Outbreak of avian botulism in a backyard poultry farming

Surto de botulismo aviário em criação de aves domésticas

ABSTRACT
The purpose of this study is to report an outbreak of avian botulism in backyard poultry farming. In 2019, a botulism outbreak in a flock of laying hens was investigated in Brazil. In the flock of 30 hens, clinical signs of botulism occurred after they ate decaying vegetables. A type C botulism outbreak was confirmed using the mouse lethality assay for detection of botulinum toxin in serum and ELISA test to detect *Clostridium botulinum* in intestinal contents and serum. Botulism in laying hens has rarely been reported. The chickens developed cyanotic comb and wattle, dyspnea, different degrees of flaccid paralysis in the neck, and detachment of feathers. No macroscopic lesions were observed, as were microscopic findings. The chicken's serum was neutralized by C antitoxin, confirming the botulism diagnosis, and also toxin was detected in intestinal contents.
Keywords: botulinum toxin, organic matter, poultry

RESUMO
O objetivo deste estudo é relatar um surto de botulismo aviário na avicultura de subsistência. Em 2019, foi investigado um surto de botulismo em um rebanho de poedeiras no Brasil. Em uma criação com 30 galinhas, os animais iniciaram sinais clínicos característicos de botulismo depois que ingerirem vegetais em decomposição. Um surto de botulismo do tipo C foi confirmado inoculando o soro de suspeitas no ensaio de letalidade em camundongos para detecção de toxina botulínica e teste ELISA para detectar Clostridium botulinum a partir do conteúdo intestinal e do soro. O botulismo em galinhas poedeiras é raramente relatado e as aves o relato desenvolveram crista cianótica e pálida, dispneia, diferentes graus de paralisia flácida do pescoço e desprendimento das penas. Não foram observadas lesões macroscópicas, assim como achados microscópicos. Durante teste laboratorial o soro das galinhas foi neutralizado pela antitoxina C, confirmando o diagnóstico de botulismo, e também foi detectada toxina no conteúdo intestinal.

Palavras-chave: toxina botulínica, matéria orgânica, galinhas

1 INTRODUCTION
Clostridium botulinum is a spore-forming anaerobic bacterium causing avian and mammalian botulism outbreaks (Prisilla et al., 2017). Avian botulism is an emerging disease in wild and domestic birds caused by ingestion of botulinum toxin produced by Clostridium botulinum (C. botulinum). Since 1917, botulism was also reported in chickens, turkeys, pheasants, ducks, and peafowls (Dohms, 2003). Clinical signs of botulism are similar both in domestic fowls and wild birds and consist of flaccid paralysis of the neck, wings, and eyelids. Clinical signs and mortality occur within hours or 1–2 days depending on the toxin doses (Son et al., 2018). Type C, particularly, and D also are responsible for most cases of avian botulism. Several outbreaks of type C botulism in wild species have been reported in several countries (Silva et al., 2018). The animal develops flaccid paralysis, reported in chickens (Gallus gallus domesticus), ducks (Cairina moschata) guinea fowl (Numida meleagris).

2 MATERIALS AND METHODS
Six hens had dead and four present classic signs of botulinum toxin poisoning from a backyard poultry farming with approximately 58 chickens 42-week-old. In the same week of the event, many vegetables were provided, the leftovers of the food remained in the hen house, even in deterioration, and chickens ingested this organic matter. Thus, four chickens were euthanized and necropsied and organ fragments were collected and fixed in 10% buffered formalin and processed for paraffin inclusion and hematoxylin-eosin staining (HE), serum aliquots and contents found in
the gizzard and ingluvium were collected as well to detection of botulinum toxin, for bioassay in mice, and ELISA test (Enzyme-Linked Immunosorbent Assay).

3 RESULTS

The farmer reported that the four sick’s hens developed cyanotic comb and wattle, dyspnea, different degrees of flaccid paralysis in the neck, and detachment of feathers after ingesting decaying vegetables. The mortality rate was observed at 10.3% (6/58), excluding birds that were euthanized. During a necropsy, no macroscopic lesions were observed, as were microscopic findings. The chicken's serum was neutralized by C antitoxin, confirming the botulism diagnosis. The toxin was detected also in contents found in the gizzard and ingluvium.

4 DISCUSSION

Botulism is a serious illness caused by Clostridium botulinum toxins, which should be considered an animal health emergency (Machado et al., 2010). Several botulism outbreaks in wild species have been previously reported as type C botulism.

Avian feces may have previously contaminated vegetables indicated as botulinum toxin carriers in the present study since C. botulinum is present in the digestive tracts of different bird species (Hardy et al., 2013). The anaerobic environment and decaying organic matter from decaying vegetables may have provided the bacteria to produce the toxin. Inadequate food (decaying); high water temperatures which pH between 7.5 and 9 and low redox potential (Eh); and animal carcasses are botulism risk factors (Vidal et al., 2013). These are considered botulism outbreaks risk factors because of the initial multiplication and toxinogenesis of C. botulinum (Sánchez, 2013). Is necessary to avoid the occurrence of these risk factors, to prevent avian mortality and disease, including endangered species (Anza et al., 2014).

The mortality rate observed here (10.34%) was lower than orders botulism outbreaks. The mortality rate of 20% in a flock of 33% in a flock of 29-week-old hens (7,500 hens) in Ireland (Shape et al., 2011) and of 30-week-old hens (12,800 hens) was reported in Sweden (Skarin et al., 2013).

It has been demonstrated that the susceptibility of broilers decreased with age (Dohms & Cloud, 1989). The outbreak reported here occurred at 42 weeks. This age difference could explain the lower mortality rate.

A botulism outbreak then occurred rapidly and was characterized by flaccid paralysis and sudden mortality of the birds (Circella et al., 2019). The bacteria produce toxins that damage the chicken's nervous system leading to paralysis with affected birds displaying signs such as reluctance.
to walk and apparent lameness, paralyzed neck, with some loss of feathers in this area (Souillard et al., 2017) as we identified in the hens of this study.

The toxin is a zinc metalloprotease that consists of a light chain, heavy chain, and translocation domain. The cleavage of these proteins prevents the release of acetylcholine into the neuronal muscular junction and results in a flaccid paralysis known as botulism (Hill et al., 2015; Popp et al., 2012).

Anamnestic data and clinical signs are necessary but lead to a presumptive diagnosis of botulism. Other analyzes are necessary, such as necropsy and histopathology. In the suspected botulism cases, it is important to carry out these two analyzes to exclude other diseases, since macro and microscopic lesions are not present. No macroscopic lesions were observed but other studies also were visualized generalized congestion of the organs (Hill et al., 2015; Sato et al., 2016). However, in general, definitive diagnosis, which is required to advise on recurrence risks, is essential analysis such as MLA, ELISA test, and Polymerase Chain Reaction (PCR) (Circella et al., 2019). The definitive diagnosis in that outbreak was supported with MLA and ELISA test, the toxin identified as responsible for the disease outbreak was type C.

5 CONCLUSIONS

Against epidemiological aspects related to access and ingestion decaying organic matter, cases of deaths with acute evolution, development of flaccid paralysis, absence of macro and microscopic lesions, and detection of toxin in the serum of the animals, it is concluded that the access decaying vegetables is a botulinum toxin possible infection source.
REFERENCES


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