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*Original article*

# Assessment of sand accumulation in the gastrointestinal tract and its excretion with stool in silesian foals

N. Siwińska<sup>1</sup>, A. Łuczka<sup>2</sup>, A. Żak<sup>1</sup>, M. Słowikowska<sup>1</sup>, A. Niedźwiedź<sup>1</sup>

<sup>1</sup> Department of Internal Medicine and Clinic of Diseases of Horses, Dogs and Cats,

<sup>1,2</sup> Faculty of Veterinary Medicine, University of Environmental and Life Sciences,  
Pl. Grunwaldzki 47, 50-366 Wrocław, Poland.

## Abstract

The aim of this study was to assess sand accumulation in the gastrointestinal tract and fecal sand excretion in Silesian foals using three diagnostic methods and taking into account the sex and age of the animals. Another aim of the study was to compare the three diagnostic methods. The study was carried out on 21 clinically healthy Silesian foals (10 females and 11 males) from 9-28 weeks old grazed on permanent pasture. The sand intake was assessed using a sedimentation test, abdominal ultrasonography and a quantitative evaluation of sand per 100 g of stool. In the sedimentation test, the sand was palpable in the stool of 57.1% of the horses, and clearly visible in 42.9% of the animals. The ultrasound examination revealed the presence of sand in the gastrointestinal tract in 66.7% of the horses. It was limited to a single location in 60% of the horses, while it was present in several regions in 40% of the horses. The mean amount of sand was  $0.14 \pm 0.33$  g per 100 g of stool. It did not exceed 0.1g in 71.4% foals, while it ranged from 0.1-0.5 g in 23.8% foals. In 4.8% of the animals, it amounted to 1.6 g per 100 g of stool. There was no correlation between age and gender and the results. There was a positive correlation between the ultrasound examination and the sedimentation test.

Sand may be accumulated in the gastrointestinal tract of foals without any clinical signs. The amount of sand excreted in the stool is not an indicator of the amount of sand accumulated in the gastrointestinal tract. An abdominal ultrasound examination should be combined with a sedimentation test for more specific results.

**Key words:** sand content, sand accumulation, abdomen ultrasonography, sedimentation, horses

## Introduction

The continuous intake of sand (and dirt) with feed without its equivalent excretion in stool may cause its accumulation in the digestive tract and lead to digestive disorders (Magdesian 2005). Sand colic is the most common symptom of sand intake, as the accumulating sand may lead to mechanical irritation of the gastric and intestinal mucosa, decreasing peristalsis and narrowing the intestinal lumen. Other symptoms of sand accumulation include diarrhoea, a decrease in body mass and increase in body temperature  $> 38.6^{\circ}\text{C}$ , which usually suggests gastrointestinal inflammation (Hotwagner et al. 2008, Hart et al. 2013). Although sand accumulation is most commonly described in adult horses, symptoms associated with it may also occur in young animals even in their first week of life, as reported by McAuliffe (2004). Sand accumulation predominantly affects horses in geographical areas with sandy (light) soils and little rain. This phenomenon almost exclusively affects animals that spend most of their time on poor condition pastures in dry areas, as well as in paddocks and boxes in stables. Hay collected from fields with numerous molehills may also contain a lot of sand and may be associated with this pathology.

There are numerous reports of sand accumulation in adult horses. Several studies have been carried out to determine the success of various diagnostic and therapeutic methods, including conservative and surgical treatment, prognosis and both short and long-term outcomes (Korolainen and Ruohoniemi 2002, Korolainen 2003, Husted et al. 2005, Hotwagner et al. 2008, Hart et al. 2013, Kilcoyne et al. 2017). Most of the studies included horses of different ages and breeds, kept under various conditions, with clinical signs of sand accumulation (Korolainen and Ruohoniemi 2002, Korolainen 2003, Hotwagner et al. 2008, Hart et al. 2013). There is only one study assessing sand accumulation, carried out by Ruohoniemi et al. (2001) which included two six-week old foals. To our best knowledge, none of the available studies assesses the prevalence and diagnostics of asymptomatic sand accumulation in the gastrointestinal tract of foals. This study is also the first study assessing foals of one breed, from one stable and kept under uniform conditions.

The aim of this study was to assess the level of sand accumulation in the gastrointestinal tract and its excretion in healthy Silesian foals kept in rotational grazing systems using three diagnostic methods and taking into consideration the sex and age of the horses. Another aim of the study was to compare the three diagnostic methods.

## Materials and Methods

### Study group

The study was carried out on a group of 21 Silesian foals (including 10 mares and 11 geldings) in August 2017 kept at the Książ Stallion Stud Farm. The foals were from nine to 28 weeks old (mean 19 weeks old, standard deviation  $[\pm] 5.33$ ). The horses were kept with their mothers in a common stable without boxes. The horses grazed on grassy paddocks all day long and also received concentrates and roughage twice a day. The paddocks contained good quality low grass without visible molehills. The obtained feed also appeared free from contamination and was administered in a common manger and feeding racks.

The foals included in this study underwent a full clinical examination, which included an assessment of the body temperature, respiratory rate, heart rate, mucous membranes, the capillary refill time and intestinal motility. Based on the clinical examination and owner anamnesis, gastrointestinal disease was ruled out. In order for the foals to be included in the study, the mares had to be healthy prior to and in the perinatal period (normal pregnancy, delivery and normal placenta). The following methods were used to assess sand accumulation: a sand sedimentation test using a disposable rectal glove, an abdominal ultrasound and quantitative assessment of the presence of sand in stool. All the examinations were carried out on non-fasting unseated animals. The examinations were performed on all the animals at the same time of day when the animals returned from grazing before receiving a concentrate feed. All of the examinations performed in this study were non-invasive and are routinely performed in everyday medical practice. In accordance with the existing law applicable in Poland, Experiments on Animals Act of January 15<sup>th</sup> 2015 (Journal of Laws of the Republic of Poland, 2015, item. 266), non-invasive clinical studies do not require ethical approval.

### Sample collection

Five fresh stool samples were collected from the top layer of the stool uncontaminated by the ground. In total, twenty-one samples were collected from 21 foals. The samples were divided into two. One half was used for the quantitative analysis of sand in the stool sample, while the other half was used for the sedimentation test. Both tests were performed immediately after the sample collection.

### Sedimentation test in a rectal glove

A 20 g fresh stool sample was placed in a rectal glove. One litre of lukewarm water was then added, and

the content was thoroughly mixed in order to obtain a uniform consistency. The glove was tied and suspended for 60 minutes in order for the sand sediment to collect at the bottom of the glove. The procedure was repeated for each stool sample. After 60 minutes, the fingertip of each glove was assessed visually and via palpation. The samples were assessed using a 0-4 scale created by the authors, where 0 indicated no sand; 1 – visible, impalpable traces of sand; 2 – visible and palpable small amount of sand, which collects as sediment; 3 – a larger amount of sand that is visible without palpation.

### **Abdominal ultrasound**

The ultrasound examination was carried out on standing horses held by two assistants. Six foals were excluded from the study due to lack of cooperation during the examination. Hence, 15 horses were examined. A MyLab 30 Gold Vet scanner equipped with a convex 1-5 MHz (Esaote) probe was used. The horses were not clipped for the procedure. The hair at the examination site was cleaned, washed with warm water mixed with soap containing chlorhexidine and moistened with alcohol. Ultrasound gel and a protective cover were then applied to the probe. The presence of sand in the digestive tract was assessed at three sites, according to the “fast localised abdominal sonography of the horse” protocol (Busoni et al. 2011, Southwood and Fehr 2012) used to examine the large intestine: the left flank diagnostic window, the right flank diagnostic window and the ventral window. The left/right diagnostic window refers to the space between the last rib and the iliac crest of one side of the body. The ventral window extends ventrally from the manubrium of the sternum to the fold of the groin (Busoni et al. 2011). In the ultrasound examination, sand accumulation in the digestive tract casts a strong acoustic shadow, which hinders the imaging of deeper structures. The amount of sand visible in the ultrasound examination was assessed using a three point scale created by the authors, where the sand accumulation was assessed from + (trace amounts of sand) to +++ (large amounts of sand). Accordingly, 0 – no sand visible on ultrasound, 1 – sand accumulation assessed as “+”, present in a single location, 2 – amount of sand assessed as “++” in one location or as “+” in numerous locations, 3 – amount of sand assessed as “++/+++” in numerous locations.

### **Quantitative assessment of sand accumulation**

A 60 g sample of fresh stool was placed in a plastic cup and 300 mL of lukewarm water was added. The content was then mixed and left for six hours. The solution was then sieved (using a sieve with holes

enabling the passage of sand particles and stopping larger elements) into a clean cup, and the sieve was further rinsed with 200 mL of lukewarm water to make sure all the sand particles entered the cup. The cup filled with sand and water was left to settle. The supernatant was then carefully removed, and the remaining sand mixed with water was set aside until all the water evaporated. The sand was then weighed using an Explorer Ohaus analytical balance. The obtained result was then converted to correspond to a 100 g stool sample. This methodology was devised by the authors based on the method presented by Landes et al. (2008).

### **Statistical analysis**

The obtained results were evaluated using descriptive statistics. The Pearson correlation coefficient was used to calculate the relationship between the age of the foals and the quantity of sand accumulation. The Spearman rank correlation coefficient was used to compare the correlation between age and the test results and the correlation between each diagnostic test. Fisher’s test (RxC) was applied to determine the correlation between the sex of the animals (nominal qualitative variable) and the results of the diagnostic tests. In all the analyses, a p value <0.05 indicated a statistically significant result. The statistical analyses were performed using PQStat version 1.6.6 software.

## **Results**

### **Sedimentation test**

The sedimentation test revealed a variable presence of sand in the stool in the studied horses. Trace amounts of sand in the stool samples were found in twelve out of the 21 foals (57.1%) and reached a score of 1 on the 3-score scale (0-3). In nine foals (42.9%), the sand was both palpable and visible. Eight out of the nine foals (88.9% in this group, and 38.1% of all the studied animals) reached a score of 2, while one foal (constituting 11.1% of this group and 4.8% of all the studied animals) reached a score of 3. The mean score using this scale was  $1.48 \pm 0.58$ ;  $1.54 \pm 0.55$  for stallions and  $1.4 \pm 0.66$  for mares. No statistically significant correlation was found between the age of the foals and the test results ( $r=-0.09$ ,  $p=0.696$ ) or the sex and the test results ( $p=0.696$ ).

### **Results of the abdominal ultrasound**

There were features of sand accumulation in the digestive tract in 10 (66.6%) out of the 15 examined horses. In six of those horses (60%), sand was identified

Table 1. Statistical correlations between the applied diagnostic methods used to assess sand accumulation in the equine digestive tract.

	Sedimentation test	Abdominal ultrasound	Quantitative assessment of sand accumulation per 100 g of stool
Sedimentation test	-	r=0.47*	r=0.018
Abdominal ultrasound	r=0.47*	-	r=0.307
Quantitative assessment of sand accumulation per 100 g of stool	r=0.018	r=0.307	-

\* significance level set at  $p=0.05$

in one diagnostic window, while in four horses (40%) it was recognised in two locations. Two horses scored 2 points based on the ultrasound findings suggesting the presence of small amounts of sand, while two foals had significant sand accumulation and scored 3 points. In the cases where sand was found in a single location, it was usually identified in the left flank diagnostic window (5 of 6 horses) or the right flank window (1 of 6 cases). In all four cases where sand was present in two areas, it was observed in the ventral diagnostic window and the left flank window. The mean value of intestinal sand accumulation was  $1.07 \pm 0.99$ ; with a value of  $1.14 \pm 0.98$  for stallions and  $1 \pm 1.0$  for mares. There were no statistically significant correlations between the age ( $r=-0.344$ ,  $p=0.210$ ), gender ( $p=0.999$ ) and the results of the ultrasound examination.

#### Quantitative assessment of sand accumulation in stool samples

The mean amount of sand in the stool samples in all the foals was  $0.143 \pm 0.329$  g per 100 g of stool. In 15 foals (71.4%), the amount of sand did not exceed 0.1 g per 100 g of stool, in five (23.8%) foals it ranged from 0.1-0.5 g of sand per 100g of stool, while in one foal (4.8%) it reached 1.545g of sand per 100 g of stool. There was no statistically significant correlation between the age ( $r=-0.201$ ,  $p=0.395$ ) or the gender ( $p=0.314$ ) of the studied foals and the amount of sand per 100 g of stool.

#### An assessment of the correlation between the diagnostic methods

The results of the correlations between the diagnostic methods are presented in Table 1. There was no statistically significant correlation between the sedimentation test and the quantitative assessment of sand accumulation in the stool samples ( $r=0.018$ ,  $p=0.937$ ), or between the abdominal ultrasound examination and the quantitative assessment of sand accumulation in the stool samples ( $r=0.307$ ,  $p=0.286$ ). A weak positive correlation was noted between the sedimentation test and the abdominal ultrasound examination ( $r=0.47$ ,  $p=0.05$ ).

## Discussion

These are the first findings of sand accumulation in the gastrointestinal tract and sand content in stool in a group of healthy Silesian foals using three diagnostic methods.

The research to date has focused on the assessment of sand accumulation in adult horses with clinical signs of sand accumulation, which has also been induced experimentally (Ragle et al. 1989, Landes et al. 2008). Significant sand accumulation in the digestive tract is associated with severe clinical signs and may be seen in horses with colic (Ruohoniemi et al. 2001, Korolainen and Ruohoniemi 2002, Korolainen 2003, Keppie et al. 2008, Hart et al. 2013, Kilcoyne et al. 2017). However, a small amount of sand in the gastrointestinal tract and stool in horses seems to be quite common, particularly in grazing animals. A study carried out by Husted et al. (2005) on Icelandic horses that grazed the whole year revealed that more than half of clinically healthy horses had trace amounts of sand in their stool. That study found that all the studied foals excreted some sand in their stool. The differences in the findings may be attributed to various age groups and types of management of the horses. In our study, sand was present in the gastrointestinal tract of young foals, which is in accordance with the data of Ruohoniemi et al. (2001). According to the data in this study, a lack of clinical signs does not rule out sand accumulation in the digestive tract. The amount of sand in the digestive tract required to elicit clinical signs remains undetermined and may differ between individuals, as reported by Bertone et al. (1988) and Specht and Colahan (1988). Similarly, in our study, the amount of accumulated sand differed between foals. According to Husted et al. (2005), the well-known risk factors of sand accumulation include the type of soil, type of pasture and feeding practise. The authors of the existing studies assessed the degree of sand accumulation in animals from different areas and stables (Korolainen and Ruohoniemi 2002, Keppie et al. 2008, Kilcoyne et al. 2017). Hence, the authors of this study chose to examine young horses of a single horse breed native to Lower Silesia. The study by Husted et al. (2005) was the only other study that assessed

a single breed of horse (the Icelandic horse). That studied group was uniform as all the foals were kept in the same conditions and grazed on the same pasture. Loamy soil formed from sedimentary rock predominates in the northern territories of Wałbrzych, where the National Stallion Depot at the Książ Castle is located. The horses grazed on high quality pasture covered with short green grass. Short grass may lead to the ingestion of larger quantities of soil, especially when ingested with grass roots. The feeding system itself and the quality of the feed concentrate do not seem to affect the amount of accumulated and excreted sand considering that the feed was administered in feeding mangers and racks. To date, no sex or breed predisposition to sand accumulation has been found, which is in accordance with our findings in the presented group of foals. The study by Husted et al (2005) is the only study that presents the percentage effect of gender on sand accumulation in the gastrointestinal tract. However, those authors did not present correlation coefficients. They reported a similar prevalence of sand accumulation in mares and geldings and a slightly lower prevalence in stallions. It is unclear whether the stallions were kept in different conditions to the mares and geldings.

Sand accumulation is mainly diagnosed based on auscultation of the abdominal cavity (Ragle et al. 1989), on ultrasound examination (Korolainen and Ruohoniemi 2002, McAuliffe 2004) and gastrointestinal radiography (Ruohoniemi et al. 2001, Kilcoyne et al. 2017). Abdominal radiography is considered the most sensitive method used to diagnose sand accumulation. Numerous authors described this method comprehensively in the context of diagnosis and monitoring of the treatment of sand accumulation (Ruohoniemi et al. 2001, Korolainen and Ruohoniemi 2002, Keppie et al. 2008, Kilcoyne et al. 2017). However, this technique requires the use of specialist high power equipment, usually only available in clinical conditions. If gastrointestinal ultrasonography is unavailable, an ultrasound examination is considered a practical and reliable alternative, with a specificity and sensitivity of 87.5%, as described by Korolainen and Ruohoniemi (2002).

In their study, Korolainen and Ruohoniemi (2002) compared the accuracy of radiography combined with ultrasonography for the diagnosis of sand accumulation and concluded that ultrasonography alone suffices in the diagnosis of intestinal sand accumulation. However, this technique gives limited information about the quantity of sand within the intestinal lumen due to the presence of an acoustic shadow. From the authors' experience, it may be difficult to use this technique in foals, uncooperative animals and without the use of pharmacological restraint.

Other studies (McAuliffe 2004, Hotwagner et al.

2008, Keppie et al. 2008, Hart et al. 2013) have shown that sand more commonly accumulates in the large intestine (particularly in the colon and cecum) than in the small intestine. This is due to a larger intestinal diameter, the presence of water absorption processes and a slower passage of stool in the large intestine. Hence, when performing abdominal ultrasonography, the authors used diagnostic windows to visualise segments of the colon. All of the animals in the study by Ruohoniemi et al. (2001) were diagnosed with naturally induced gastrointestinal sand accumulation, although the authors did not mention whether that accumulation was symptomatic in any of the animals. In the studied foals, the sand was located in the colon, which is consistent with previous reports (Korolainen and Ruohoniemi 2002, McAuliffe 2004). Sand was most commonly present in the left ventral colon, which is contrary to the findings of Ruohoniemi et al. (2001) in adult horses, where most of the sand accumulated in the right dorsal and transverse colon. This difference in sand location may be attributed to the different age of the animals, as well as management differences. Peristaltic movements, which may be slower in the case of sand accumulation, are also assessed in the ultrasound examination (Korolainen and Ruohoniemi 2002). In the current study, colonic peristalsis in the foals was not assessed extensively as this would prolong the study and cause stress to the young animals that were unaccustomed to medical procedures. Chemical restraint was not used in the animals due to the proven effect of alpha2-agonists on intestinal peristalsis (Smith 2008, Zullian et al. 2011). Relative assessment of peristalsis indicated normal colonic motility. This, in turn, may be a good prognostic factor for rapid excretion of the accumulated sand.

The sedimentation test using a rectal glove or bucket is a commonly used quantitative assessment of sand accumulation (Husted et al. 2005, Hukkinen 2015). This is a simple technique that does not require any specialist equipment and may be carried out in field conditions. According to Hukkinen (2015), this test is relatively accurate (over 80% sensitive) but it does not provide information regarding current accumulation of sand in the animal. Lack of sand in the sedimentation test does not rule out its presence in the gastrointestinal tract. Hence, the test should be treated as a supplementary technique (Hukkinen 2015). The sedimentation test has been described in several other studies (Husted et al. 2005, Hukkinen 2015). Its main limitation is lack of standardization. Depending on the examiner preferences, various amounts of stool and water may be used. Similarly, the result may be recorded after a shorter (20 min.) or longer (1/12/24 hours) period of time based on a subjective scale. Thus, it is difficult to compare the

results presented in this study with those of other authors (Husted et al. 2005, Hukkinen et al. 2015).

The quantitative assessment of sand in stool is unpopular and rarely used in clinical practice. The only available reference to this method describes its use on a group of eight clinically healthy horses (Landes et al. 2008). The results obtained by those authors (amount of sand in g /100g of stool) are almost 20-fold higher than those obtained in the present study. These large discrepancies may be attributed to the difference in the age of the animals in the two studies, and may point to a larger gastrointestinal sand accumulation in adult horses than in foals. In the present study, the age of the animals did not correlate with the amount of sand accumulation although their age range was quite narrow. Husted et al. (2005) studied adult horses and noted a slight linear decrease in the detection of the amount of sand accumulated in the stool with age. Similarly, the horses in the study of Landes et al. (2008) spent entire days on dry paddocks, increasing the risk of sand and soil ingestion with feed more than in the studied group of foals. Sand accumulation was much higher in one of the studied foals than the mean value for the entire group of foals, which may have been caused by a higher accidental ingestion of sand or better motility causing more rapid sand excretion compared to the remaining foals. Other results in this foal did not differ from the results obtained in the other horses. Based on their own experience, the authors believe that the quantitative assessment of sand accumulation may not be accurate as the separation of all the sand from the stool sample proved difficult.

There are few studies presenting statistical correlations between different diagnostic techniques of sand accumulation. The most significant study, by Korolainen and Ruohoniemi (2002), revealed a strong correlation between abdominal radiography and ultrasonography. In turn, Husted et al. (2005) proved a significant correlation between the sedimentation test using a rectal glove and sand decantation. In our study, a correlation between the sedimentation test and abdominal ultrasonography was observed. The remaining diagnostic methods do not seem to correlate, which may be caused by the small size of the study group. The significant correlation obtained in this study may indicate the reliability of both tests due to their complementarity.

One of the limiting factors of the current study was the fact that no abdominal radiography was performed. This was caused by two factors. Firstly, the foals would have had to be chemically restrained for the procedure. Secondly, the authors did not have access to a suitable, portable X-ray machine. Lack of chemical restraint also decreased the number of horses that underwent abdominal ultrasonography, due to lack of cooperation

of some of them. The authors plan to extend the study to include more foals, horses of other breeds, of various ages, different use and different maintenance. Other complementary diagnostic techniques will be used. It would also be interesting to study animals with clinical symptoms of gastrointestinal sand accumulation.

## Conclusion

This is the first study that assesses sand accumulation in healthy foals of a single breed. The collected data suggest that sand is accumulated in asymptomatic foals. In the authors' opinion, the sedimentation test using a rectal glove is the quickest and easiest diagnostic method. Due to its simplicity, it may be carried out in the field. However, the amount of sand excreted in the stool is not an accurate indicator of the amount of sand accumulated in the gastrointestinal tract. Abdominal ultrasonography was the most reliable and practical diagnostic tool used in this study. Hence, both methods should be used complementarily.

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