offered were reported from 80 percent (n=2600) of the respondents which represented 78 percent (n=411) of the colic horses and 80 percent (n=2189) of the non-colic horses in the study. The majority of the colic and non-colic horses, 70 percent (n=288) and 68 percent (n=1489) respectively, were not fed straw as the only roughage at any meal. The distribution of number of feedings with straw as the only roughage is shown in Figure 10.

The majority of colic and non-colic horses, 95 percent (n=496) and 89 percent (n=2430) respectively, were reported to be fed roughage in the stable. Also, a majority of colic and non-colic horses, 92 percent (n=92) and 95 percent (n=2592) respectively, were reported to be fed roughage in the paddock. The distribution of where horses were fed roughage is shown in Figure 11.

9.1.6. Feeding routines for concentrates

The number of concentrate meals fed per day were reported from 93 percent (n=3020) of respondents which represented 94 percent (n= 491) of the colic horses and 92 percent (n=2529) of the non-colic horses in the study. Thirty-four percent (n=165) of the colic horses were reported to be fed concentrate two times daily, whereas 36 percent (n=905) of the non-colic horses were reported to be fed concentrates one time daily. The distribution of the number of concentrate meals fed daily is shown in Figure 12.

The timing for feeding concentrate (fed together with forage, or before or after) were reported from 93 percent (n=3020) of the respondents in which represented 94 percent (n= 491) of the colic horses and 92 percent (n=2529) of the non-colic horses in the study. The majority of the colic and non-colic horses, 60 percent (n=290) and 62 percent (n=1518) respectively, were reported to be fed concentrates together with roughage. The distribution of when the horses were fed concentrate in relation to roughage feeding is shown in Figure 13.

9.1.7. Watering strategies

The majority of colic and non-colic horses, 99 percent (n=517) and 96 percent (n=2643) respectively, were reported to have access to water in the stable. The distribution of watering routines is shown in figure 14.

9.1.8. Change in type and amount of roughage

A change in type or batch of roughage were reported from 93 percent (n=3050) of the horses representing 95 percent (n=498) and 93 percent (n=2552) of colic and non-colic horses respectively. The majority of colic and non-colic horses had not been experiencing a change in diet during the last six months, 83 percent (n=411) and 69 percent (n=1764) respectively. The distribution of changes in type and batch of roughage is shown in Figure 15.

A change in the amount of roughage fed were reported from 90 percent (n=2936) of the respondents representing 95 percent (n=489) of colic and 89 percent (n=2447) of non-colic horses. The majority of colic (86 percent, n=421) and non-colic horses (50 percent, n=1234) were reported not to have had a change in amount of roughage. The distribution of changes in the amount of roughage fed is shown in Figure 16.

9.1.9. Change in type and amount of concentrate

A change in type and/or batch of concentrate were reported from 96 percent (n=3125) of the respondents representing 94 percent (n=492) of colic and 96 percent (n=2633) of non-colic horses. The majority of the colic and the non-colic horses had not been experiencing a change in the amount of concentrate, 92 percent (n=453) and 77 percent (n=2029) respectively. The distribution of changes in amount of concentrates fed is shown in Figure 17.

A change in the amount of concentrates fed were reported from 87 percent (n=2826) of the horses representing 88 percent (n=461) of colic and 86 percent (n=2365) of non-colic horses. The majority of the colic and non-colic horses, 88 percent (n=406) and 67 percent (n=1586) respectively, were reported to not have experienced any change in amount of concentrates fed during the last six months. The distribution of changes in amount of concentrate is shown in Figure 18.

9.1.10. Changed access to water

A changed access to water in the stable were reported from 96 percent (n=3147) of the respondents representing 97 percent (n=507) of colic and 96 percent (n=2640) non-colic horses. The majority of the colic and non-colic horses, 97 percent (n=493) and 96 percent (n=2536) respectively, were reported to not have had a change in access to water in the stable during the last six months. The distribution of how horses had access to water in stable is shown in Figure 19.

9.1.11. Changed type of water source

A change in type of water source in the stable were reported from 93 percent (n=3029) of the respondents, representing 94 percent (n=492) and 93 percent (n=2537) of the colic and the non-colic horses respectively. The majority of the colic and the non-colic horses had not experienced any change in the type of water source in the stable, 95 percent (n=468) and 92 percent (n=2333), respectively. The distribution of changes in type of water sources in the stable is shown in Figure 20.

A change in type of water source in the paddock were reported from 86 percent (n=2799) of the respondents representing 84 percent (n=441) of colic and 86 percent (n=2358) non-colic horses. The majority of the colic and the non-colic horses had not experienced a change in the type of water source in the paddock, 93 percent (n=411) and 82 percent (n=1944) respectively. The distribution of changes in type of water source in the paddock is shown in Figure 21.

9.1.12. Number of trainings per week

Number of trainings per week was reported from 94 percent (n=3056) of the respondents representing 93 percent (n=485) colic and 84 percent (n=2571) non-colic horses. Forty-one percent (n=198) and 39 percent (n=1005) of colic and non-colic horses respectively, were

trained three to four times per week. The distribution of number of trainings per week is shown in Figure 22.

9.1.13. Individual factors

Age was reported for 99 percent (n=520) colic and 99 percent (n=2721) non-colic horses. The majority of the colic and non-colic horses, 51 percent (n=264) and 55 percent (n=1491) respectively, were reported to be between 7-15 years old. The distribution of age is shown in Figure 23.

Gender of the horse were reported from a total of 99 percent (n=3238) of the respondents representing 99 percent (n=519) colic and 99% (n=2719) non-colic horses. The majority of the colic and non-colic horses, 51 percent (n=268) and 52 percent (n=1420) respectively, were reported to be geldings. The distribution of gender is shown in Figure 24.

Breed type was reported for a total of 98 percent (n=3191) of the horses representing 99 percent (n=516) of the colic and 98 percent (n=2675) of the non-colic horses respectively. The majority of the colic- and non-colic horses, 59 percent (n=305) and 60 percent (n=1604) respectively, were reported to be warm-blooded horses. The distribution of breeds is shown in Figure 25.

Body condition score was reported by 94 percent (n=3060) of the respondents representing 92 percent (n=483) of the colic and 94 percent (n=2577) of the non-colic horses. On a scale of 0-5, the largest proportions of colic and non-colic horses, 51.6% (n =252) and 59.0% (n=2675) respectively, had a body condition score of 3. Distribution of other BCS is shown in Figure 26.

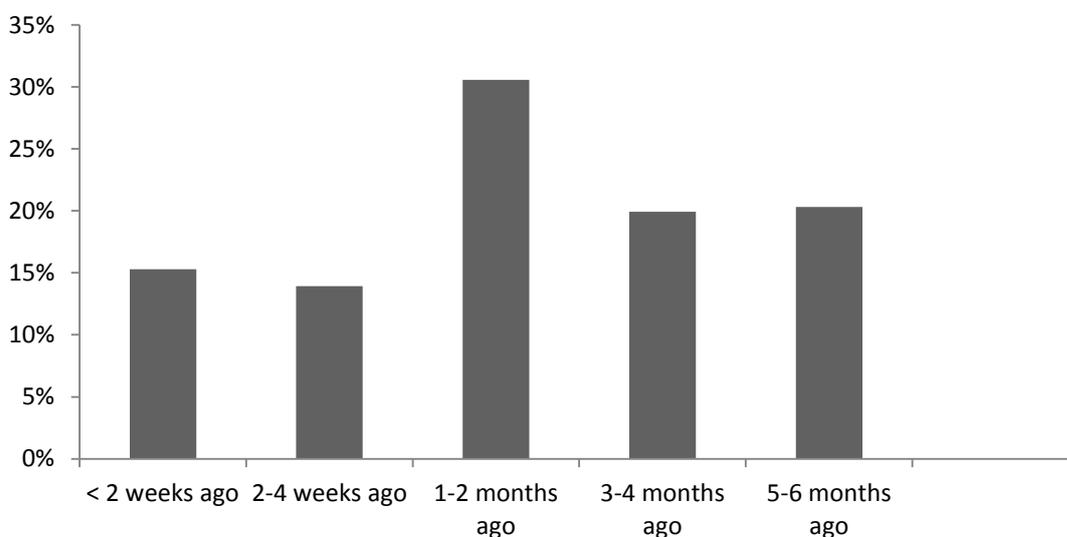


Figure 1. Distribution of when the horses in the study suffered from colic, in relation to time point of questionnaire response.

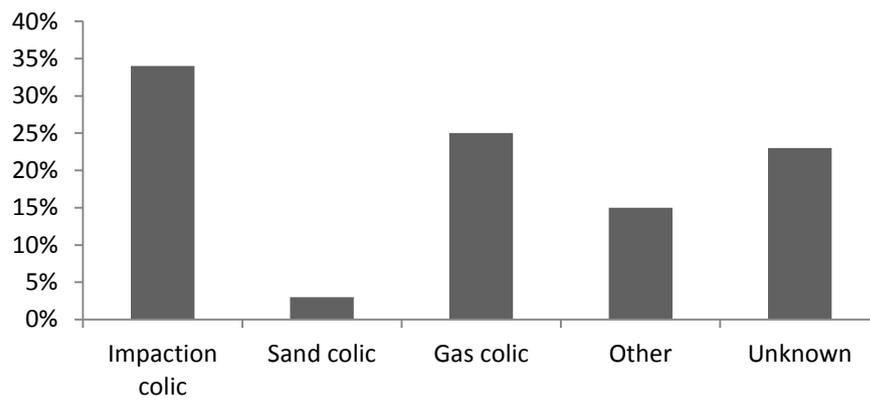


Figure 2. Distribution of the type of colic among colic horses in the study.

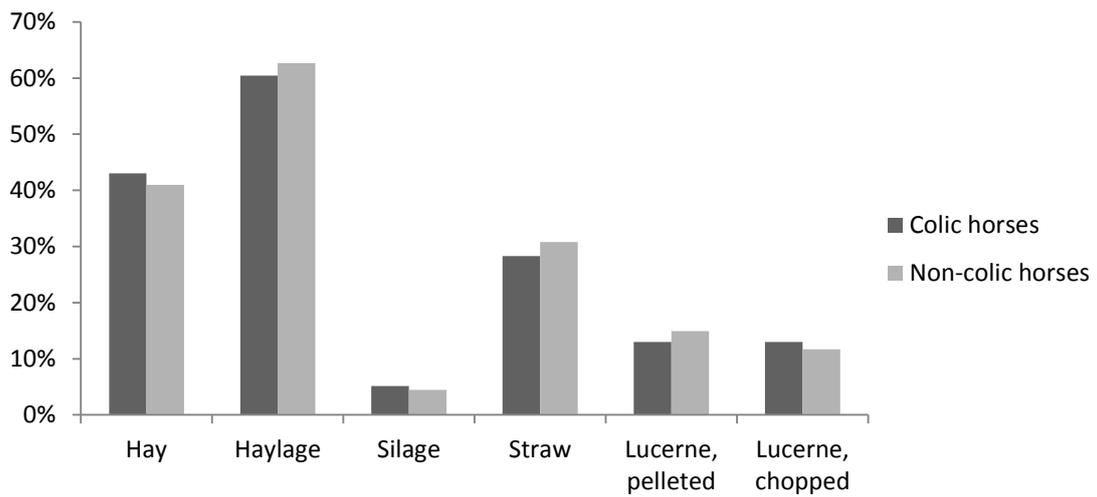


Figure 3. Distribution of type of roughage fed to the horses in the study.

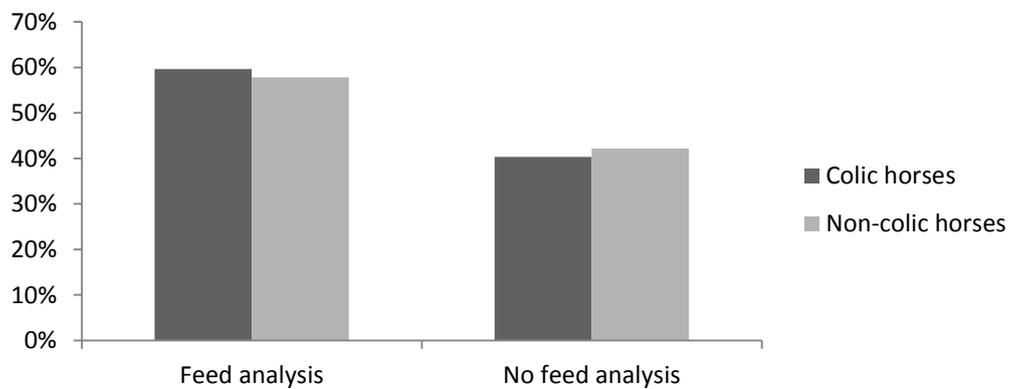


Figure 4. Distribution of feed analysis of the roughage used among the horses in the study.

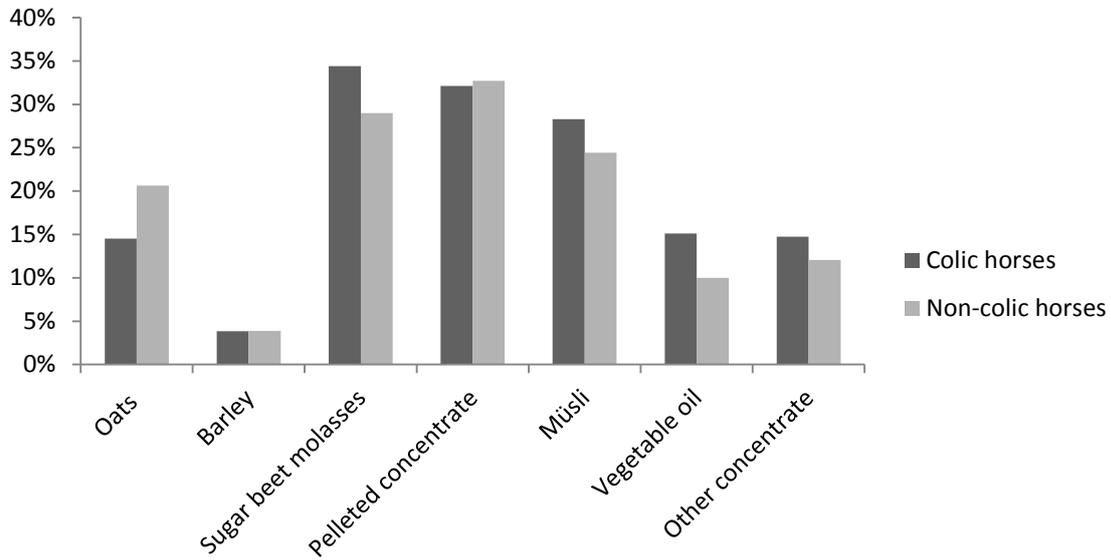


Figure 5. Distribution of type of concentrate fed to horses in the study.

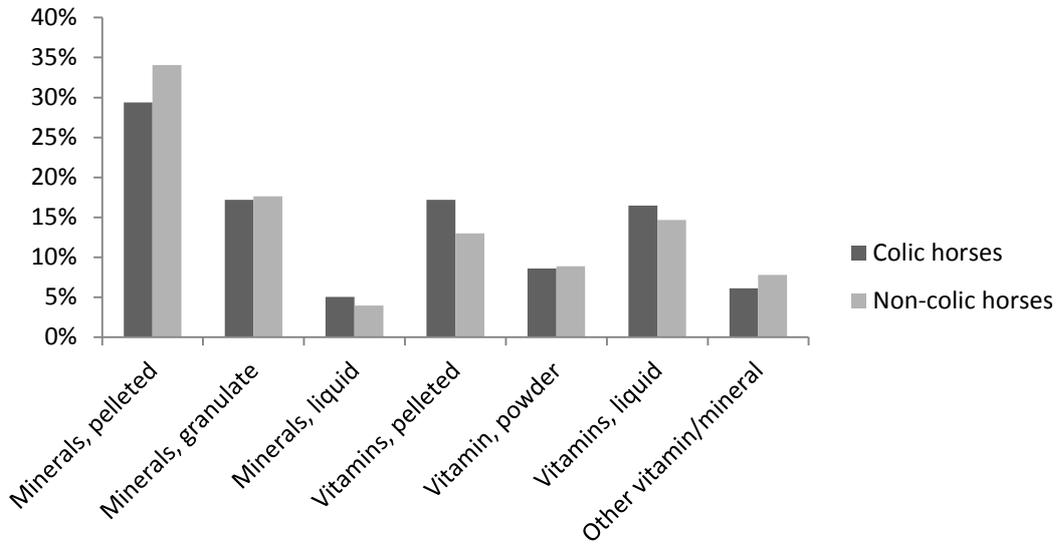


Figure 6. Distribution of type of feed supplements fed to horses in the study.

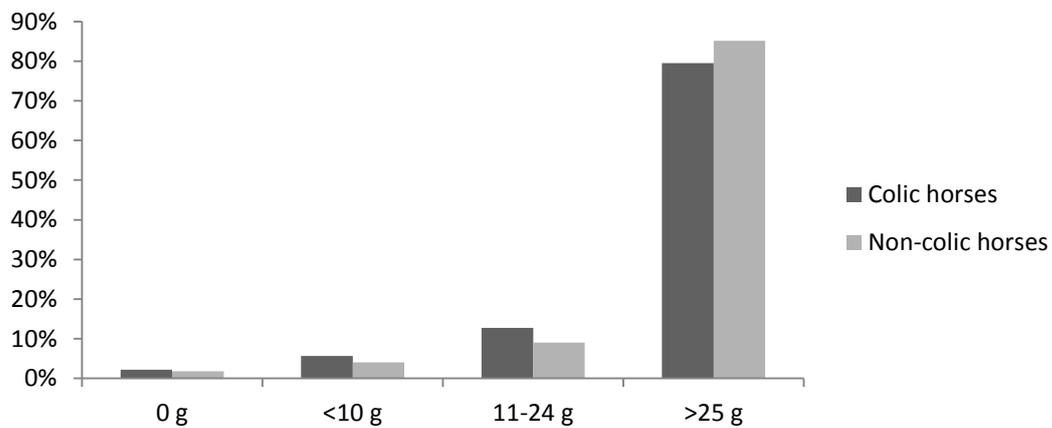


Figure 7. Distribution of daily salt intake of the horses in the study.

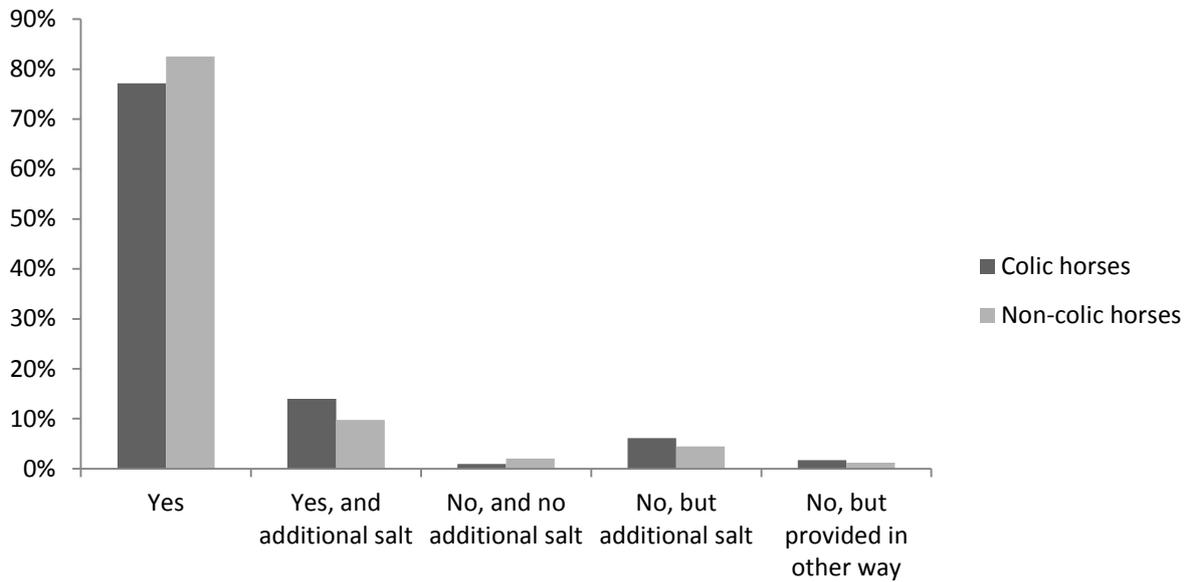


Figure 8. Distribution of access to salt by the horses in the study.

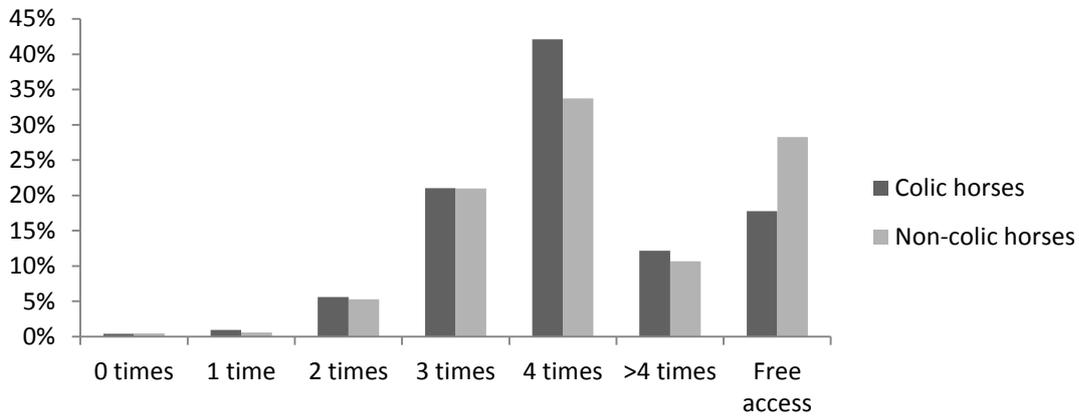


Figure 9. Distribution of number of roughage meals fed daily to the horses in the study.

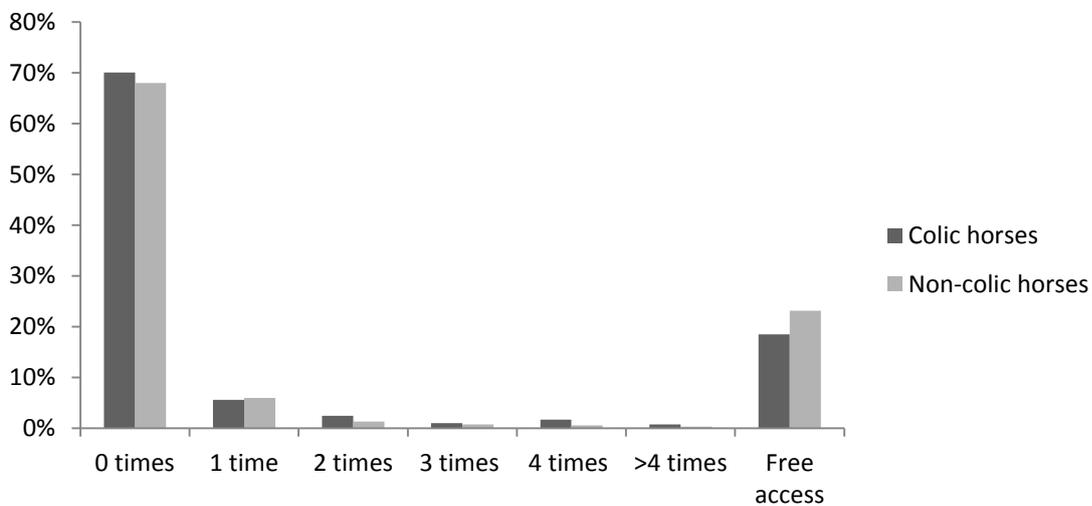


Figure 10. Distribution of number of feedings per day where straw was the only roughage offered to the horses in the study.

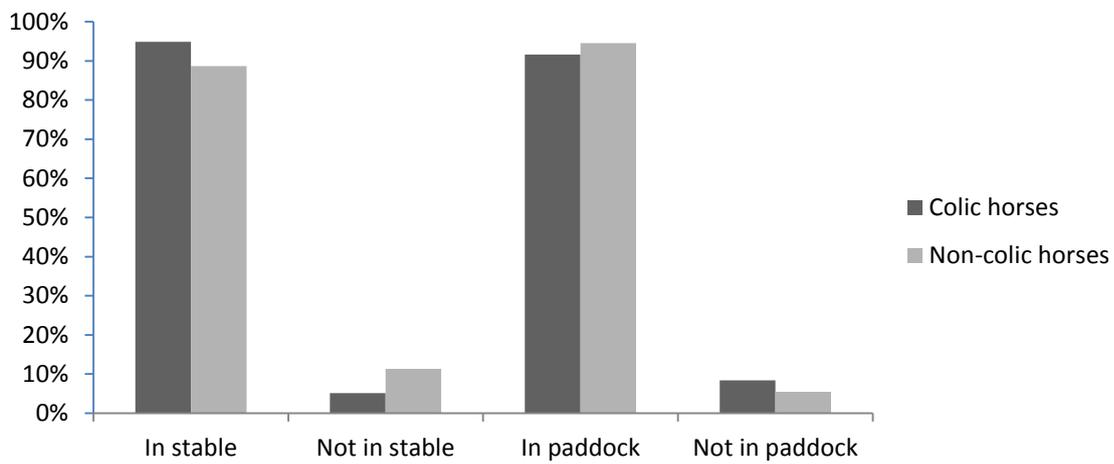


Figure 11. Distribution of where the horses in the study were fed roughage (stable and/or paddock).

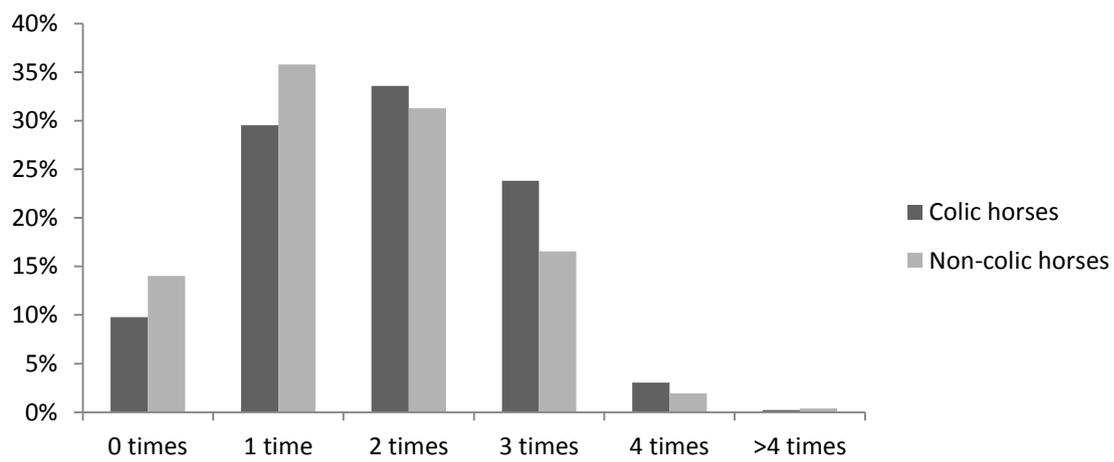


Figure 12. Distribution of number of concentrate meals fed daily to the horses in the study.

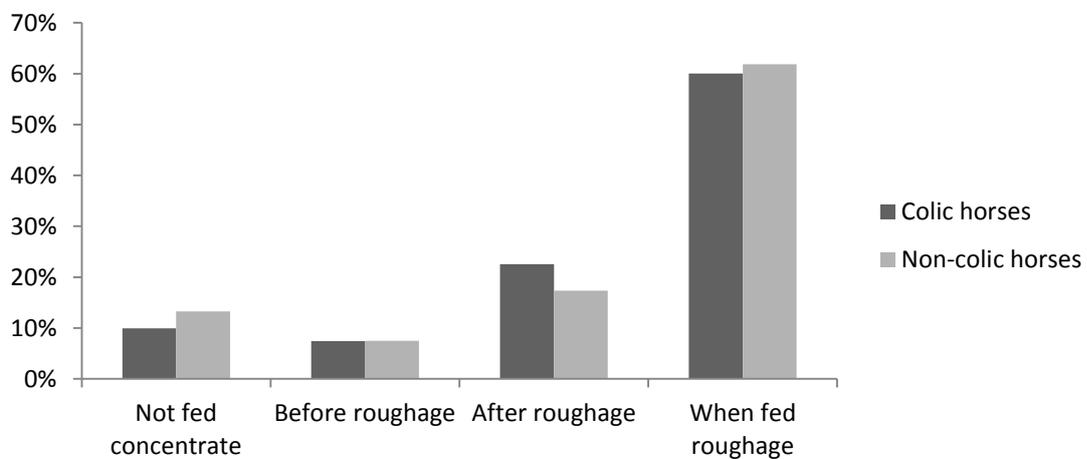


Figure 13. Distribution of when the horses in the study were fed concentrates (in relation to roughage feedings).

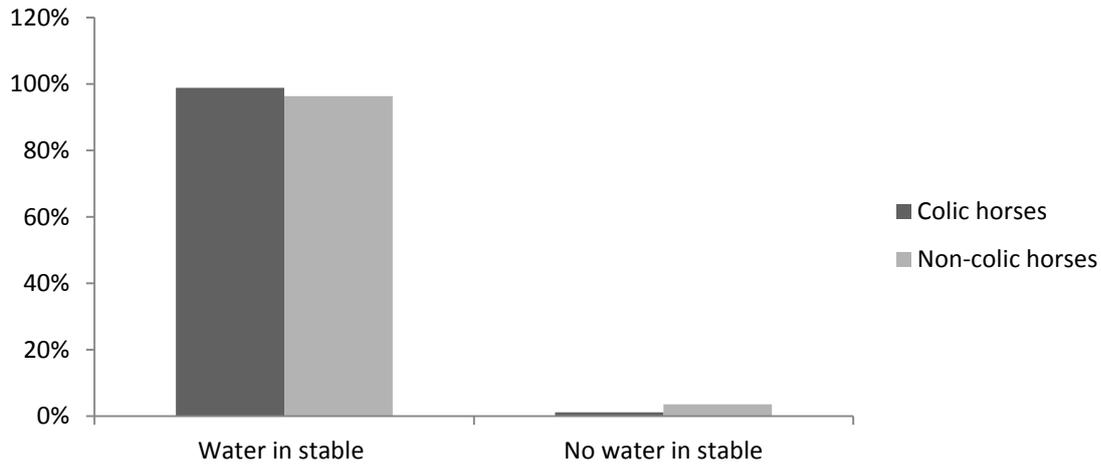


Figure 14. Distribution of access to water in the stable for the horses in the study.

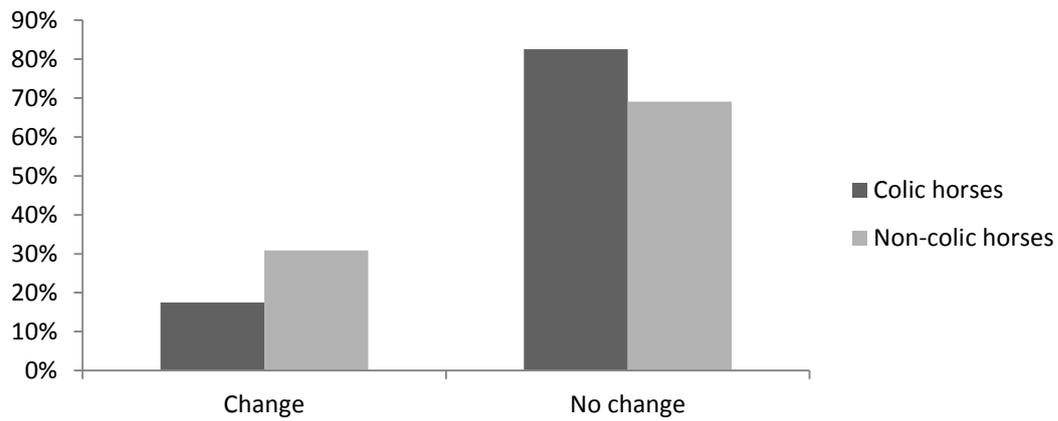


Figure 15. Distribution of changes in type and/or batch of roughage among the horses in the study.

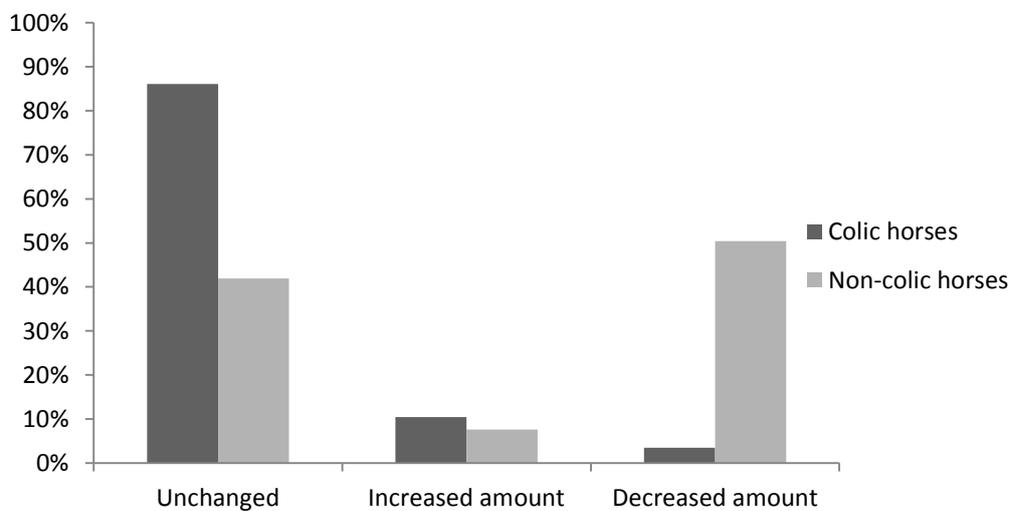


Figure 16. Distribution of changes in the amount of roughage fed among the horses in the study.

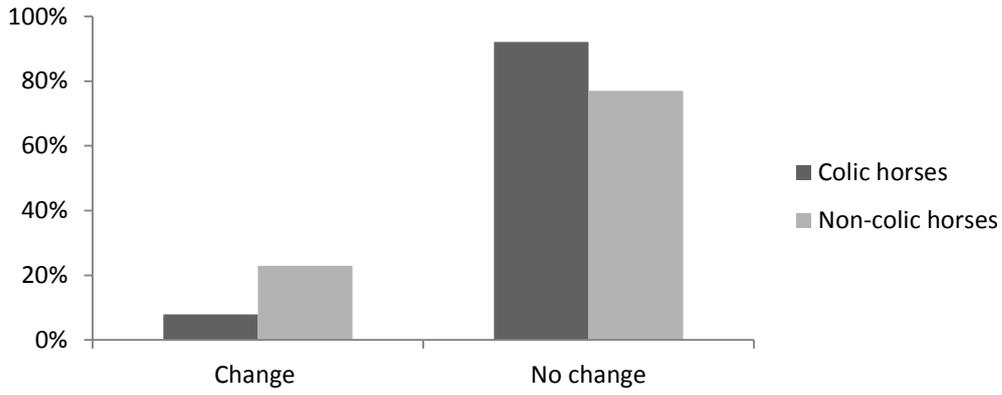


Figure 17. Distribution of changed type and/or batch of concentrate among the horses in the study.

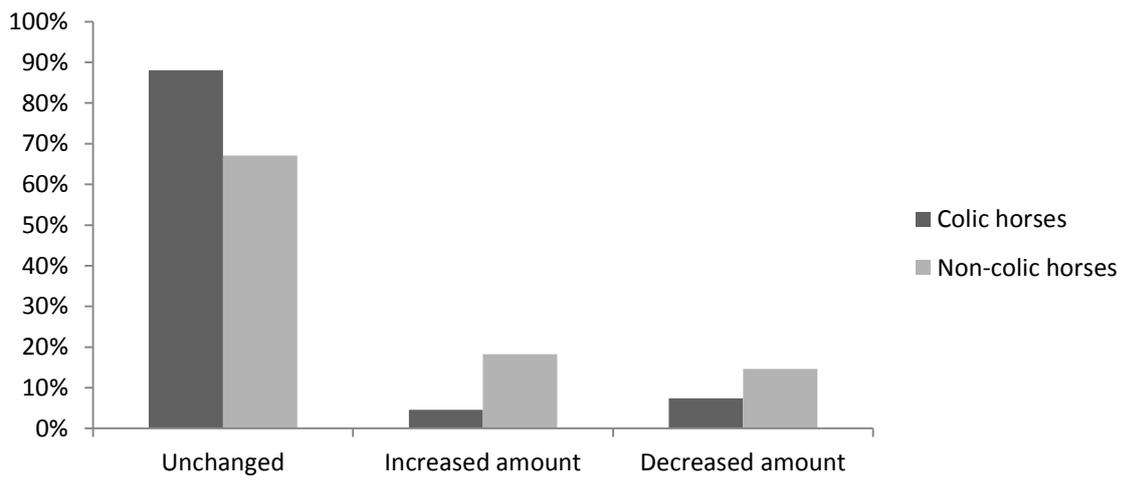


Figure 18. Distribution of changes in the amount of concentrates fed among the horses in the study.

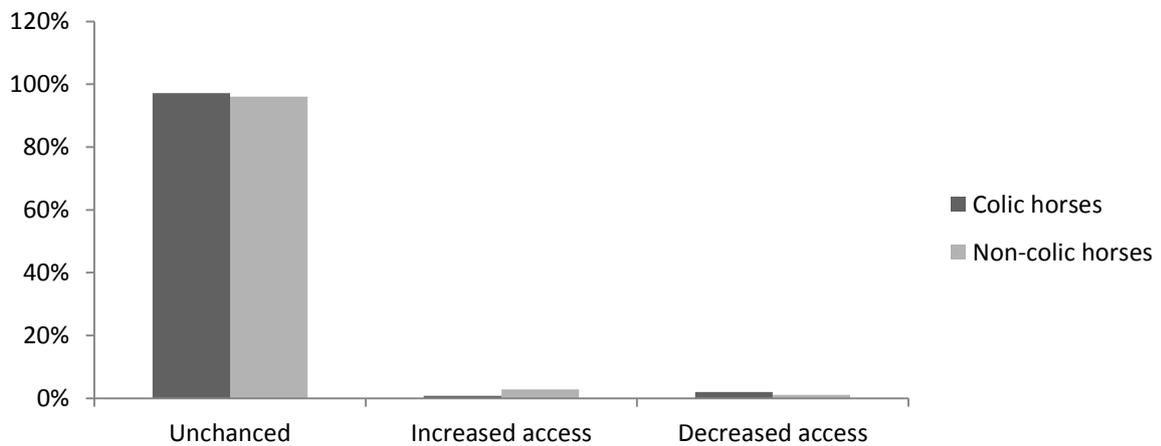


Figure 19. Distribution of changes in access to water in the stable among the horses in the study.

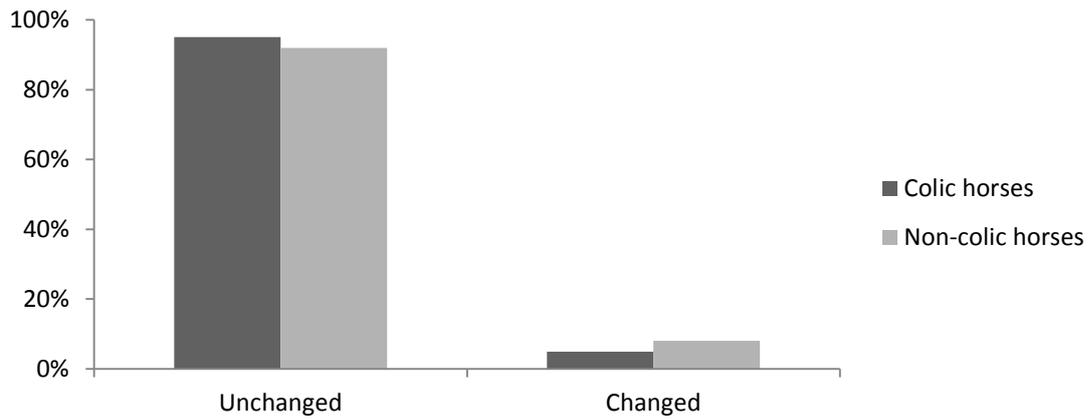


Figure 20. Distribution of changes in type of water source in the stable among the horses in the study.

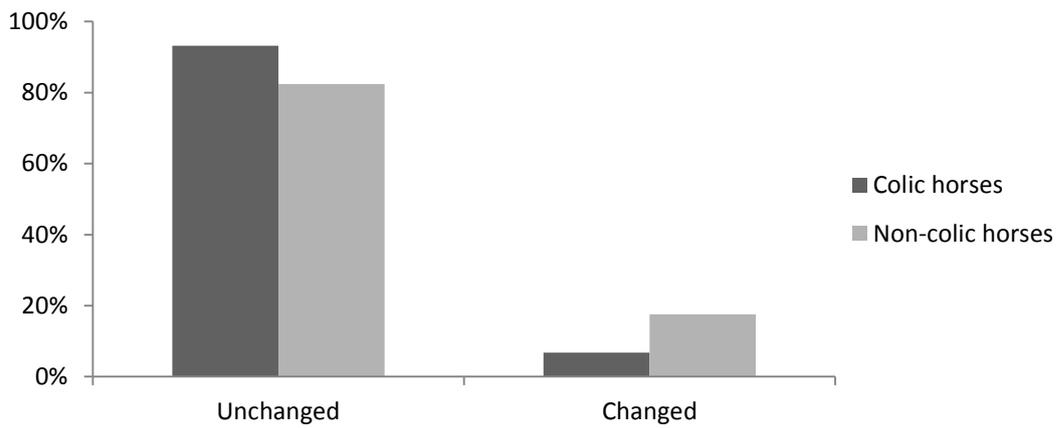


Figure 21. Distribution of changes in type of water source in the paddock among the horses in the study.

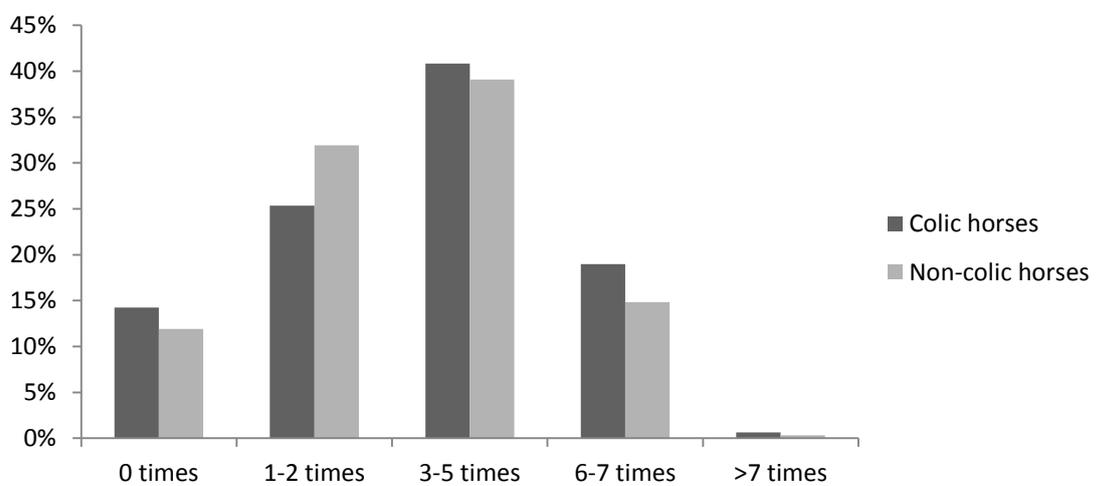


Figure 22. Distribution of number of trainings per week among the horses in the study.

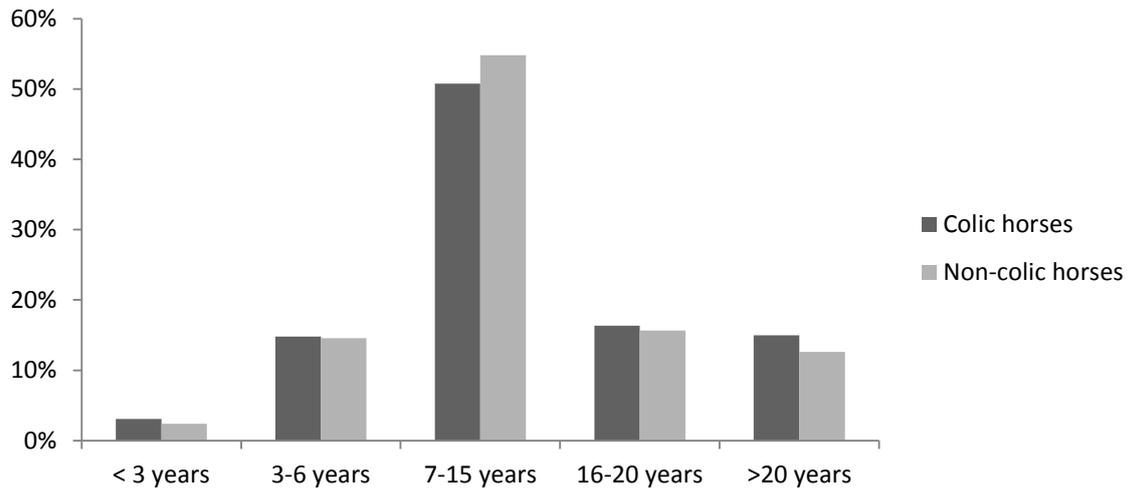


Figure 23. Distribution of age among the horses in the study.

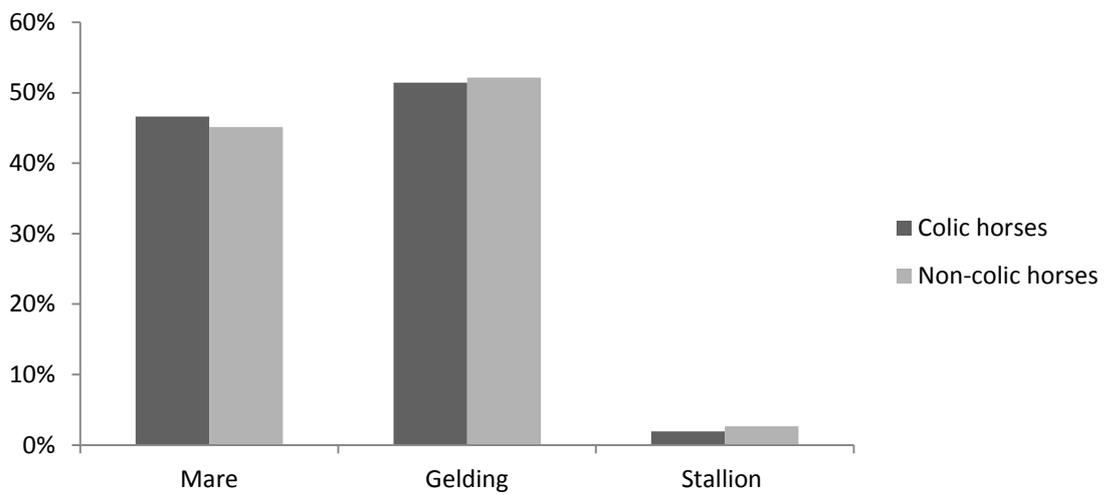


Figure 24. Distribution of gender among the horses in the study.

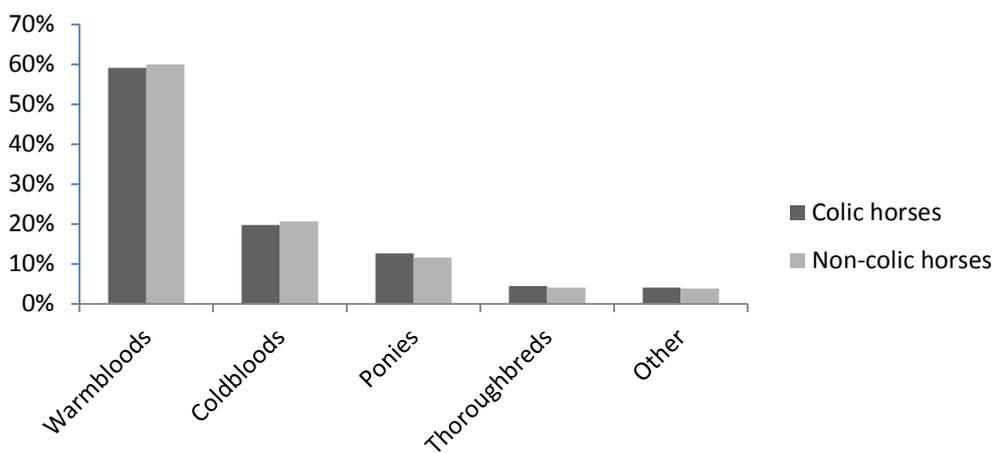


Figure 25. Distribution of breed type among the horses in the study.

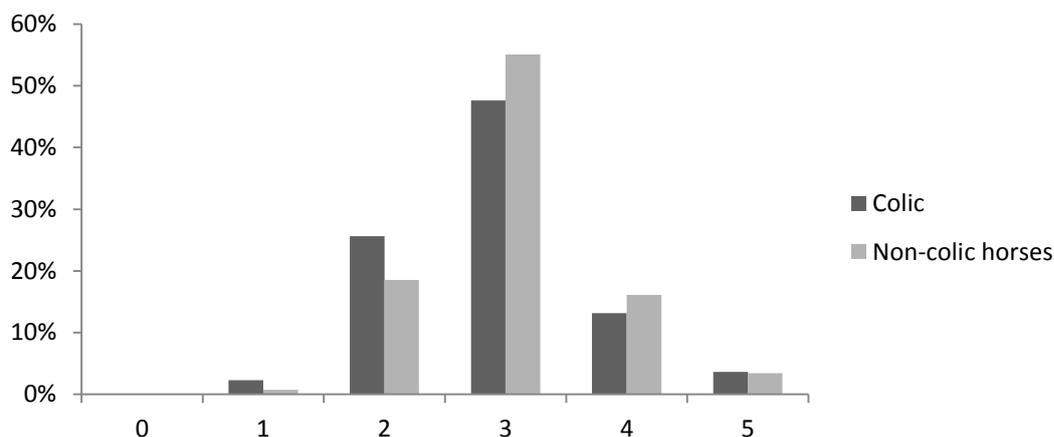


Figure 26. Distribution of body condition score among the horses in the study.

9.2. Results from univariate analysis

A total of 18 class variables and 10 continuous variables had a univariate significance level of $p < 0.005$ and were therefore considered in the multivariate model development. These variables and their significance level are summarized in Table 2 and 3 for continuous and class variables, respectively.

9.2.1. Type and amount of roughage

When investigating the average amount of forage per 100 kg BW and day, an association was detected between decreased presence of colic for every 1 kg increase in amount of forage per 100 kg BW fed daily (Table 2). No association was detected between presence of colic and specific type of roughage fed.

9.2.2. Type and amount of concentrate

When investigating the average amount of concentrate feed per 100 kg BW and day, an association was detected between an increased presence of colic for every 1 kg increase in amount of barley, molassed sugar beet pulp and müsli per 100 kg BW fed daily. Also, an association was detected between a decreased presence of colic for every 1 kg increase in amount of oats and pelleted concentrate per 100 kg BW fed daily (Table 2). No association was detected between presence of colic and type of concentrate fed.

9.2.3. Type and amount of feed supplements

When investigating the average amount of supplement feed per 100 kg BW and day, an association was detected between a decreased presence of colic for every 1 ml increase in amount of liquid minerals per 100 kg BW fed daily (Table 2). Neither an increased amount of liquid vitamins, pelleted vitamins nor pelleted minerals increased or decreased the presence of colic (Table 2). The daily intake of salt was shown to be associated with presence of colic ($p = 0.006$) as horses having a daily salt intake of < 25 g had higher presence of colic relative to horses having a daily intake > 25 g (Table 3). If the horse had access to a salt block or not was shown to be associated with presence of colic ($p = 0.005$), and an increased presence of colic was shown for horses fed salt mixed with concentrates in addition to salt block or as the only source (Table 3).

9.2.4. Feeding routines for roughage

Number of roughage meals fed daily was shown to be associated with presence of colic ($p < .0001$), and horses having free access to roughage were shown to have a decreased presence of colic, while the other number of feedings were shown to have an increased presence of colic (Table 3). Number of meals per day with straw as the only roughage offered was shown to be associated with presence of colic ($p = 0.004$) and horses fed straw as the only roughage offered 2, 3, 4 and >4 times per day were shown to have increased presence of colic, whereas horses offered straw as the only roughage 1 time or had free access to straw were shown to have a decreased presence of colic (Table 3). If the horse was fed roughage or not in the stable ($p < .0001$) and in the paddock ($p = 0.009$) was associated with presence of colic. Horses that were not fed roughage in the stable had increased presence of colic whereas horses not fed roughage in the paddock had decreased presence of colic (Table 3). However, no association was shown between presence of colic and how (on ground, in hay net, in feed rack, on feeding table) the horses were fed roughage in the stable and in the paddock.

9.2.5. Feeding routines for concentrates

The number of concentrate meals fed daily was associated with presence of colic ($p = 0.0001$), and the presence of colic increased with the number of feedings with concentrate fed daily (Table 3). The timing for feeding concentrate (in relation to feeding roughage) was associated with presence of colic ($p = 0.005$). All the different timings for feeding concentrate were associated with an increased presence of colic, where horses fed concentrate after being fed roughage had ~~an~~ the highest presence of colic relative to horses not fed concentrate at all (Table 3). However, meals when feeding only concentrates were fed without any roughage was not shown to be associated with presence of colic.

9.2.6. Watering strategies

If the horse was watered or not in the stable ($p = 0.006$) was associated with presence of colic, where horses not watered in the stable had an increased presence of colic relative to horses watered in the stable (Table 3). However, no association were shown between colic and if the horses were watered in the paddock or not, the type of water source provided in the stable and in the paddock and the type of automatic waterer used in the stable and in the paddock.

9.2.7. Change in type and amount of roughage

A change in type or batch of roughage during the last six months was associated with an increased presence of colic ($p < .0001$) (Table 3). However, specific changes made in roughage feeding during this period (change from one batch and/or type of roughage to another batch and/or type of roughage) was not shown to be associated with presence of colic. A change in the amount of roughage, both increased and decreased, was associated with decreased presence of colic relative to not having a change in amount of roughage (Table 3).

9.2.8. Change in type and amount of concentrate

A change in the amount of concentrates fed was shown to be associated with an increased presence of colic ($p < .0001$) (Table 3). However, a specific type of change in concentrate (change from one batch and/or type of concentrate to another batch and/or type of concentrate) was not shown to be associated with presence of colic. A change in amount of

concentrate, both increased and decreased, was shown to be associated with decreased presence of colic ($p<.0001$) relative to not having a change in amount of concentrate (Table 3).

9.2.9. Changed access to water

A changed access to water in the stable, both decreased and increased, was associated with decreased presence of colic ($p=0.01$) (Table 3). No association between presence of colic and changed access of water in the paddock was found ($p=0.88$).

9.2.10. Changed type of water source

A change in type of water source in the stable was shown to be associated with decreased presence of colic ($p=0.02$). No association was detected between presence of colic and changes in specific types of water sources in the stable ($p=0.95$) (Table 3). A change in type of water source in the paddock was associated with increased presence of colic ($p<.0001$) (Table 3). However, no association was detected between presence of colic and changes in specific types of water sources in the paddock ($p=0.70$).

9.2.11. Number of trainings per week

Number of trainings per week was found to be associated with presence of colic, where horses that were trained >6 times per week were associated with increased presence of colic, and horses that were trained <3 times per week, were associated with decreased presence of colic, both relative to horses not trained at all ($p=0.02$) (Table 3). However, neither discipline nor training intensity of the horse was associated with presence of colic.

9.2.12. Individual factors

A body condition score <3 and >4 was found to be associated with an increased presence of colic ($p<.0001$) (Table 3). The horses with a BCS >3 were commonly fed a number of concentrates and supplement feeds. No association between presence of colic and age ($p=0.38$), gender ($p=0.57$) or breed type ($p=0.93$) were shown.

Table 2. Continuous variables associated with presence of colic in the univariate analysis

Amount of feed/ 100 kg BW	Colic horses (%)	Mean	SD	Non-colic horses (%)	Mean	SD	OR	95% CI		p-value
								Lower bound	Upper bound	
Forage (kg/day)	469 (16.8)	2.19	0.78	2323 (83.2)	2.28	0.76	0.8	0.73	0.96	0.01
Oats (kg/day)	77 (11.9)	0.13	0.09	571 (88.1)	0.14	0.17	0.3	0.06	1.08	<.0001
Barley (kg/day)	20 (16.1)	0.15	0.14	104 (83.9)	0.13	0.12	1.9	0.16	22.022	0.006
Molassed sugar beet pulp (kg/day)	172 (18.4)	0.05	0.04	764 (81.6)	0.05	0.05	18.6	1.36	258.11	<.0001
Pelleted concentrate (kg/day)	161 (15.8)	0.14	0.12	860 (84.2)	0.14	0.14	0.9	0.35	2.29	0.02
Müsli (kg/day)	142 (18.1)	0.15	0.20	642 (81.9)	0.12	0.12	3.6	1.49	9.25	<.0001
Minerals, pelleted (g/day)	224 (15.9)	21.88	18.74	1186 (84.1)	20.09	14.74	1.0	0.10	1.01	<.0001
Vitamins, pelleted (g/day)	42 (19.0)	15.68	8.63	179 (81.0)	16.21	13.74	1.0	0.99	1.03	0.0007
Minerals, liquid (ml/day)	16 (19.8)	6.43	6.40	65 (80.2)	12.05	25.72	0.9	0.94	1.03	<.0001
Vitamins, liquid (ml/day)	54 (17.3)	7.17	10.02	258 (82.7)	5.08	5.20	1.0	1.00	1.06	<.0001

Table 3. Class variables associated with presence of colic in univariate analysis

	Number of horses		OR	95% CI		p-value
	Colic horses (%)	Non-colic horses (%)		Lower bound	Upper bound	
Body condition score						<.0001
0	0 (0)	1 (0.04)	-	-	-	
1	12 (2.5)	17 (0.7)	4.3	2.03	9.08	
2	134 (27.6)	508 (19.5)	1.6	1.27	2.02	
3	252 (51.9)	1530 (58.9)	Ref.			
4	69 (14.2)	447 (17.2)	0.9	0.70	1.25	
5	19 (3.9)	95 (3.7)	1.2	0.73	2.02	
Number of trainings per week						0.02
0 times	69 (14.2)	356 (13.8)	Ref.			
1-2 times	123 (25.3)	821 (31.9)	0.8	0.56	1.06	
3-5 times	198 (40.8)	1005 (39.0)	1.0	0.75	1.37	
6-7 times	92 (18.9)	381 (14.8)	1.2	0.88	1.76	
> 7 times	3 (0.6)	8 (0.3)	1.9	0.50	7.47	
Number of roughage meals per day						<.0001
0	2 (0.4)	12 (0.4)	Ref.			
1	5 (0.9)	16 (0.6)	1.9	0.31	11.37	
2	29 (5.6)	145 (5.3)	1.2	0.26	5.65	
3	109 (21.0)	573 (20.9)	1.1	0.25	5.17	
4	218 (42.1)	922 (33.7)	1.4	0.32	6.41	
>4	63 (12.2)	292 (10.7)	1.3	0.28	5.93	
Free access	92 (17.8)	772 (28.3)	0.7	0.16	3.25	
Number of straw feedings						0.004
0	288 (70.1)	1489 (68.0)	Ref.			
1	23 (5.6)	130 (5.9)	0.9	0.58	1.45	
2	10 (2.4)	29 (1.3)	1.8	0.86	3.70	
3	4 (0.9)	16 (0.7)	1.3	0.43	3.89	
4	7 (1.7)	12 (0.5)	3.0	1.18	7.73	
>4	3 (0.7)	7 (0.3)	2.2	0.57	8.62	
Free access	76 (18.5)	506 (23.1)	0.8	0.59	1.02	
Roughage fed in the stable						<.0001
Yes	496 (91.6)	2430 (88.6)	Ref.			
No	27 (5.2)	311 (11.3)	2.4	1.57	3.52	

Table 3 (Continued)	Number of horses		OR	95% CI		p-value
	Colic horses (%)	Non-colic horses (%)		Lower bound	Upper bound	
Roughage fed in the paddock						0.009
Yes	479 (91.6)	2592 (94.5)	Ref.			
No	44 (8.4)	149 (5.4)	0.6	0.44	0.88	
Number of concentrate feedings/ day						0.0001
0	48 (9.8)	355 (14.1)	Ref.			
1	145 (29.3)	891 (35.5)	1.2	0.84	1.68	
2	165 (33.6)	784 (31.2)	1.5	1.09	2.18	
3	117 (24.0)	422 (16.8)	2.1	1.43	2.97	
4	15 (3.1)	49 (2.0)	2.3	1.18	4.35	
>4	1 (0.2)	10 (0.4)	0.7	0.09	5.91	
Feeding of concentrate						0.02
Not fed concentrate	48 (9.9)	326 (13.3)	Ref.			
Before fed roughage	36 (7.5)	184 (7.5)	1.3	0.83	2.12	
After fed roughage	109 (22.6)	426 (17.4)	1.7	1.20	2.51	
Together with roughage	290 (60.0)	1518 (61.9)	1.3	0.93	1.80	
Free access to saltblock						0.005
Yes	402 (77.2)	2242 (82.5)	Ref.			
Yes, and salt in concentrate	73 (14.0)	266 (9.7)	1.5	1.16	2.03	
No saltblock or salt	5 (1.0)	55 (2.0)	0.5	0.20	1.27	
No, but salt in concentrate	32 (6.1)	121 (4.5)	1.5	0.98	2.21	
No, provided salt in other way	9 (1.7)	32 (1.2)	1.6	0.74	3.31	
Daily intake of salt						0.03
0 g	10 (2.2)	43 (1.8)	1.3	0.65	2.60	
< 10 g	26 (5.6)	98 (4.1)	1.5	0.95	2.31	
11-24 g	59 (12.7)	218 (9.0)	1.5	1.11	2.05	
>25 g*	368 (79.5)	2051 (85.1)	Ref.			
Access to water in the stable						0.006
Yes	517 (98.8)	2643 (96.4)	Ref.			
No	6 (1.2)	98 (3.6)	3.2	1.39	7.31	
Change of roughage (type, batch)						<.0001
No	411 (82.5)	1764 (69.1)	Ref.			
Yes	87 (17.5)	788 (30.9)	2.5	0.37	2.61	
Changed amount of roughage						<.0001
Unchanged	421 (86.1)	1234 (50.4)	Ref.			
Increased	51 (10.4)	187 (7.6)	0.7	0.48	0.92	
Decreased	17 (3.5)	1026 (41.9)	0.03	0.02	0.06	
Change of concentrate (type, batch)						<.0001
No	39 (7.9)	604 (22.9)	Ref.			
Yes	453 (92.1)	2029 (77.1)	3.0	0.21	3.41	

*= free access to saltblock

Table 3 (Continued)	Number of horses		OR	95% CI		p-value
	Colic horses (%)	Non-colic horses (%)		Lower bound	Upper bound	
Changed amount of concentrate						<.0001
Increased	34 (7.4)	347 (14.7)	0.2	0.12	0.30	
Decreased	21 (4.6)	423 (17.6)	0.4	0.27	0.55	
Unchanged	406 (88.1)	1586 (67.3)	Ref.			
Change in water access (stable)						0.01
Increased	4 (0.8)	75 (2.8)	0.2	0.10	0.75	
Decreased	10 (2.0)	29 (1.1)	1.8	0.86	3.66	
Unchanged	493 (97.2)	2536 (96.1)	Ref.			
Change in water source (stable)						0.02
No	468 (95.2)	2333 (92.0)	Ref.			
Yes	24 (4.8)	204 (8.0)	0.6	0.38	0.91	
Change in water source (paddock)						0.005
No	411 (93.2)	1944 (82.4)	Ref.			
Yes	30 (6.8)	414 (17.6)	2.3	0.23	3.50	

9.3. Results from multivariate analysis

Two final multivariate models were created, by using stepwise selection, for both the class variables and the continuous variables found to be associated with presence of colic (table 2 and 3). Seven class variables were included in the first final model (Table 4) and three continuous variables were included in the second final model (Table 5).

9.3.1. Change in type of water source in the paddock

A change in water source in the paddock were included in the final model with a significance level of $p=0.002$. The results showed that horses having an change of water source in the paddock were more than two times more likely to be colic horses ($p=0.002$) relative to horses not having a change of water source in the paddock (Table 4).

9.3.2. Number of concentrate meals fed daily

The number of concentrate feedings per day were included in the final model with a significance level of $p=0.04$. The results showed that horses fed concentrate three times daily were 2.1 times more likely to be colic horses ($p=0.005$) relative to horses not receiving any concentrate (Table 4) and these horses were also shown to be fed the highest amount of starch rich concentrate (oats, pelleted concentrate and müsli) per 100 kg BW and day.

9.3.3. Daily intake of salt

The daily intake of salt was included in the final model with a significance level of $p=0.008$. The results showed that horses reported having an intake of 11-24 grams daily were more than two times more likely to be colic horses relative to horses having an daily salt intake of >25 g ($p=0.04$) (Table 4).

9.3.4. Change in type and amount of roughage

Both change in type and amount of roughage were included in the final model with significance levels of $p=0.003$ and $p<.0001$, respectively. The results showed that horses where roughage type was changed were more than two times more likely to be colic horses ($p<.0001$) relative to horses where no change in roughage type had been present (Table 4). The results also showed that horses reported having an increased or a decreased amount of roughage were less likely to be colic horses ($p<.0001$) relative to horses where the amount of roughage was not changed (Table 4).

9.3.5. Change in type and amount of concentrate

Both change in type and amount of concentrate were included in the final model with a significance level of $p<.001$ and $p<.0001$, respectively. The results showed that horses reported to have had a changed type of concentrate were more than three times more likely to be colic horses ($p<.0001$) relative to horses not having any change in type of concentrate. The results also showed that horses reported to have had an increased amount of concentrate were less likely to be colic horses ($p=0.0003$) relative to horses not having an increased amount of concentrate (Table 4).

9.3.6. Amount feed/ 100 kg BW fed daily

The amount of forage (hay, haylage and silage), müsli and liquid vitamins per 100 kg BW fed daily were the only three feeds included in the final model (Table 5). Colic horses had a lower average consumption of forage (2.19 ± 0.78 kg/100 kg BW/ day) than non-colic horses (2.28 ± 0.76 kg/100 kg BW/ day). For each 1 kg increase/ 100 kg BW in the amount of forage consumed, the risk of colic decreased with 20% ($p=0.007$). Colic horses had a higher average consumption of müsli (0.15 ± 0.20 kg/100 kg BW/ day) than non-colic horses (0.12 ± 0.12 kg/100 kg BW/ day). For each 1 kg increase/100 kg BW in the amount of müsli consumed, the risk of colic increased over four times ($p=0.005$). Colic horses had a higher average consumption of liquid vitamins (7.17 ± 10.92 ml/100 kg BW/ day) than non-colic horses (5.08 ± 5.20 ml/100 kg BW/ day). For each 1 ml increase/ 100 kg BW in the amount of liquid vitamins consumed, the risk of colic increased by 3 % ($p=0.03$).

Table 4. Class variables associated with presence of colic in multivariate analysis

Model 1	Number of horses		OR	95% CI		p-value
	Colic horses (%)	Non-colic horses (%)		Lower bound	Upper bound	
Number of concentrate feedings/ day						
0	48 (9.8)	355 (14.1)	Ref.			
1	145 (29.3)	891 (35.5)	1.2	0.84	1.68	0.40
2	165 (33.6)	784 (31.2)	1.5	1.09	2.18	0.30
3	117 (24.0)	422 (16.8)	2.1	1.43	2.97	0.005
4	15 (3.1)	49 (2.0)	2.3	1.18	4.35	0.10
>4	1 (0.2)	10 (0.4)	0.7	0.09	5.91	1.00
Daily intake of salt						
0 g	10 (2.2)	43 (1.8)	2.2	0.71	6.76	0.3
< 10 g	26 (5.6)	98 (4.1)	0.6	0.25	1.72	0.06
11-24 g	59 (12.7)	218 (9.0)	2.5	1.37	4.41	0.04
>25 g*	368 (79.5)	2051 (85.1)	Ref.			
Change of roughage (type, batch)						
No	411 (82.5)	1764 (69.1)	Ref.			
Yes	87 (17.5)	788 (30.9)	2.1	1.65	2.70	<.0001
Changed amount of roughage						
Unchanged	17 (3.5)	1234 (50.4)	Ref.			
Decreased	421 (86.1)	1026 (41.9)	0.03	0.02	0.06	<.0001
Increased	51 (10.4)	187 (7.6)	0.7	0.48	0.92	<.0001
Change of concentrate (type, batch)						
No	39 (7.9)	604 (22.9)	Ref.			
Yes	453 (92.1)	2029 (77.1)	3.5	2.46	4.86	<.0001
Changed amount of concentrate						
Unchanged	406 (88.1)	1586 (67.3)	Ref.			
Decreased	21 (4.6)	423 (17.6)	0.3	0.17	0.73	0.70
Increased	34 (7.4)	347 (14.7)	0.2	0.08	0.31	0.0003
Change in water source (paddock)						
No	411 (93.2)	1944 (82.4)	Ref.			
Yes	30 (6.8)	414 (17.6)	2.9	1.98	4.29	0.002

*= free access to salt block

Table 5. Continuous variables associated with presence of colic in the multivariate analysis

Model 2	Colic horses (%)	Mean	SD	Non-colic horses (%)	Mean	SD	OR	95% CI		p-value
								Lower bound	Upper bound	
Forage (kg/ 100 kg BW/ day)	469 (16.8)	2.19	0.78	2323 (83.2)	2.28	0.76	0.8	0.73	0.96	0.007
Müsli (kg/ 100 kg BW/ day)	142 (18.1)	0.15	0.20	642 (81.9)	0.12	0.12	4.4	1.58	12.4	0.005
Vitamins, liquid (ml/100 kg BW/ day)	54 (17.3)	7.17	10.02	258 (82.7)	5.08	5.20	1.03	1.00	1.06	0.03

10. Discussion

10.1. Change in type and amount of feed

A change in type and/ or batch of roughage was shown to be associated with colic, which was expected based on similar findings from other studies (Cohen *et al.*, 1999; Cohen *et al.*, 1995; Cohen and Peloso, 1996; Cohen and Gibbs, 1999; Tinker *et al.*, 1997). Horses reported to have had a changed type and/ or batch of roughage were more than two times more likely to be colic horses ($p < .0001$; OR=2.1; 95% CI=1.65-2.70) relative to horses not having a change in type of roughage in the current study (Table 3). The most common change of roughage among the horses in this study was a change in type and/or batch of haylage or hay. Unfortunately, a change of batch and/or type of haylage is not well documented in previous studies, which might be due to the fact that haylage is not used in the same extent in *e.g.* the USA as for the horse population in this study. A change of type and/or batch of hay is on the other hand well documented in the previously mentioned studies, which have reported that a change of batch and/or type of hay (Cohen *et al.*, 1999; Hillyer *et al.*, 2002; Hudson *et al.*, 2001; Tinker *et al.*, 1997) increase the risk of colic which could explain the results in this study. Previous studies have shown that horses having a change in the amount of hay were at higher risk of colic relative to horses not having a change in the amount of hay (Tinker *et al.*, 1997; Hudson *et al.*, 2001). In the present study, the findings show that horses having a change in the amount of roughage, decrease or increase, were less likely to be colic horses relative to horses not having a change in the amount of roughage. However, the previous studies investigated a change in the amount of hay and not a change in total roughage amount (including hay, haylage, silage, straw and lucerne) as in this study. Therefore, in order to compare the results with previous findings it would be necessary to know the specific type of roughage that was changed and also the specific amount increased or decreased to be able to draw any further conclusions.

The results from this study also showed that horses reported as having a changed type and/or batch of concentrate were more than three times likely to be colic horses ($p < .0001$; OR=3,5; 95% CI=2.46-4.86) relative to horses not having a change in type and/or batch of concentrate. This result is supported by the findings from previous studies where a recent change in type of concentrate or grain has been shown to increase the risk of colic (Hudson *et al.*, 2001; Hillyer *et al.*, 2002). Besides being supported by similar results from previous studies, the results from the present study are in agreement with the negative effects of changes and alterations of environment and function of the horse GIT from a biological perspective (de Fombelle *et al.*, 2001). A change in specific types of concentrates was not shown to be associated with colic in this study, probably because there were too many different combinations of changes made among the horses, resulting in very few horses having the exact same change. However, when looking into the different combinations of changes it was possible to see a pattern, that the

majority of colic horses had had a change from one starch rich feed to another. However, a change from one feed to another in itself does not mean that the risk of colic increases. To know why a change in type and/or batch increases the risk of colic also requires further information about the change and feeds. In the end, the effect of the change depends on whether the microorganisms in the intestine have adapted to the new environment or not caused by the change (Bailey *et al.*, 2003; de Fombelle *et al.*, 2003; Berg *et al.*, 2005; Varloud *et al.* 2007). This would require information about how long the period for a certain change was and also the amount changed from one day to another. However, this information was not possible to retrieve from the questionnaire responses.

In this study, horses having an increased amount of concentrate were shown to be less likely to have colic relative to horses not having any change in concentrate feeding. These findings are contradictory to the findings reported from previous studies (Hudson *et al.*, 2001; Hillyer *et al.*, 2002) where horses having a changed amount of concentrate were more likely to be colic horses. However, it might be due to the fact that there was a higher proportion of non-colic horses in the present study reported having an increased amount of concentrate in the diet, relative to the proportion of colic horses. Also, it could be that the change in amount of feed was so small that it did not have any biological relevance and therefore no influence on the risk of colic relative to the changes in amounts made in previous studies. Unfortunately, no information regarding the specific amount of concentrate before and after the change was obtained from the questionnaire, which would be necessary in order to make any further conclusions of this result.

10.2. Amount of feed per 100 kg BW fed daily

The amount of feed per 100 kg body weight (BW) and day was shown to be associated with presence of colic in this study, as colic horses had a lower average consumption of forage (2.19 ± 0.78 kg/100 kg BW/ day) than non-colic horses (2.28 ± 0.76 kg/100 kg BW/ day) and the results showed that for each 1 kg increase in the amount of forage consumed per 100 kg BW, the risk for colic decreased with 20 percent. A lower daily average consumption of forage for colic horses relative to non-colic horses has also been shown in other studies (Tinker *et al.*, 1997; Kaya *et al.*, 2009), indicating that a higher inclusion of forage in diet promotes a good GIT health and reduces the risk of colic. The previous studies investigating the association between colic and the amount of forage fed daily did not consider the BW of the horse although the horses included in these studies have been horses of various breeds with a large variation in body weight. This makes it difficult to draw any conclusion regarding if the average amount of forage in their studies was sufficient to meet the lowest forage requirements. However, in order to make final conclusions regarding the amount of forage fed daily it is important to know the dry matter (DM) content of the forage. In this study, no information regarding the DM content of the forage could be collected and therefore it was not possible to draw conclusions of whether the horses were provided with a sufficient amount of forage or not. The result that every 1 kg increase in amount of forage per 100 kg BW reduced the risk of colic is also supported by the biological effect of forage on the horse's digestive tract. When horses having a diet high in forage or that are fed only forage the microbes produce a higher percentage of acetic acid and a smaller proportion of propionic acid, not causing a decreased pH and therefore preventing the emergence of harmful bacteria (Varloud *et al.*, 2004). Also, a diet with a high inclusion of forage, which has a high content of fiber, has a high water holding capacity. The majority of the colic horses in this study were reported to have suffered from impaction colic. As this type of colic can be caused by an insufficient amount of water and a low water content of the feed is possible that these horses

were provided an insufficient amount of water and/or were fed a too low amount of forage in the diet.

Colic horses were also shown to have a higher average consumption of müsli (0.15 ± 0.20 kg/100 kg BW/ day) than non-colic horses (0.12 ± 0.12 kg/100 kg BW/ day), and the results showed that for each 1 kg increase/100 kg BW in the amount of müsli, the risk of colic increased over four times. A higher consumption of müsli have not been showed to be associated with colic in previous studies, but a higher consumption of concentrate and a high amount of other starch-rich feeds (Hudson *et al.*, 2010; Hillyer *et al.*, 2002; Kaya *et al.*, 2009) has been shown to be associated with colic. Grains, particularly oats, barley and wheat, are common ingredients in commercial concentrate mixtures such as müsli-feeds, which means that starch content may be high in these feeds. As mentioned before, starch have a negative effect on the horse GIT and a high inclusion of starch-rich feeds increases the proportion of propionic acid and lactic acid and lower the proportion of acetic acid, resulting in a decrease in pH which can result in digestive disturbances like colic (Shiraz-Beechey *et al.*, 2008), which support the findings in this study that an increased amount of müsli increases the presence of colic.

The amount of liquid vitamins was also shown to be associated with a higher presence of colic. Colic horses had a higher average consumption of liquid vitamins (7.17 ± 10.92 ml/100 kg BW/ day) than non-colic horses (5.08 ± 5.20 ml/100 kg BW/ day). For each 1 ml increase/100 kg BW in the amount of liquid vitamins consumed, the risk of colic increased by 3 percent. Liquid vitamins have not been reported as a risk factor for colic previously. Why liquid vitamins were associated with a higher presence of colic in this study is not clear. It might be that liquid vitamins is a common denominator for the colic horses in this study and was therefore included in the final model. However, it would be interesting to investigate the impact of liquid vitamins further to see if there is any biological explanation for the increased presence of colic.

10.3. Feeding routines for concentrates and roughage

The number of daily feedings with concentrates was shown to be associated with presence of colic (Table 3) and was also included in the final model as one of the main factors affecting the presence of colic (Table 4. In this study, horses fed concentrates three times daily were 2.1 times more likely to suffer from colic relative to horses not receiving any concentrate. The result from this study supports the results from previous studies which showed that <3 concentrate feedings per day increased the risk of colic relative to not receiving any concentrate (Tinker *et al.*, 1997; Hudson *et al.*, 2001). When reviewing the amount of concentrate fed daily for the horses reported to be fed concentrate three times daily, it showed that these horses were also the ones fed the highest amount of starch rich concentrate (oats, pelleted concentrate and müsli) per 100 kg BW and day. The general feeding recommendations is that if the diet has a high inclusion of starch rich feeds the feed should be divided into smaller proportions throughout the day (Harris and Arkel, 2005). However, when having such a high incorporation of starch in the diet it might not be enough to just divide the ration into smaller portions throughout the day. This could be explained by that if horses have an inclusion of too large amount of starch rich concentrate in the diet it becomes too much for the amylase system in the small intestine to handle. Therefore, it might not be enough to divide the ration and feeding recommendations for the amount of starch (150 g starch/ 100 kg body weight) should therefore be applied.

Roughage feeding strategy was associated with presence of colic in this study. Horses fed roughage in smaller portions throughout the day had higher presence of colic relative to horses having free access to roughage. Because horses are strict herbivores and hindgut fermenters it is important that they receive sufficient amount of roughage. The horse GIT is adapted to a continuous flow of forages containing large amounts of water, soluble proteins and structural carbohydrates (Frape, 2010) which makes a free access of roughage more optimal from a biological point of view. Also, horses having free access to roughage are more likely on a diet based on a high proportion of roughage, or even forage only diet, and at most only a small percentage of concentrate included. Whereas horses provided with smaller proportions of roughage per day, a restricted feeding, are more likely to have a smaller proportion of roughage in the diet. It could also be that these horses are provided a part of their daily ration of roughage when provided one of their concentrate meals (when having a large amount of concentrate included in the diet). The strategy of dividing the amount of roughage into many small portions might not be considered to be equivalent, or enough to promote a healthy GIT of the horse to the same extent as free access of roughage does. For these horses it would be good to either provide the horse with roughage having a lower nutritional content or add straw to increase the amount of roughage in the ration to promote a healthy GIT of the horse. A long time between feedings could also be the explanation for why horses not fed roughage in the stable had a higher presence of colic relative to horses provided roughage in the stable in the current study.

Although straw is commonly used as supplement in feed rations to horses, the number of feedings with only straw as roughage was shown to be associated with an increased presence of colic. Horses having free access to straw and horses that were fed straw only at one meal per day had decreased presence of colic, whereas horses fed straw as the only roughage at more than one up to four meals per day had an increased presence of colic. Some horses cannot be fed free access of forage (hay, haylage or silage) as their consumption would exceed their nutritional needs. Therefore, replacing some of the forage with straw is a commonly used strategy for horse owners to decrease the nutritional value but still provide the horse with a continuous intake of fiber (Frape, 2010). Replacing too many meals per day with straw as the only roughage might not be optional, as straw has a lower nutritional value of protein and energy than forage and can therefore not replace forage to any high extent. Also, due to the lower content of protein and calcium in straw, a lower buffering effect is expected which may have a negative effect on the environment of the GIT (Frape, 2010). However, the number of feedings with straw as the only roughage was not included in the final model, and therefore not considered one of the main factors for presence of colic among the horses in this study.

10.4. Change in type of water source in the paddock

Insufficient or limited access to water has been shown to be associated with an increased risk of colic (Goncalves *et al.*, 2002). In this study, horses that had a change of water source in the paddock were almost three times more likely ($p=0.0020$; OR=2.9; 95% CI=1.98-4.29) to be colic horses relative to horses not having a change of water source in the paddock. Change in water source in the paddock has been investigated before in conjunction to colic, but no association with presence of colic has been found (Hudson *et al.*, 2001). However, previous studies have reported an increased risk of colic for horses having a changed and decreased amount of water intake in the paddock (Kaya *et al.*, 2009), horses without a continuous supply of water in the paddock (Reeves *et al.*, 1996) and horses provided water from buckets and automatic waterer in the paddock (Kaneene *et al.*, 1997). Even though no association was found between a change of a specific type of water source and colic in the present study, the

information on these changes could be used to explain the results. The majority of the colic horses in this study had had a change from tub (regular or protected from freezing) to bucket and/or automatic waterer in the paddock, which means that the horses might have changed from a water source with open surface to a water source with decreased surface and water flow. Also, as mentioned earlier, the majority of the horses in this study was fed haylage as roughage and/ or had also been subjected to a change from roughage with lower DM content (haylage) to roughage with a higher DM content (hay). When making this type of change, from a diet with low DM to high DM, the water consumption is higher or will be altered (Williams *et al.*, 2014) for the horses that have a change in type of forage from a forage with a low DM to a forage with an increased DM. Altered water consumption together with a decreased or limited flow rate, as a result of a change in water source in paddock, might result in that horses cannot provide themselves with sufficient amounts of water and therefore may be at increased risk of presence of colic.

10.5. Daily intake of salt

The daily intake of salt was considered as one of the main factors for colic as it was included in the final model. In this study, horses having an intake of 11-24 grams of salt were more likely ($p=0.0387$; $OR=2.5$; $95\%CI=1.37-4.41$) to be colic horses relative to horses having an intake of >25 g. This result might be explained by the fact that the intake of salt did not fulfill the requirements of the horse for salt. Replacement of loss of salt related to exercise when horses perform heavy work is required (Jansson *et al.*, 2010). Therefore, information regarding the number of trainings per week and training intensity for the colic horses having a daily intake of 11-24 grams of salt was studied. Fifty percent ($n=27$) of the colic horses that were told to be fed 11-24 grams of salt per day were trained 3-5 times per week. Among the horses trained 3-5 times per week 52 percent ($n=14$) were told to have a training intensity of moderate to heavy work. This indicates that these horses might get too little salt in relation to the losses of salt linked to training. However, as the question regarding the amount salt fed daily did not specify whether it was the amount of additional salt or including the salt lick there might have been a difference in how horse owners interpreted and responded to this question. All horses reported to be provided with 11-24 g salt per day where also reported to have free access to a saltlick, which means that the horse owners most likely interpreted the question as how much additional salt was provided in the diet. This might show that even if the horses were supplemented with salt, it was not sufficient to cover the losses of salt when trained. The concentration of salt in the body fluid is also one of the controlling factors for the total fluid volume in the body. When horses don't receive enough salt it might lead to a decreased ability to maintain fluid balance in the body followed by a decreased water intake (Berglund *et al.*, 2003). As a decreased or limited water intake has been shown to be associated with an increased risk of colic, this could explain why a higher presence of colic was present among these horses in the study, as the main type of colic reported from the respondents was impaction colic.

10.6. Other nutritional-related management factors

Assessment of BCS is a helpful tool to determine if the feed ration covers the energy requirement of the horse, or if the ration may even provide too much energy (Carroll & Huntington, 1988). The results from this study indicated that horses with a BCS lower than 3 (less than desired) and higher than 4 (more than desired) had a higher presence of colic relative to horses having a desired BCS (BCS 3 according to Carroll and Huntington, 1988). Associations between presence of colic and a certain BCS have not been reported in previous studies. A possible explanation for higher presence of colic in horses having a BCS <3 may be

that these horses could be “hard keepers”, which are horses that are naturally prone to be thin and therefore have difficulties to gain weight and keep a good BCS, not able to keep a sufficient BCS on only roughage. To compensate for this, and in order to keep the horse in a sufficient body condition, they feed concentrate which can have a negative effect on the GIT health. This is because starch is not digested in the small intestine and therefore continue into the colon where the starch is broken down by microorganisms. This might lead to an increased fermentation and an increased formation of lactate and propionate, causing a decreased pH and may cause conditions like colic (Julliand *et al.*, 2001). For horses with a BCS >3 it is likely that the higher presence of colic is explained by that they have a daily intake of energy that exceeds their needs. Also, when reviewing the data for these horses they were commonly fed a number of concentrates and supplement feeds which might explain the association of BCS >3 with increased presence of colic. From the data it was also shown that there were more horses having a BCS over three that did not have a feed analysis performed on the roughage fed relative to the number horses that had a feed analysis on the roughage fed. No roughage analysis together with a large variation of concentrates and supplements could easily result in a ration with too much energy, which might be the explanation of the high BCS for these horses. However, it may be difficult to judge the BCS and it requires much experience to be carried out and assessed correctly. Although drawings of horses with different BCS were included in the questionnaire to facilitate for the respondent to give a correct answer for the BCS of their horse, it is still not optimal and maybe it would have been better to attach a text description for each of the scores on the scale. Also, different horse breeds and types have different body types which should be taken into account when judging BCS. For this reason it may not be possible to draw any conclusions about whether the BCS is associated with presence of colic or not and/or to which extent it may be associated with presence of colic.

10.7. Experimental design and execution

Questionnaire based studies are always coupled with uncertainty of the answers received and with no information on who chooses to respond. This means that the data retrieved may be biased in an unknown direction. However, as the questionnaire was advertised through the website “Hästsverige”, a Swedish website that conveys knowledge about horses and horse management based on research, it probably reached horse owners with a special interest in horse-related research, and maybe also a target audience with interest in feed-related problems. Another disadvantage with questionnaires can be that the response rates can be low, which was not the case in this study. High response rate increase the credibility of the results, and also show the relevance of a subject. Also, some questions might have been asked in a way that made the respondents interpret the questions and then respond in a way that was not intended. This was probably the case for the question about salt intake, where some could have specified the total amount of salt per day, while others indicated the amount of salt supplemented in addition to access to salt block. Also, it is not possible to ask supplementary questions or to add questions retrospectively when realizing that more information was needed in order to make final conclusions.

It has also been some difficulties to find articles investigating the association between colic and nutrition-related factors as many were lacking sufficient presentations of the results e.g. missing p-values. Therefore there had to be a severe selection among articles to use as both background and to support the findings in the present study.

10.8. Future research

The majority of previous studies performed on nutrition-related risk factors for colic in horses have been conducted in the USA and UK, which may also be a reason for some of the contradictory results in this study. Therefore, it would be necessary to further investigate nutrition-related factors for colic in Sweden, as there may be large differences in feeding and feeding routines of horses in different parts of the world. One example is that the previous studies investigated the impact of amount, changes in and feeding strategies for hay, whereas the use of roughage with a larger variation in dry matter content, haylage and silage, is more common in Sweden. It would also be of interest to further investigate changes in feeding and feeds, especially how changes between different types of specific feeds and water sources affect the GIT and hence the presence of colic. Liquid vitamins fell out as one of the main factors associated with colic in this study, which has not been shown in previous studies and therefore it may be interesting to further investigate whether or not it is of biological relevance for the risk of colic in horses. Other ways to further investigate nutrition-related risk factors for colic is to perform other questionnaires focused more strongly around a specific factor, like changes in roughage feeding, making room for more detailed and specific questions. It would then also be of interest to complement the questionnaire with information regarding the nutritional and hygienic quality of the feed.

11. Conclusion

The finding in this study indicates that the amounts of feeds and supplements, as well as changes in feeding and watering practices are the main factors affecting the presence of colic in horses. Also, that the equine gastrointestinal health is best served by a slow and constant intake of a high-fiber, low-starch diet and a sufficient amount of water.

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14. Appendix 1

Questionnaire: Feeding- and watering routines as risk factors for colic in horses

Different types of colic is one of the common health problems that horses suffer. In the studies done about risk factors for colic have feed and feeding procedures shown to be important. Therefore, conducted in the spring of 2016 a master thesis at the Swedish University of Agricultural Sciences (SLU), with the aim to investigate precisely routines for feeding the horses to further increase awareness of the factors that increase the risk of colic. That means we want to have as many answers possible, both from horse owners with horses that had colic and from horse owners with horses who have never had colic. It is important for us to be able to make a good comparison.

The investigation carried out by a web questionnaire. One horse per survey applies, which means that an owner of several horses are free to fill in the questionnaire several times, but does not include more than one horse in one survey. The survey is available online t. o. m. 2016-02-29

For questions regarding the study or the questionnaire you can contact the student conducting thesis work, Katrin Lindroth, via email at kali0013@stud.slu.se or by calling at telephone number 070-8306174. Thank you for your participation!

Click on the arrow to the right to proceed to the survey!

1. How would you describe your horse?

a) How old are your horse?

- <3 years old
- 3-6 years old
- 7-15 years old
- 16-20 years old
- >20 years old

b) What gender is your horse?

- A mare
- A gelding
- A stallion

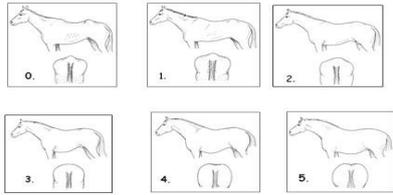
c) What breed is your horse?

- Appaloosa
- Arabian
- Connemara
- Dartmoor
- Thoroughbred
- Fjord horse
- Frieser
- Gotland ponie
- Haflinger
- Icelandic horse
- Coldblooded trotter
- Lipizzaner
- Morgan horse
- New forest
- North-Swedish horse
- Swedish warmblood (SWB)
- Shetland ponie
- Warm blooded trotter
- Welshponie
- Quarterhorse
- Crossbred ponie
- Crossbred horse
- Other breed
- If other breed, specify what breed:_____

d) What is the withers hight of your horse? (Enter the height in cm)

e) Which body condition score (BCS) best fits your horse according to the following images below?

- 0
- 1
- 2
- 3
- 4
- 5



f) How much does your horse weight? (Enter the weight in kg): _____

Help: Approximate weights for different breeds of horses: Icelandic: 280-400 kg, Arabian: 350-450 kg, Thoroughbred Horse: 400-600 kg, Warm-blooded trotter: 400-600 kg, Swedish Warmblood (SWB): 450-700 kg, Ardennes: 700-800 kg.

g) How long have you had your horse?

- 0-6 months
- 7-12 months
- 1-5 years
- > 5 years

h) What disciplines do your horse perform? Choose one or more options

- Dressage
- Show jumping
- Eventing
- Hobby riding/ Walking
- Riding school
- Exhibition
- Breeding
- Western
- Working equitation
- Endurance
- Racing
- Trotting
- Driving
- Company
- Academic riding
- Tournaments and/ or archery
- Other

i) What training intensity best fits your horse training?

- Are not trained
- Light work (leisure riding)
- Medium work (riding school, part leisure riding)
- Hard work (Low- and intermediate-level eventing, some trotting and racing, more difficult classes in jumping)
- Very hard work (horse racing training, elite level eventing, endurance racing)

j) How often do you train your horse per week?

- 0 times per week
- 1-2 times per week
- 3-5 times per week
- 6-7 times per week
- >7 times per week

2. a) What type and amount (kg per day) of roughage are your horse fed?

NOTE! Enter the amount in kg of one or more roughage fed to your horse. If the horse is not fed roughage, leave blank and move on to question 4

- Hay: ____kg
- Haylage (>50% DM): ____kg
- Silage (<50%DM): ____kg
- Straw: ____kg
- Lucerne, pelleted: ____kg
- Lucerne, chopped: ____kg

b) Is there an available feed analysis on the roughage?

- Yes, and can be provided
- Yes, but cannot be provided
- No

3. What are the strategies for feeding roughage?

Help: Roughage includes forage and straw

a) How many times per day is your horse fed roughage?

- 0 times
- 1 time
- 2 times
- 3 times
- 4 times
- >4 times
- Free access

b) How many times per day is your horse fed straw as the only roughage? (i.e. no hay, haylage or silage)

- 0 times
- 1 time
- 2 times
- 3 times
- 4 times
- >4 times
- Free access

c) What is the *maximum* time (in hours) between two feedings of roughage?

- Not fed roughage
- 0-2 hours
- 2-4 hours
- 4-8 hours
- 8-12 hours
- >12 hours
- Free access
- Don't know

d) How is the roughage fed in the stable? Choose one or more options

- Not fed in the stable
- On the floor/ bedding
- In hay net
- In automatic feeder

e) How is the roughage fed in the stable? Choose one or more options

- Not fed in the paddock
- On the ground
- In hay net
- In automatic feeder
- On a feeding rack
- On a feeding table

4. What type and amount (kg per day) of concentrate are your horse fed?

NOTE! Enter the amount in kg for one or more types of concentrate that you're feed your horse. If the horse is not fed any concentrates, leave blank and move on to question 6

Help: 1 L oats weights 0,5 kg, 1 L barley weights 0,6 kg, 1 L dry molassed sugar beet pulp weights 0,4 kg, 1 L Pelleted concentrate weights 0,5 kg (big pellets), 1 L müsli weights 0,4 kg, 1 dl oil weights 0,1 kg

- Oats:____kg
- Barley:____kg
- Molassed sugar beet pulp (Dry weight):____kg
- Pelleted concentrate:____kg
- Müsli:____kg
- Vegetable oil:____kg
- Other:____kg
- Here you can specify the brand of pelleted concentrate and müsli fed, or the type of other feedfed:_____

5. What are the strategies for feeding concentrate?

a) How many times per day is your horse fed concentrate?

- 0 times
- 1 time
- 2 times
- 3 times
- 4 times
- >4 times

b) How many times per day are your horse fed *only* concentrate without access to roughage (and no straw or pasture)?

- 0 times
- 1 time
- 2 times
- 3 times
- 4 times
- >4 times

c) When is the horse fed concentrate (in relation to roughage)?

- Are not fed concentrate
- Before fed roughage
- After fed roughage
- When fed roughage

6a) Does the horse has free access to a salt block?

- Yes
- Yes, and are provided additional salt in the concentrate
- No, do not have access to a saltlick and are not provided salt in other way
- No, but are provided salt in the concentrate
- No, salt are provided in other way
- If the horse is provided in other way, please specify in what way:_____

b) What is the horse daily consumption of salt (g)?

Help: Free access to saltlick: 25-30 g/ day

- 0 g
- <10 g
- 11-24 g
- 25-30 g

7. What type and amount (g or ml per day) of feed supplements is your horse fed?

NOTE! Enter the amount for one or more additional feed you feed your horse and enter the amount in grams of the pellets or granules and in ml if it is a liquid supplement. If the horse is not fed any supplementary feeds, leave blank and continue to question 7

Help: 1 dl pelleted minerals weights approximately 100 g, 1 dl pelleted vitamins weights approximately 80 g. Example of minerals is Calcium and phosphorus Example of vitamins is Vitamin E, B and Biotin.

- Minerals, pelleted: _____g
- Minerals, granulate “powder”: _____g
- Minerals, liquid; _____ml
- Vitamins, pelleted: _____g
- Vitamins, granulate “powder”: _____g
- Vitamins, liquid: _____g
- Other _____g/ml
- Here you can specify the brand of the minerals and vitamins fed, or the type of other mineral _____ or _____ vitamin fed: _____

8. How does the horse have access to water?

a) How does the horse have access to water in the stable? Choose one or more options

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

b) How does the horse have access to water in the paddock? Choose one or more options

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From an automatic waterer
- From a frostless automatic waterer
- Other source: _____

c) If the horse has access to an automatic waterer in the stable, what type of automatic waterer? Leave blank if automatic waterers are not used

- Tongue
- Float
- Pipe valve

d) If the horse has access to an automatic waterer in the paddock, what type of automatic waterer? *Leave blank if automatic waterers are not used*

- Tongue
- Float
- Pipe valve

Help:



9. Did your horse suffer from colic the last six months?

- Yes
- No

10. a) When did your horse suffer from colic?

- Less than two weeks ago
- 2-4 weeks ago
- 1-4 months ago
- 3-4 months ago
- 5-6 months ago

b) What type of colic did your horse suffer from?

- Constipation colic
- Sand colic
- Gas colic
- Other type of colic
- Don't know

c) Was your horse on pasture when suffering from colic?

- Yes
- No

Help: *With pasture means the horse gets all or part of their daily food intake from pasture grasses. Being outside in the paddock in winter i.e. from November to March are not counted as pasture although it is grass in the pasture.*

d) Has the horse been through one of these changes 0-4 weeks before it got colic? Leave blank if none of the options is correct.

- Changed paddock
- Changed flock
- Changed stable
- Changed training intensity

10. Had there been any changes in how your horse were fed roughage 0-4 weeks before suffering from colic?

a) Had there been any change in type and/or batch of roughage during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Hay
- Haylage (> 50 % dry matter content)
- Silage (< 50 % dry matter content)
- Straw
- Lucerne, pelleted
- Lucerne, chopped
- Other

Change to (Choose one or more options):

- Hay
- Haylage (> 50 % dry matter content)
- Silage (< 50 % dry matter content)
- Straw
- Lucerne, pelleted
- Lucerne, chopped
- Other

b) Had there been any change in amount of roughage during this period?

- No
- Yes, increased amount fed per day
- Yes, decreased amount fed per day

11. Had there been any changes in how your horse was fed concentrate 0-4 weeks before suffering from colic?

a) Had there been any change in type and/or batch of concentrate during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Oats
- Barley
- Molassed sugar beet pulp
- Müsli
- Vegetable oil
- Other

Change to (Choose one or more options):

- Oats
- Barley
- Molassed sugar beet pulp
- Müsli
- Vegetable oil
- Other

b) Had there been any change in amount of concentrate during this period?

- No
- Yes, increased amount fed per day
- Yes, decreased amount fed per day

12. Had there been any change in how your horse had access to water 0-4 weeks before suffering from colic?

a) Had there been any change in the access (number of hours / day) to water in the paddock during this period?

- No
- Yes, increased numbers of hours
- Yes, decreased numbers of hours

b) Had there been any change in the access (number of hours / day) to water in the stable during this period?

- No
- Yes, increased numbers of hours
- Yes, decreased numbers of hours

13. Had there been any change in water source (bucket, water cup, etc.) in the stable during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

Change to (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

14. Had there been any change in water source (bucket, water cup, etc.) in the paddock during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

Change to (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

15. Had there been any changes in how your horse was fed roughage the last six months?

a) Had there been any change in type and/or batch of roughage during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Hay
- Haylage (> 50 % dry matter content)
- Silage (< 50 % dry matter content)
- Straw
- Lucerne, pelleted
- Lucerne, chopped
- Other

Change to (Choose one or more options):

- Hay
- Haylage (> 50 % dry matter content)
- Silage (< 50 % dry matter content)
- Straw
- Lucerne, pelleted
- Lucerne, chopped
- Other

b) Had there been any change in amount of roughage during this period?

- No
- Yes, increased amount fed per day
- Yes, decreased amount fed per day

16. Had there been any changes in how your horse was fed concentrate during the last six months?

a) Had there been any change in type and/or batch of concentrate during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Oats
- Barley
- Molassed sugar beet pulp
- Müsli
- Vegetable oil
- Other

Change to (Choose one or more options):

- Oats
- Barley
- Molassed sugar beet pulp
- Müsli
- Vegetable oil
- Other

b) Had there been any change in amount of concentrate during this period?

- No
- Yes, increased amount fed per day
- Yes, decreased amount fed per day

17. Had there been any change in how your horse had access to water *the last six months*?

a) Had there been any change in the access (number of hours / day) to water in the paddock during this period?

- No
- Yes, increased numbers of hours
- Yes, decreased numbers of hours

b) Had there been any change in the access (number of hours / day) to water in the stable during this period?

- No
- Yes, increased numbers of hours
- Yes, decreased numbers of hours

18. Had there been any change in water source (bucket, water cup, etc.) in the stable during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

Change to (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

19. Had there been any change in water source (bucket, water cup, etc.) in the paddock during this period?

- Yes
- No

If Yes, specify what type of change below:

Change from (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

Change to (Choose one or more options):

- Don't have access to water in the stable
- From tub
- From a frostless tub
- From a waterer
- From a frostless waterer
- Other source: _____

In particularly interesting cases, there may be reason to ask further questions to supplement the survey responses or find out more information about the horse. Information submitted here will not be used for any purpose other than to complete the survey and will not be disclosed to any third party. Mail Addresses, phone numbers or addresses will not be used for any type of promotional messages, or spam, or anything that does not have the right to do this study. As the survey is anonymous, it is completely optional to provide details.

NOTE! If you enter your email address we will send you the link to the mater thesis while finished.

20. a) May we contact you to complement the questionnaire or ask further questions?

- Yes
- No

If yes, answer the questions below:

b) How can we contact you? Choose one or more options

- By email
- Over the phone

c) What is your contact information? (Please include your name, email address and / or phone number)

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