

## Zinc Phosphide Intoxication in Three Horses: A Case Report

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### Abstract

Three adult crossbred Thoroughbred horses died in June 2018 at a ranch near Hamedan, Iran. Rigor mortis was evident with no signs of post-mortem bloating in the carcasses. The neck muscles appeared excessively cyanotic in necropsy. In addition, widespread petechiae were observed on the inner surface of the thoracic intercostal muscles. Dropsy was visible in the inner surface of the abdominal wall and peritoneum. The stomach content was watery and dark gray. Furthermore, gastric mucosa was also gray. The livers had a dark appearance and the adjacent duodenum was gaseous and blackened. The kidneys showed autolysis with areas of medullary hyperemia. In addition to decomposition, a specific odor similar to the garlic aroma or natural gas could be smelled from the carcasses. Tissues and contents of various organs, as well as food and water samples, were collected for diagnostic tests. Preliminary toxicological studies showed detectable amounts of phosphide in the samples collected from gastric contents of the animals, and complementary tests confirmed the presence of zinc phosphide in the concentrate feed.

**KEYWORDS:** Equine, Histopathology, Phosphine, Poison, Rodenticide

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### Case History

On June 28, 2018, the sudden death of three 8-year-old crossbred Thoroughbred horses, including two mares and one stallion was reported from a barn at Qerghler, a village in Hamedan, Iran. The animals were necropsied and thoroughly examined by a large animal internal medicine specialist shortly after the notification call. According to the farm owner's

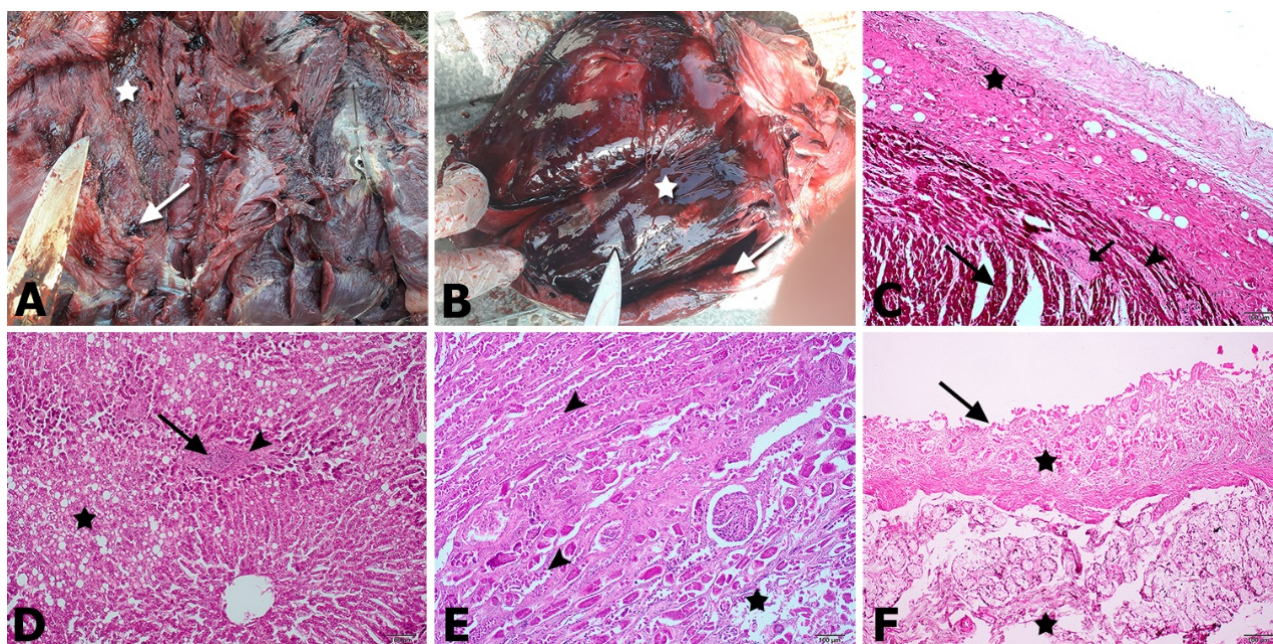
declaration and based on the daily inspections of the farm guard, the animals had probably died during the previous evening.

### Clinical Presentations

The dead horses were laid on the left lateral sides with no signs of post-mortem bloating.

No signs of trauma, contusion, subcutaneous inflammation, and emphysema were observed or palpated. Complete rigor mortis was reversing with the loosening of the proximal part of the neck. There was no bleeding and/or fluid discharge from the natural orifices. The ocular conjunctivas were hyperemic and edematous. Moreover, the neck muscles appeared cyanotic with obvious venous congestion. The inner surface of the abdominal wall and parietal peritoneum were edematous. Furthermore, hypostasis was evident on the left side of all the internal organs in all carcasses. In addition to the odor caused by the decomposition of the carcasses, a specific odor similar to the smell of natural gas or fresh garlic was noticeable.

Although the lungs were normal in size, numerous spots of pinpoint petechial hemorrhage were observed on the neighboring inner surface of the inter-costal muscles. Black gray discolorations were found in the tracheal mucosa. Hearts were normal in terms of size and texture. However, the superficial veins of the hearts were congested along with the imbibition of blood into the surrounding myocardial tissues. The heart muscles were hyperemic with the ventricles containing dark unclotted blood with blood imbibition of the endocardium ([Figure 1](#)). The medulla of moderately autolyzed kidneys was slightly hyperemic.



**Figure 1.** Gross (A, B) and microscopic view (C-F) of organs of horses poisoned by zinc phosphide. **A)** Skeletal muscles of the neck appear cyanotic (asterisk) with profound venous congestion (arrow). **B)** Myocardium of the heart shows hyperemia (arrow), ventricles contain dark unclotted blood and endocardium tinged with blood (asterisk). **C)** Epicardium of the heart appear hyperemic (asterisk), myocardial fibers show necrosis (large arrow) and arterial (small arrow) and venous (arrow-head) blood vessels are congested. **D)** Liver demonstrates severe hepatocellular necrosis (asterisk) with portal venous congestion (arrow) and mild perivascular filtration of leukocytes (arrowheads). **E)** Kidney shows interstitial edema (asterisk) and diffuse coagulative necrosis of the renal tubular cells (arrows). **F)** Mucosa and submucosa of stomach show severe inflammation; edema and necrosis (asterisks) with sloughing of the gastric epithelium (arrow). C-D  $\times 100$ , Hematoxylin& Eosin.

The stomach contents were gray, runny, and odorous, and the gastric mucosa was deeply gray. Next to the blackly discolored liver, the duodenum was heavily darkened and gaseous.

Death was estimated to have occurred 12-18 h before based on rigor mortis and hemoglobin imbibition around the heart veins. According to the necropsy findings, acute heart failure due to gastrointestinal poisoning was probably the etiologic cause of the death of the animals.

Following dissection, appropriate tissue specimens were collected from the hearts, livers, kidneys, lungs, spleens, gastrointestinal tracts, and neck muscles and were sent for histopathological examination. In addition, gastric contents, tissue samples of liver and kidney, as well as drinking water (filled in a capped bottle) and consumed food (concentrate and forage separately) were collected for toxicological analysis. The metacarpal bone was collected aseptically and submitted for microbiological examination.

## **Diagnostic Testing**

### **Toxicology**

The stomach contents of horses were evaluated for the presence of phosphine (PH<sub>3</sub>) using silver nitrate-impregnated strips (Bumrah *et al.*, 2015). The discoloration and darkening of the test strips confirmed the presence of a detectable amount of PH<sub>3</sub> in the samples. Toxicological analysis revealed zinc phosphide as the toxic agent. The zinc content in the concentrated sample of the manger was measured as 962 ppm.

### **Histopathology**

Severe inflammation of gastric mucosa and submucosa, as well as the diffuse necrosis of the gastric epithelium, submucosa, and muscularis propria, were observed. The liver was severely inflamed with extensive centrilobular and periportal necrosis, sinusoidal and portal

vascular congestion along mild perivascular leukocytes filtration. Despite the noted findings, the destruction of liver tissue over time made diagnosis difficult. In the heart sections, pericardial congestion, interstitial connective tissue edema, and leukocytes infiltration with mild to moderate necrosis of myocardial fibers were observed. However, moderate tissue destruction and mild coagulative necrosis of cardiac muscle cells were noted in the heart of one of the mares. Myofibrillar necrosis and connective tissue inflammation along with moderate tissue destruction and putrefaction were found in the skeletal muscles. Moreover, cloudy swelling and diffuse coagulative necrosis of renal tubular cells with interstitial edema were observed in the kidneys. In the lungs, interstitial and alveolar edema, diffuse hyperemia, and hemorrhage of the interstitium, and a mild neutrophilic infiltration were noted ([Figure 1](#)).

### **Assessments**

The results of the silver nitrate test for the presence of PH<sub>3</sub> in the contents of the stomach of horses and the subsequent measurement of zinc in the feed concentrate confirmed zinc phosphide intoxication. The diagnosis of this poisoning usually takes place based on clinical signs, history of exposure to zinc phosphide, and toxicological findings. The results of blood biochemical tests, necropsy, and histopathological examination are nonspecific (Plumlee, 2001). However, the necropsy and histopathological findings of the present report were consistent with few previous reports of zinc phosphide intoxication in horses (Ingram, 1945; Drolet *et al.*, 1996; Olivares *et al.*, 2002; Bazargani *et al.*, 2008).

Zinc phosphide is a relatively old and inexpensive rodenticide that has been used since the 1910s. Cases of intentional and accidental poisoning with zinc phosphide have been reported in humans and animals (Gupta, 2018). Zinc phosphide is hydrolyzed in aqueous media and

releases  $\text{PH}_3$ , which is an absorbable toxic gas and has a specific odor similar to garlic, rotten fish, or natural gas. The low pH of the gastric environment accelerates zinc phosphide conversion to  $\text{PH}_3$  (Plumlee, 2001; Gupta, 2018). It has been shown that  $\text{PH}_3$  inhibits cellular respiration in mitochondria (Sciuto *et al.*, 2016). The toxic dose of zinc phosphide in horses remains unknown. However, the lethal dose in other domestic species has been reported to be 20-40 mg/kg bw. Phosphide is a stomach irritant and often causes vomit. Horses are unable to vomit the food already taken. As a result, they cannot even tolerate doses lower than those the other animals can do (Plumlee, 2001).

Clinical signs of acute zinc phosphide poisoning usually occur within 15 min to 4 h after oral uptake and might be even delayed for up to 18 h (Plumlee, 2001; Fox *et al.*, 2018; Nagy *et al.*, 2019). Acute symptoms reported in horses include colic, sweating, muscle tremors, incoordination, respiratory distress, cardiac arrhythmia, and depression (Ingram, 1945; Drolet *et al.*, 1996; Olivares *et al.*, 2002; Bazargani *et al.*, 2008; Nagy *et al.*, 2019). Tremors and involuntary movements of hind limbs due to neural stimulation of motor organs have been reported as clinical manifestations in some horses (Drolet *et al.*, 1996; Bazargani *et al.*, 2008; Nagy *et al.*, 2019). Subsequently, the horse lies down and may have convulsions prior to death. The  $\text{PH}_3$  gas at concentrations of 7 ppm can cause severe poisoning (Plumlee,

2001). Nasogastric intubation can help discharge the toxic gas and may pose the risk of poisoning to clinical staff, especially in closed confined spaces (Drolet *et al.*, 1996).

Although no specific antidote for gastrointestinal intoxication with phosphine has been reported yet (Plumlee, 2001; Gupta, 2018; Constable *et al.*, 2016), gastric lavage of poisoned horses in the early stages would help them to survive (Easterwood *et al.*, 2010). In addition, oral administration of 2% sodium bicarbonate solution can be useful in decelerating the release of  $\text{PH}_3$ . Furthermore, supportive therapies are of value; for example, intravenous administration of fluids can help stabilize the cardiovascular system and counteract acidosis. In addition, intravenous administration of dextrose and oral corn syrup has been used to maintain blood glucose levels (Easterwood *et al.*, 2010; Nagy *et al.*, 2019).

After a detective investigation based on the above-mentioned findings, the offender confessed to intentional poisoning.

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## **Conflict of Interest**

None.

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## مسمومیت با فسفید روی در سه راس اسب: گزارش بالینی

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در تیرماه سال ۱۳۹۷ تعداد ۳ راس اسب بالغ دو خون در محل یک دامداری در نزدیکی همدان تلف شدند. در بازرسی لاشه‌ها جمود نعشی رخ داده بود ولی نشانه‌ای از نفخ پس از مرگ مشاهده نشد. در کالبدگشایی، عضلات ناحیه گردن لاشه‌ها پرخونی سیاه‌رنگی را نشان می‌داد. به علاوه، در عضلات بین دنده‌ای در سطح داخلی قفسه سینه نیز نقاط خونریزی سرسوزنی فراوان مشاهده شد. ادم در سطح داخلی دیواره شکم و پرده صفاقی وجود داشت. محتویات معده هر سه اسب به شدت آبکی و مخاط آن‌ها به رنگ خاکستری بود. کبدها به وضوح سیاه رنگ بودند و دوازدهه در مجاورت کبد پر گاز و به شدت سیاه رنگ شده بود. کلیه‌ها تا حدی اتولیز را نشان می‌دادند و در بعضی نقاط آن‌ها اندکی پرخونی مرکزی مشاهده شد. علاوه بر بوی ناشی از فساد، بوی خاصی شبیه به بوی سیر یا گاز شهری از لاشه‌ها استشمام می‌شد. نمونه‌های بافتی و محتویات چند عضو برای آزمایش‌های تشخیصی جمع‌آوری شدند. بررسی‌های اولیه سم‌شناسی مقادیر قابل شناسایی فسفین را در نمونه‌های محتویات معده حیوانات تلف شده نشان داد و آزمایش‌های تکمیلی، نوع سم موجود در کنسانتره این اسب‌ها را فسفید روی تایید کرد.

**واژه‌های کلیدی:** تک‌سمی، هیستوپاتولوژی، فسفین، سم، جونده‌کش