Original Article

Evaluation of the Hairballs in the Gastrointestinal Tract and Urinary Stones in Razi Institute Laboratory Rabbits

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Background: Usually, the daily self-grooming by rabbits leads to fur accumulation in the animal’s stomach. Since rabbit hair is looser than other animals and constantly licks their body, the fur can be pulled out easily. On the other hand, rabbits are susceptible to urinary stone formation.

Objectives: This study was designed to investigate the presence of hairballs and urinary stones in Razi Institute Laboratory rabbits.

Methods: During the 1 year, the albino Dutch laboratory rabbit colony, in research, breeding, and production of the Laboratory Animals Department of Razi Institute, including 106 males, 287 females, and 166 kittens, were monitored. After the necropsy of the selected animals, the gastrointestinal tract (stomach and intestines) were examined for the presence of hair and hairballs. Then the urinary system (kidneys, ureter, urinary bladder, and urethra) was examined for any urinary stones.

Results: No symptoms of anorexia, lethargy, abdominal pain, weight loss, decrease and abnormal stools were observed in them, and also no mortality occurred in the whole colony. All samples’ stomach was full, indicating enough eating. No gas or congested spots, or hemorrhage were observed in the intestines. The amount and consistency of stool in the intestines were normal. In none of the samples, hairballs were observed, but in most rabbits’ stomachs (both sexes), a small amount of hair was observed in the stomach contents. Also, no symptoms of urinary stones were observed in the colony of the studied rabbits.

Conclusion: Balanced diet, supply of nutritional requirements, and the absence of any stressors in breeding environments have played a key role and prevented many diseases, such as hairballs and urinary stones. No observation of urinary stones in this study could lead to the hypothesis that infection with the bacteria that cause urinary stones in the studied rabbits was eliminated or non-pathogenic, indicating specific pathogen-free animals. However, bacterial and other infectious agent monitoring should be specialized.

Keywords: Gastrointestinal tract, Hairballs, Rabbit, Stones, Urinary tract

ABSTRACT

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1. Introduction

Rabbits are herbivorous animals. Also, they are named fibrevores animals due to their special high-fiber diet. In laboratory rabbits, food is available to the animals without restriction; therefore, the animals’ stomachs are never empty and occupies more than 15% of the gastrointestinal tract contents (Quesenberry & Carpenter, 2011; Fallahi & Mansouri, 2015).

Usually, the daily self-grooming by rabbits leads to fur accumulation in the animal’s stomach. Since rabbit hair is looser than other animals and constantly licks their body, the fur (also known as hair, cony, coney, comb, or lapin) can be pulled out easily. Therefore, some hair may enter the mouth and pace through the stomach. Also, the fur spilled on the food is always eaten. On the other hand, hair plucking in the last third of pregnancy to make a nest box is common, and some plucked hairs may be eaten. Trichophagia is a condition in which excessive hair is eaten (Quesenberry & Carpenter, 2011). Eaten hair in the stomach becomes hairballs or fur balls that, after a while, due to enlargement in the stomach, remain and cannot pass through the pyloric valve to enter the duodenum. This condition is also called trichobezoar (Figure 1) (Mondal et al., 2006; Quesenberry & Carpenter, 2011). Rabbits with trichobezoar develop anorexia, lethargy, movement inability, reduced mobility, abdominal pain, weight loss, abdominal distention, lack of self-cleaning, and stool pieces become smaller than usual (Fukumura et al., 2012; Nowland et al., 2015). The formation of hairballs in the stomachs of other animals, such as sheep, is also common, especially in woolly sheep. In humans, hair eating occurs following a mental disorder called Rapunzel syndrome (Godara et al., 2015).

Urinary stones may be found in the kidneys, urinary bladder, and urinary tract of rabbits of any age and breed (mostly middle-aged and older rabbits) (Figure 2). Rabbit urine is concentrated and sometimes creamy due to its calcium carbonate and ammonium magnesium phosphate crystals and occasionally dark red due to the existence of porphyrin and may be mistaken for hematuria due to blood in the urine (Lee et al., 1978; Pinto Filho et al., 2016). The rabbit’s urine pH is alkaline and between 8 and 9. The alkalinity of rabbit urine is due to the high alkaline carbonates in the diet formulation. The urinary stones comprise magnesium, ammonium, phosphate, oxalate, carbonate, uric acid, urate, and cysteine. The urinary stones are usually made up of calcium carbonate and oxalate. Urinary stones are more common in male rabbits due to the length of the urinary tract (King, 2006; Nowland et al., 2015). Among the causes of urinary stones in rabbits, genetic factors, hormonal and nutritional imbalances, infections, especially bacteria in the urinary bladder, and kidney diseases are the most important causative factors (Quesenberry & Carpenter, 2011). On the other hand, some nutritional deficiencies, such as Vitamin B6 deficiency, also increase oxalate secretion and lead to urolithiasis (Quesenberry & Carpenter, 2011). It is well documented that some breeds, such as the dwarf lop, are susceptible to urinary stone formation. Therefore, laboratory strains should not be obtained from them (White, 2001).

Urease-producing bacteria, such as Proteus mirabilis, Klebsiella pneumonia, Staphylococcus aureus, Pseudomonas aeruginosa, and Serratia, are always associated with stone formation and recurrence. These bacteria break down urea into ammonium and carbon dioxide, causing the urine to become alkaline and form phosphate salts. Intestinal microflora could prevent and or induce the formation of kidney stones. Oxalobacter formigenes is an anaerobic gram-negative bacterium that reduces oxalate in the gastrointestinal tract and has been shown to avoid calcium oxalate stones. In patients with kidney stones, bacteria of the genus Bacteroides are found to be 3-4 folds more than normal. Therefore, their role in the occurrence of stones is possible. Gram-positive bacteria of Eubacterium have shown the opposite effect on oxalate formation, and Escherichia coli has the opposite effect on citrate formation (Wang et al., 2021). Clinical signs of rabbits with urinary stones include lethargy, anorexia, loud, painful, gritted teeth, hunching, reluctance to move, pushing the abdomen to the ground, reduced neonatal care, force, and painful urination, and the presence of blood in the urine (hematuria) can lead to death (Quesenberry & Carpenter, 2011). Also, in obese animals, the natural contraction of the urinary bladder and its normal position is disturbed due to the presence of fat around the kidneys and urinary ducts, and the bladder lacks a contractile state for complete excretion of urine. Therefore, a significant amount of urine remains inside the bladder, providing the basis for stone formation. On the other hand, the resulting pain from arthritis, pododermatitis, and spinal problems leads to incomplete urination and causes urination retention (White, 2001).

High dietary calcium is involved in the formation of urinary stones. Prolonged consumption of high-calcium diets causes calcification (calcium accumulation) in the kidneys and aorta (Quesenberry & Carpenter, 2011). Urine is the main way of excreting calcium in rabbits. If dietary calcium is high, urinary calcium excretion is also increased. In rabbits, 45%-60% of the calcium that
enters their body is excreted in the urine, and 20% is excreted in the feces. In other mammals, such as rats, 93% is excreted in the feces and only 2% in the urine. Also, blood calcium in rabbits is 30%-50% higher than in other mammals (Quesenberry & Carpenter, 2011).

Oxalate is metabolized in the intestines by bacteria. Therefore, insufficient or destroyed intestines microflora consequently leads to oxalate accumulation in the kidneys and urinary tract which can cause stones (Wang et al., 2021).

This study was designed to investigate the presence of hairballs and urinary stones in Razi Vaccine and Serum Research Institute Laboratory rabbits.

2. Materials and Methods

During 1 year (June 2021-February 2022), the albino Dutch laboratory rabbit colony, in the Department of Research, Breeding and Production of Laboratory Animals, Razi Vaccine and Serum Research Institute, including 106 males, 287 females, and 166 kittens, were monitored. In two turns with an interval of 6 months, 45 female and 12 male rabbits aged 9-10 months (2-2.2 kg) were randomly selected. The animals were apparently healthy and negative for external parasites and had no specific disease. The rabbits were fed with the standard pellet of laboratory rabbits, and they had access to water ad libitum. The rabbit breeding system was conventional, and the material of the cages was aluminum with a mesh floor, and the tray under it was made of stainless steel. There was one female rabbit with neonates in each cage, and the male rabbits were kept in separate cages. The temperature of the breeding room was 22-24°C, with humidity in a range of 45%-55%. The light/dark cycle was 12:12 hours per day, and the light intensity was less than 325 lux.

The selected animals were transferred to the necropsy room, and first, according to the ethical principles, euthanasia (by combining ketamine at 600 mg/kg and xylazine at 30 mg/kg by intramuscular injection) was performed (Baneux et al., 1986). After the necropsy, the gastrointestinal tract (stomach and intestines) was exam-
ined for hair and hairballs. Then the urinary system (kidneys, ureter, urinary bladder, and urethra) was examined for any urinary stones. Also, the diets used by the studied rabbits were analyzed to compare the nutritional requirements of these animals.

Table 1 presents the nutritional requirements of laboratory rabbits and the analysis of the diet used by the rabbits in this study.

### Rabbit nutritional requirements and diet analysis

<table>
<thead>
<tr>
<th>Material</th>
<th>Rabbit Nutritional Requirements</th>
<th>Analysis of Rabbit Diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein (%)</td>
<td>15-17</td>
<td>18.2</td>
</tr>
<tr>
<td>Energy (kcal/lb)</td>
<td>950</td>
<td>912</td>
</tr>
<tr>
<td>Crude fat (%)</td>
<td>2</td>
<td>2.4</td>
</tr>
<tr>
<td>Crude fiber (%)</td>
<td>10-12</td>
<td>11.17</td>
</tr>
<tr>
<td>Calcium (%)</td>
<td>0.45</td>
<td>0.5</td>
</tr>
<tr>
<td>Phosphorus (%)</td>
<td>0.37</td>
<td>0.31</td>
</tr>
<tr>
<td>Arginine (%)</td>
<td>0.76</td>
<td>1.02</td>
</tr>
<tr>
<td>Histidine (%)</td>
<td>0.38</td>
<td>0.39</td>
</tr>
<tr>
<td>Isoleucine (%)</td>
<td>0.76</td>
<td>0.89</td>
</tr>
<tr>
<td>Ileucine (%)</td>
<td>1.2</td>
<td>1.34</td>
</tr>
<tr>
<td>Lysine (%)</td>
<td>0.92</td>
<td>0.88</td>
</tr>
<tr>
<td>Methionine (%)</td>
<td>0.6</td>
<td>0.57</td>
</tr>
<tr>
<td>Phenylalanine (%)</td>
<td>0.76</td>
<td>0.84</td>
</tr>
<tr>
<td>Threonine (%)</td>
<td>0.61</td>
<td>0.63</td>
</tr>
<tr>
<td>Tryptophan (%)</td>
<td>0.23</td>
<td>0.23</td>
</tr>
<tr>
<td>Valine (%)</td>
<td>0.84</td>
<td>0.88</td>
</tr>
<tr>
<td>Vitamin B6 (mg/kg)</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Vitamin B2 (mg/kg)</td>
<td>4</td>
<td>4.85</td>
</tr>
<tr>
<td>Choline (g/kg)</td>
<td>1.2</td>
<td>1.34</td>
</tr>
</tbody>
</table>

3. Results

Hairballs

During one year of examination in the colony of albino Dutch rabbits, no symptoms of anorexia, lethargy, inability to move, decreased mobility, abdominal pain and distension, weight loss, lack of self-cleaning, or abnormal stools were observed. Also, no mortality occurred in the whole colony. Of 45 female and 12 male rabbits, no abnormalities were observed in the contents of the stomach and intestines after necropsy. All samples’ stomach was full, indicating eating had been enough (Figure 3). No gas or congested spots, or hemorrhage were observed in the intestines. The amount and consistency of stool in the intestines were normal. After opening the stomach and various parts of the intestines, in none of the samples, hairballs were observed, but in most rabbits’ stomachs (both sexes), a small amount of hair was observed in the stomach contents (Figure 4).

Urinary stones

The recorded data revealed no symptoms of urinary stones, including lethargy, anorexia, painful gnashing of teeth, hunchbacked, inability to move, pushing the ab-
demen to the ground, reduced neonatal care, or painful urination were observed. In selected animals’ necropsies, concentrated creamy and sometimes dark red urine were observed (Figure 5). In none of the kidney, urinary bladder, and urinary duct samples in all the studied rabbits, no stones and gravel were observed, even in obese animals (Figure 6).

4. Discussion

Causes of trichophagia in rabbits include low fiber and essential amino acids, high carbohydrates, starches, and sugars in the diet, as well as the presence of any stressors, lack of mobility, decreased water intake, anorexia, and concurrent diseases, especially coccidiosis, enterotoxemia, and E. coli infections (Circella et al., 2021; Mondal et al., 2006; Quesenberry & Carpenter, 2011).

Reducing fiber reduces the movement of ingested substances in the digestive tract. Eating and gastrointestinal motions are interdependent. When movement decreases or stops, stool production also decreases or stops. Therefore, if the rabbit does not eat for only 12 hours, this issue should be investigated urgently (Quesenberry & Carpenter, 2011). In the case of domestic rabbits, carpet fur may also be eaten. In Angora rabbits, trichophagia is much more common and even leads to death (Mondal et al., 2006). Mondal et al. examined the prevalence of trichobezoar in four breeds of angora rabbits in sub-temperate Himalayan conditions in a 5-year study. They found that 28.6% of rabbit deaths were due to trichobezoar and its complications. In that report, in most samples, trichobezoar was a large, single mass in the stomach of rabbits. The stomach contents in affected animals with trichobezoar were watery, and the amount of stool in the intestines was low and very firm. Hairballs obstructed the pyloric valve, and pathomorphological lesions were observed in the stomach, liver, lung, heart, and kidneys.
(Mondal et al., 2006). The crude fiber required in the rabbit diet is 10%-14% (National Research Council Subcommittee on Laboratory Animals, 1995). Digestible and non-digestible fibers are necessary for rabbit nutrition and have metabolic and physical benefits. As in hay, long-strand fibers promote healthy dentition because the plant silicates and large particles keep the constantly growing molar surfaces in proper occlusion. The long fibers also propel ingested fur through the digestive tract and reduce the risk of trichobezoars in the stomach. The larger particles from non-digestible fiber stimulate gut motility and enterocyte turnover (Nowland et al., 2015). Wang et al. published a report on recent developments in the mechanisms of urinary stone formation (Wang et al., 2021). Circella et al. published a report on the occurrence of a 0.5-2 cm stone in the urethra in the preputial sac of a rabbit, which was made of calcium carbonate and was successfully removed by surgery (Circella et al., 2021).

In this study, the stomach was completely full in all necropsied rabbits, indicating adequate appetite and complete eating. No hairballs were observed in the stomach and intestines, even in small sizes. Only a small amount of hair was observed in the contents of the stomach, which was normal and usual. The diet analysis found that the amount of fiber, essential amino acids, calcium and phosphorus, and vitamins, especially vitamin B6, are in the normal range and supply the nutritional requirements of rabbits. Also, the amount of energy in the diet was not more than normal, so the amount of carbohydrates in the diet was not high. All the rabbits had normal mobility, water, and food intake. The amount, size, and strength of rabbits’ daily stools are normal, indicating proper digestive activity. In none of the kidney, urinary bladder, and urinary duct samples, no stones and gravel were observed, even in obese animals. During the examination period, symptoms of anorexia, lethargy, gnashing of teeth, reluctance to move, abdominal distension and pain, pushing the abdomen to the ground, painful urination, and the presence of blood in the urine (hematuria), decreased weight and any special diseases were not observed in rabbits and the environmental conditions of breeding salons were favorable and far from stressors.

5. Conclusion

A balanced diet and supply of nutritional requirements, especially protein, energy, fat, fiber, minerals, amino acids, and various vitamins, as well as the absence of any stressors in breeding environments, has played a key role and has prevented many infectious and non-infectious diseases such as hairballs and urinary stones. The absence of urinary stones in this study could lead to the hypothesis that infection with urease-producing bacteria that cause urinary stones in the studied rabbits was eliminated or non-pathogenic, which could indicate specific pathogen-free animals. However, bacterial and other infectious agent monitoring should be specialized.

Ethical Considerations

Compliance with ethical guidelines

The Ethics Committee of the Razi Vaccine and Serum Research Institute has approved all study protocols.

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Authors’ contributions

Conceptualization, methodology, supervision, project administration, original draft preparation and visualization: Roozbeh Fallahi; Review and editing: Navid Dadashpour Davachi.

Conflict of interest

The authors declared no conflict of interest.

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مقاله پژوهشی
بررسی وجود توپ‌های مویی در دستگاه گوارش و سنگ‌های ادراری در خرگوش‌های آزمایشگاهی

مؤَنّسَه رازی

روزبه فلاحی، ن. نوید داداش پور دوآچی

بخش تحقیقات، تولید و پرورش حیوانات آزمایشگاهی، مؤسسه تحقیقات واکسن و سرم‌سازی رژی، سازمان تحقیقات آموزشی و ترویج کشاورزی کرج، ایران.

زید جمع‌آوری: خرگوش‌ها در طول روز روز علت‌های مختلفی چون کاهش نیروی احیایی چون کاهش نیروی احیایی که ایجاد می‌کند می‌تواند منجر به تجمع مو در معده حیوان باشد. از طرفی، خرگوش‌ها مستعد تشکیل سنگ‌های ادراری هستند.

این مطالعه به منظور بررسی وجود توپ‌های مویی و سنگ‌های ادراری در خرگوش‌های آزمایشگاهی مؤسسه رازی انجام شد.

هدف این مطالعه مانند بررسی وجود توپ‌های مویی و سنگ‌های ادراری در خرگوش‌های آزمایشگاهی مؤسسه رازی انجام شد.

روش کار: در طول 1 سال، کلینیک خرگوش‌های آزمایشگاهی در زمان انجام آزمایش سلامتی در بخش پزشکی حیات وحشی مؤسسه رازی به دستگاه گوارش و دستگاه ادراری مورد بررسی قرار گرفت. از نظر وجود مو و توپ‌های مویی و در نظر هرگونه سنگ بهداشتی در کلینیک خرگوش‌های آزمایشگاهی موی و سنگ‌های ادراری مشاهده نشده.

نتایج: در هیچ‌کدام از آزمایشگاه‌ها، وجود سنگ‌های ادراری مشاهده نشده. در نظر بودن مقدار مویی که در آن‌ها مشاهده شده، احتمال وجود سنگ‌های ادراری بسیار امروزه بالا و در بیشتر آزمایشگاه‌های آزمایشگاهی محقق است.

نتیجه‌گیری: در این مطالعه، مشاهده علائم سلولی و سنگ‌های ادراری در خرگوش‌های آزمایشگاهی نمایان نبود. این نتایج نشان می‌دهد که به وسیله نیروی احیایی که در خرگوش‌های آزمایشگاهی موجود است، معده در تمام نمونه‌ها پر می‌باشد که نشان می‌دهد که خرگوش‌ها به خوبی غذا می‌خورند و در روده‌ها گاز و نقاط پرخون و یا خونریزی مشاهده نشده. مقدار و کربن مصرف در روده‌ها به مقدار غذای مصرف شده معادل است. در هیچ‌کدام از نمونه‌ها، توپ‌های مویی مشاهده نشده است.

مقدار مویی که در معده‌ها مشاهده شده، احتمالاً باعث بهبود در بهبود سلولی و معده می‌شود. به بینی‌گیری نیروی احیایی که در بدن وجود دارد، معده در تمام نمونه‌ها پر می‌باشد و در روده‌ها گاز و نقاط پرخون و یا خونریزی مشاهده نشده.

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