



NOTE

Pathology

Symmetric undivided diplopagus with cardiac malformation in a Japanese wild boar (*Sus scrofa leucomystax*)

Tomoki NAKAMURA¹), Yuji SUNDEN¹)*, Tomohiro IMAGAWA²), Takeshi TSUKA²), Kenichi SHIRAIISHI³) and Takehito MORITA¹)¹Laboratory of Veterinary Pathology, Joint Department of Veterinary Medicine, Tottori University, 4-101, Minami, Koyama, Tottori 680-8553, Japan²Laboratory of Veterinary Diagnostic Imaging, Joint Department of Veterinary Medicine, Tottori University, 4-101, Minami, Koyama, Tottori 680-8553, Japan³Hunting Club of Tottori Prefecture, 68-1, Mitani, Hino, Tottori 689-4502, Japan*J. Vet. Med. Sci.*

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ABSTRACT. A captured Japanese wild boar (*Sus scrofa leucomystax*) fetus was dicephalic. The fetus had two heads, but one body from the cranial neck region. Computed tomography imaging revealed that the two crania merged at the occipital bone, and the vertebral bodies between the atlas and the seventh thoracic vertebra were deformed. The fetus was found to have two tongues and laryngopharynges, but its esophagus and trachea were not duplicated. Each head contained a cerebrum and cerebellum, but the brains merged at the obex of the medulla oblongata, and the cervical spinal cord had duplicated ventral clefts. The heart was composed of three atria and four ventricles. This is the first report of a dicephalus with cardiac malformation in a wild boar.

KEY WORDS: dicephalus, diplopagus, wild boar

Diplopagus (conjoined twins) is a congenital anomaly that arises in monozygotic twins at an early developmental stage [9]. Several types of diplopagus have been reported, which are classified according to whether they are separated or attached, and symmetric or asymmetric, and according to their site of attachment [2, 3]. Dicephalus is an attached and symmetric-type malformation involving duplication of the head [2]. In this report, we describe a dicephalic fetus in a captured Japanese wild boar (*Sus scrofa leucomystax*).

A malformed fetus was found in the uterus of a wild boar captured in Tottori Prefecture, Japan, on 9 May, 2018. The wild boar was captured using a binding trap as part of a program for the extirpation of injurious animals in Tottori Prefecture. The dead malformed fetus was taken to the Veterinary Medical Center of Tottori University. The fetus was a symmetric undivided diplopagus with two heads (Fig. 1). It was 18.3 cm long, weighed 346.9 g, and was estimated to be at 50–51 days of gestation (Tsuji *et al.*, Joint Meeting of the Primate Society and the Mammal Society of Japan, Okayama, Japan, Sep 2013, poster presentation, P-170). No littermates were identified. Macroscopically, the fetus had two heads that joined in the cervical region. Each head had one snout, one nasus, a pair of eyes, and a pair of ears. The fetus was subjected to computed tomography (CT) imaging, then immersed in 10% neutral-buffered formalin. Subsequently, it was dissected and the major organs, including the brains and the spinal cord, were fixed in 10% neutral buffered-formalin. The organs were embedded in paraffin and cut into 3–4 μ m thick sections, which were stained with hematoxylin and eosin (HE).

The CT images showed that each head had separate maxillae and mandibles, but shared a spinal column and axial skeleton. The two skulls were fused at the level of the occipital bone and shared a foramen magnum (Fig. 2). In the cervical spinal column, there were no spinous processes, and the fourth and fifth thoracic spinous processes were fused (Fig. 2). In addition, the spinous processes after the tenth thoracic spine were missing. The vertebral bodies from the atlas to the seventh thoracic vertebra were composed of two nodular tori, but the right side of the fourth thoracic vertebral body was not apparent (Fig. 3).

The oral cavity of each head contained a cleft palate, a tongue, and a laryngopharynx, but there was no duplication of the esophagus and trachea. The mucosal surface of the ventral midline of the trachea was slightly raised. Dissection of the skull bones revealed duplication of the encephalon. Each head contained an olfactory bulb, a pituitary gland, a cerebrum, a cerebellum, a mesencephalon, and a pons. The brains merged at the level of the obex of the medulla oblongata. Although the appearance of the spinal cord was unremarkable, the couple of cleft-like structures were observed in the ventral part of the cervical spinal cord on the cross-section. Inspection of the thoracic and abdominal organs revealed normal anatomic structure, except in the case of the

*Correspondence to: Sunden, Y.: sunden@tottori-u.ac.jp

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Fig. 1. Gross appearance of the fetus. Formalin-preserved. Bar=2 cm.

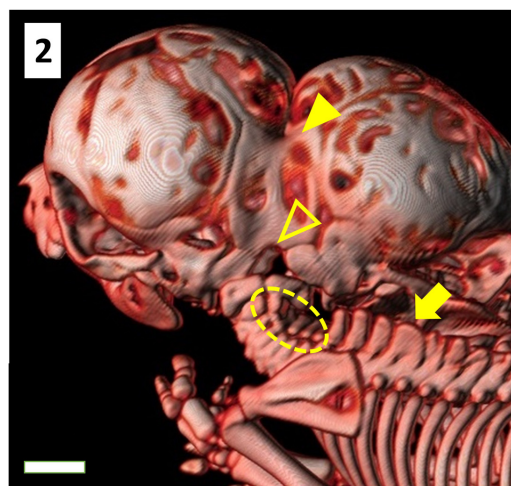


Fig. 2. CT reconstruction of the left caudal surface of the crania and spinal column. The two cranial bones are fused at the level of the occipital bone (arrowhead) and form a single foramen magnum (open arrowhead). From the axis to the seventh cervical spine, the spinous processes are missing (dashed line). The spinous processes of the fourth and fifth thoracic spine are fused (arrow). Bar=1 cm.

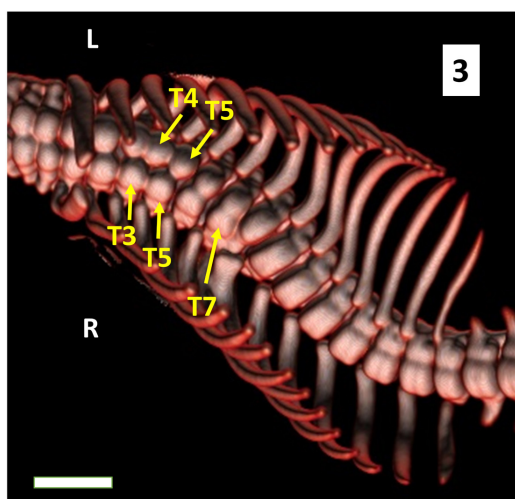


Fig. 3. CT reconstruction of the ventral vertebral bodies. The ventral vertebral bodies from the atlas to the seventh thoracic vertebra (T7) have two nodular tori. However, the right-sided nodule of T4 is not apparent, unlike those of T3, 5, and 7. L: left, R: right. Bar=1 cm.

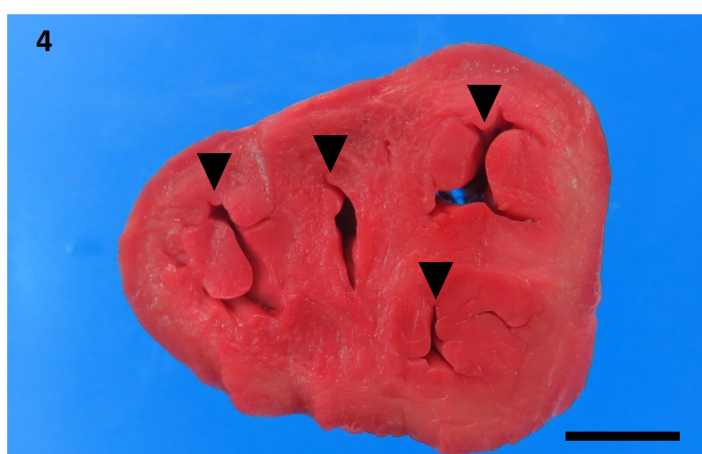


Fig. 4. Cross-section through the heart. Four ventricular cavities (arrowheads) are apparent. Bar=0.5 cm.

heart. The heart had an irregular, rounded shape and contained three atrial auricles. In addition, one aorta, two pulmonary arteries, and two ductus arteriososi were observed; however, one of the ductus arteriososis was not patent. When the heart was cut open, four ventricles (Fig. 4), partial defects of the ventricular walls, and three atrial cavities were identified.

Most of the internal organs were unremarkable at the microscopic level, except for the spinal cord. From the first cervical to the fourth thoracic spinal cord, two ventral median fissure-like structures were observed (Fig. 5), but there were no lesions suggesting duplication or malformation of the central canal, or of the dorsal or ventral horns. In addition, two ventral spinal arteries were found at the level of the first cervical spinal cord (Fig. 5).

On the basis of the above findings, the fetus was diagnosed as a symmetric undivided diplopagus, with two heads and four eyes. Diplopagus is a rare congenital malformation that occurs in monozygotic twins [3]. In domestic animals, this malformation has been reported in the cow [2, 5], pig [3, 7], dog [6], and sheep [9]. However, to our knowledge, this is the first report of an undivided diplopagus with cardiac malformation in a wild boar. It is generally recognized that monozygotic twins result from the fission of the embryo early in development. There are two hypotheses for the development of a diplopagus: abnormal fusion and incomplete fission [10]. The fusion hypothesis states that a diplopagus results from the joining of two formerly separate monozygotic embryos [10], while the fission hypothesis states that a diplopagus results from incomplete fission of an early embryo [10].

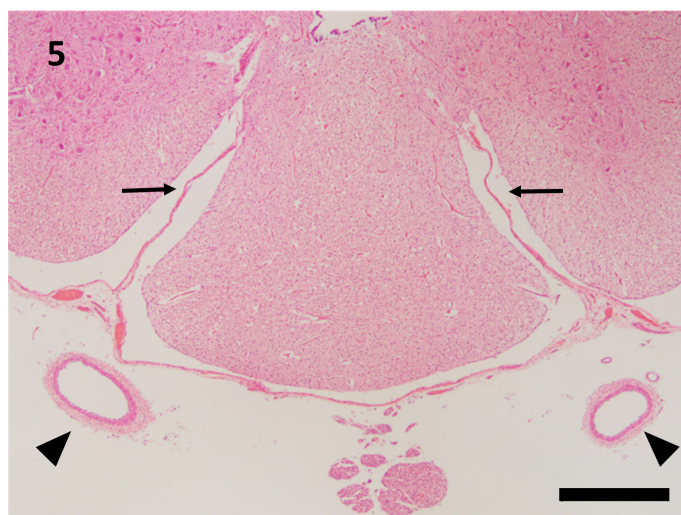


Fig. 5. Histopathology of the first cervical spinal cord. Duplicated ventral median fissure-like clefts (arrows) and ventral arteries (arrowheads). Hematoxylin and eosin staining. Bar=500 μ m.

In domestic animals, viral infection, toxic plants, and other toxins are considered to be causes of generalized congenital abnormalities [1]. In particular, classical swine fever virus (CSFV) infection has been reported to cause congenital malformations, including cerebellar hypoplasia and dysmyelination, in piglets [8]. Wild boars, which are closely related to pigs, are also reported to be susceptible to infection with this virus [4]. Therefore, congenital malformations may also occur as a result of CSFV infection of wild boar fetuses. In the present case, however, hypoplasia of the viscera and encephalitis, which are typical sequelae of viral infection, were not observed. In addition, CSFV RNA was not detected using reverse transcription-polymerase chain reaction (RT-PCR) in formalin-fixed and paraffin-embedded tissues (liver, spleen, kidney and brain; data not shown). In the pig, teratogenesis following the ingestion of plant-derived toxins, such as conium, mimosine and nicotiana, or chemical agents, including metrifonate and methallibure, has been reported [1]. However, the development of diplopagus following viral infection or poisoning has not been reported. We could not ascertain whether in this instance the dam had been exposed to such agents, because it was captured in the wild. It is probable that components of the head and heart were duplicated at an early developmental stage in the present case, however, the detailed mechanisms remain to be clarified.

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