



Case Report

**Debilitating Cutaneous Poxvirus Lesions
on Two Captive Houbara Bustards
(*Chlamydotis undulata*)**

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SUMMARY. Poxvirus was isolated from cutaneous nodules on two young Houbara bustards (*Chlamydotis undulata*) bred in captivity in Saudi Arabia. Birds were emaciated and presented nodules on tibiotarso-tarsometatarsal joints, toes, and the carpal joint. Diagnosis was confirmed by histopathology, virus isolation on inoculated chorioallantoic membranes of embryonated chicken's eggs, and electron microscopy. Progressive leg lesions were extensive and interfered with walking, significantly debilitating the birds. Successful excisions of these lesions were performed.

RESUMEN. *Reporte de Caso*—Lesiones cutáneas debilitantes por virus de viruela en dos hubaras (*Chlamydotis undulata*) mantenidas en cautiverio.

El virus de viruela fue aislado de nódulos cutáneos en dos hubaras (*Chlamydotis undulata*) mantenidas en cautiverio en Arabia Saudita. Las aves se mostraron emaciadas y con nódulos presentes en las articulaciones tibiotarsal y tarsometatarsal, en los dedos y en la articulación del carpo. El diagnóstico fue confirmado por histopatología, aislamiento viral en huevos embrionados inoculados en la membrana corioalantoidea y por microscopía electrónica. Las lesiones progresivas de las extremidades inferiores se hicieron extensivas hasta provocar alteración de la marcha y debilitamiento significativo de las aves. La remoción quirúrgica de las lesiones fue exitosa.

Abbreviation: CAM = chorioallantoic membrane

The Houbara bustard (*Chlamydotis undulata*) is a medium-sized bustard of slender appearance, measuring 55–65 cm and weighing 900–2400 g. This bird has been classified as vulnerable by the International Union for Conservation of Nature and Natural Resources (6). Some populations are thought to be seriously declining. This bird is one of the major concerns of the National Commission for Wildlife Conservation and Development in Saudi Arabia. A captive breeding program was initiated in 1986, with the aim of restoring wild populations. Immature birds intended for reintroduction are closely monitored. Although absolute assurances are difficult to achieve, the aim of

the veterinary management is to reduce the likelihood of accidentally translocating diseases to potential wild hosts.

The cutaneous form of avian pox infection has been reported in many bird species (8). The present report concerns the first isolation of poxvirus from a captive Houbara bustard with debilitating pox lesions affecting cursorial locomotion.

MATERIALS AND METHODS

Case history. In July 1993, two 4-month-old bustard chicks born in captivity were found to have cutaneous nodular lesions on the tibiotarso-tarsome-



Fig. 1. Pox lesions on a juvenile Houbara bustard tibiotarso-tarsometatarsal joint.

tatarsal joints of both legs. Cutaneous nodules were also found on toes, on areas of the skin, and on the carpal joint. Lesions were elevated, closed, warm, and relatively smooth when palpated. Development of the disease was subacute, with progressive swelling of nodules. After 2 weeks the necrotic tops of the nod-

ules on the toes and wings could be removed, leaving bleeding, swelling erosions that dried out progressively. Leg lesions did not diminish in size and developed into large semipendulant masses (30 mm wide by 41 mm high by 40 mm long for the largest one) (Fig. 1). After one month birds were unable to walk

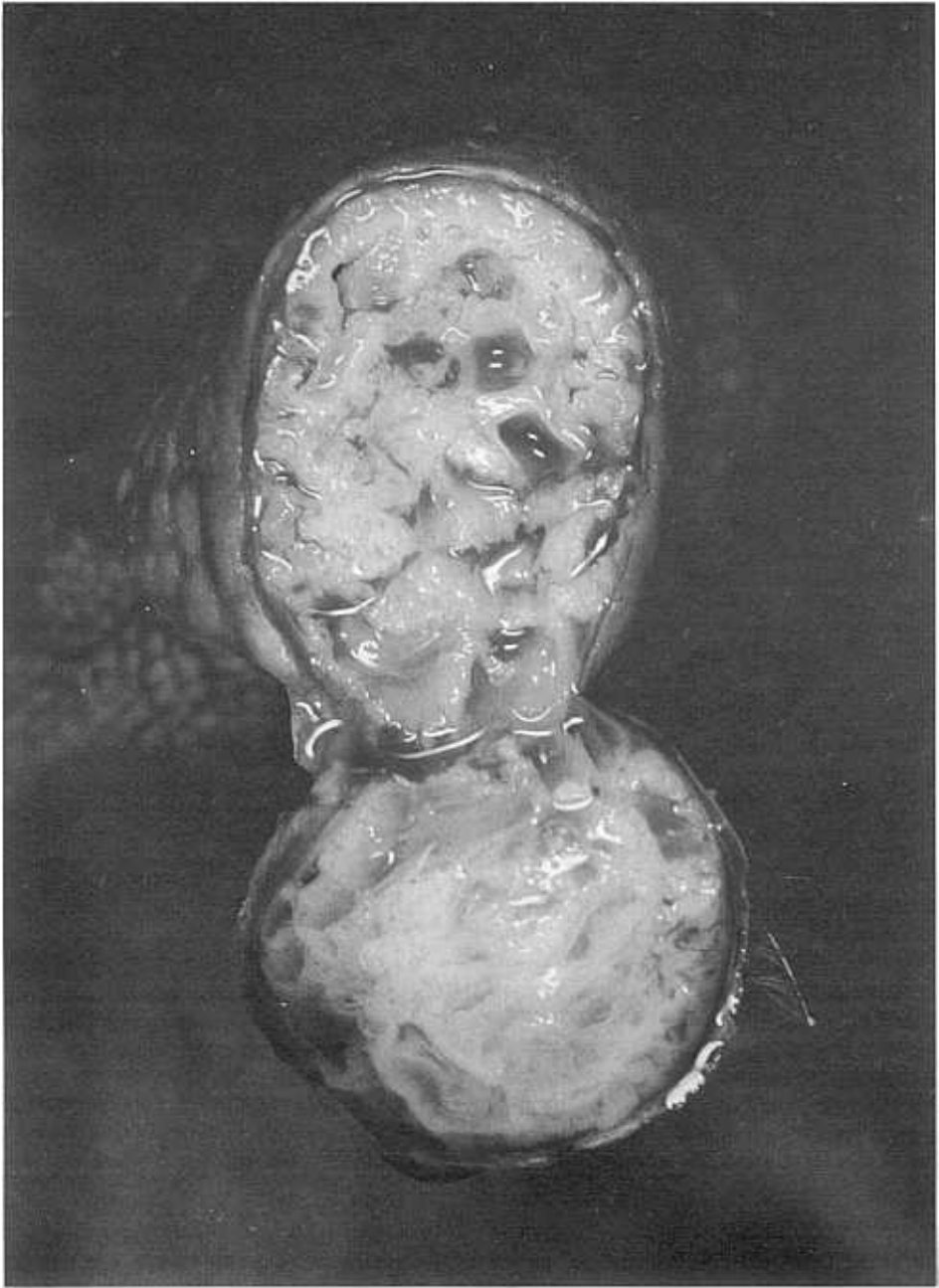


Fig. 2. Cut surface of a debilitating cutaneous pox lesion on the tibiatarso-tarsometatarsal joint of a juvenile Houbara bustard.

or reach their food. As no signs of regression were observed, surgical excisions were carried out to restore locomotion.

Laboratory procedures. Chosen samples of excised cutaneous lesions were fixed in 10% buffered formalin, embedded in paraffin, sectioned at 5 μm ,

and stained with hematoxylin and eosin. Swabs taken from the lesions were incubated aerobically on Columbia blood agar (Oxoid, Basingstoke, England) and MacConkey agar (Difco, Detroit, Mich.) at 37 C for 72 hr. Fresh excised tissues were frozen for virological examination. These samples and the formalin-fixed

material were shipped to the Department of Veterinary Pathology, Section of Diseases of Laboratory and Special Animals, of the Veterinary Faculty of Utrecht University, The Netherlands, for examination. Poxvirus was isolated from the skin samples and propagated in chicken's eggs. Seven days after infection the chorioallantoic membranes (CAM) were harvested. For electron microscopy, the CAM were ground in a mortar with sterile sand and distilled water. The homogenate was clarified by centrifugation at $200 \times g$ for 30 min. Virus particles in the supernatant were absorbed onto carbon-coated collodion film mounted on 400-mesh nickel grids for 10 min. After washing with distilled water the grids were stained with 2% phosphotungstic acid, pH 6.8. The grids were examined with a Philips CM 10 electron microscope. The homogenate was also used for an agargel-precipitation test against antibodies to fowl pox and canary poxvirus.

RESULTS AND DISCUSSION

No bacteria were isolated from pox lesions. Macroscopically, the cut surface of excised cutaneous lesions consisted mostly of a yellowish caseous necrotic mass. Tibiotarsal nodules were smooth, solid, and covered by skin. In cross-section the lesions consisted of central areas of caseous necrosis surrounded by a thick and firm capsule (Fig. 2). Histologically, the lesions consisted of a massive proliferation of the ectodermal epithelium and edema in the interepithelial connective tissue. In the cytoplasm of the swollen and vacuolized epithelial cells, typical Bollinger bodies were observed. Intracytoplasmic inclusion bodies characteristic of poxvirus were observed microscopically 7 days postinoculation in the CAM. Numerous pox virions were observed with the electron microscope in the negatively stained preparations from infected CAM (Fig. 3). An agargel-precipitation test carried out on homogenates of CAM preparations revealed a positive precipitation with canary poxvirus antibodies and no precipitation with fowl pox antibodies. Immunoprecipitation has been used for differential identification of fowl and pigeon poxviruses from those of other avian viral diseases (11). A substantial degree of host specificity exists among some avian poxviruses, especially those that infect wild birds (10). However, an antigenic proximity might exist between the strain isolated in the Houbara bustard and the canary strain of poxvirus.

In some cases the location and/or size of the pox lesion can cause severe debilitation. In gen-

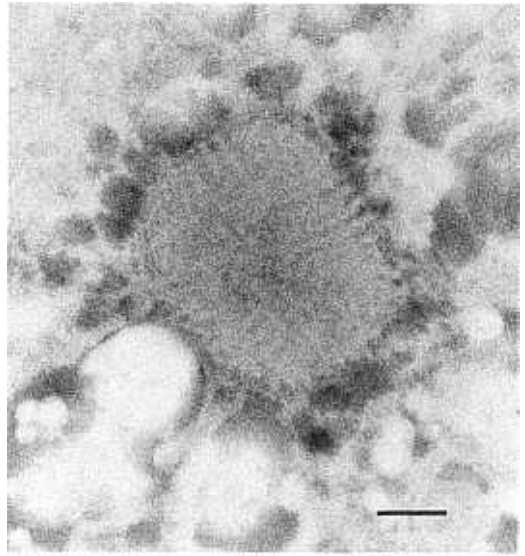


Fig. 3. Electron micrograph of the poxvirus from a Houbara bustard grown on the chorioallantoic membrane of an embryonated chicken egg. Phosphotungstic acid. Bar = 100 nm.

eral, a preponderance of pox lesions on the eyes may contribute to death because the birds may not be able to find food or escape predators (2). Lesions affecting legs are rarely debilitating (7). In Houbara bustards, however, progressive emaciation typical of poxvirus infection, as well as a mechanical incapacity to perform full extensions of both legs contributed to their inability to walk normally. Houbara bustards are primarily cursorial (3). Thus, in the wild, food is normally collected by walking from plant to plant and searching for suitable items. Although no pox lesions have been described on wild Houbara bustards, it can be assumed that the physical presence of large lesions on the tibiotarsal-tarsometatarsal joints of wild birds could be severe enough to cause fatal debilitation.

Poxvirus infections may occur in stable, captive avian populations if the disease is regularly brought in by free-ranging wild birds or insect vectors (1). Avipoxvirus is not capable of penetrating intact epithelia (5); other vectors such as biting insects are necessary. Mosquitos were present during the period of the year when this disease occurred in the Houbara bustard chicks (July), and could be responsible for transmission.

In general, surgical excision of developing pox lesions is not recommended as they are known to regress naturally. In the present case excision allowed the restoration of movement in the tibiotarso-tarsometatarsal joint. Lesions on toes and wings regressed as they also do in some fowl pox-infected chickens (7), with the recovered birds being immune (4) as they mature into adulthood. Persistent cutaneous lesions in wild birds (8) and latent infections in poultry flocks (9) have been demonstrated.

In order to prevent accidental translocation of the disease to the wild population, the two birds were not introduced into the wild. Eventually further studies will clarify the degree of antigenic proximity between the strain isolated from these Houbara bustards and the canary pox strain.

REFERENCES

1. Clubb, S. L. Avian pox in cage and aviary birds. In: Zoo and wild animal medicine. M. E. Fowler, ed. W. B. Saunders Co., Philadelphia. pp. 213-220. 1986.
2. Docherty, D. E., R. I. R. Long, E. L. Flickinger, and L. N. Locke. Isolation of poxvirus from debilitating cutaneous lesions on four immature grackles (*Quiscalus* sp.). *Avian Dis.* 35:244-247. 1991.
3. Etchecopar, R. D., and F. Hue. L'outarde houbara (*Chlamydotis undulata*). In: *Les oiseaux du Nord de l'Afrique*. N. Boubée et Cie, Paris. pp. 202-203. 1964.
4. Fenner, F., P. A. Bachmann, E. P. J. Gibbs, F. A. Murphy, M. J. Studdert, and D. O. White. *Veterinary virology*. Academic Press, Orlando, Fla. p. 404. 1987.

5. Gerlach, H. Viral diseases. In: *Clinical avian medicine and surgery*. G. J. Harrison and L. R. Harrison, eds. W. B. Saunders Co., Philadelphia. pp. 408-433. 1986.

6. IUCN. IUCN red list of threatened animals. IUCN, Gland, Switzerland and Cambridge, England. 1990.

7. Karstad, L. Pox. In: *Infectious and parasitic diseases of wild birds*. J. W. Davis, R. C. Anderson, L. Karstad, and D. O. Trainer, eds. Iowa State University Press, Ames, Iowa. pp. 34-41. 1971.

8. Kirmse, P. Pox in wild birds: annotated bibliography. *J. Wildl. Dis.* 3:14-20. 1967.

9. Tripathy, D. N., and L. E. Hanson. Immunity to fowlpox. *Am. J. Vet. Res.* 36:541-544. 1975.

10. Tripathy, D. N. Pox. In: *Diseases of poultry*. B. W. Calnek, H. J. Barnes, C. W. Beard, W. M. Reid, and H. W. Yoder, Jr., eds. Wolfe Publishing Ltd., London, England. pp. 583-596. 1991.

11. Uppal, P. K., and P. R. Nilakantan. Studies on the serological relationships between avianpox, sheep pox, goat pox and vaccinia viruses. *J. Hyg. Camb.* 68: 349-358. 1970.

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