Abdominal ectopia cordis in an aborted calf without chromosomal aberrations

Abdominale ectopia cordis bij een geaborteerd kalf zonder chromosomale afwijkingen

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ABSTRACT

Ectopia cordis is a rare congenital heart disease characterized by partial or complete displacement of the heart out of the thoracic cavity. Apart from cattle, the condition has also been described in humans and is frequently associated with Cantrell’s pentalogy. It is classified into five types: cervical, cervicothoracic, thoracic, abdominal and thoracoabdominal. The prognosis is poor and ectopia cordis may be linked to the presence of unbalanced chromosome alterations. In this report, a case of abdominal ectopia cordis is described in an aborted calf, in which no unbalanced structural chromosomal variants could be identified.

SAMENVATTING

Ectopia cordis is een zeldzame congenitale hartaandoening die gekarakteriseerd wordt door een gedeeltelijke of volledige verplaatsing van het hart buiten de thoracale ruimte. Naast het rund, is deze aandoening ook beschreven bij de mens en ze wordt regelmatig geassocieerd met ‘de pentalogie van Cantrell’. Er zijn vijf types van ectopia cordis beschreven: cervicaal, cervicothoracaal, thoracaal, abdominaal en thoraco-abdominaal. De prognose is slecht en daarnaast kan ectopia cordis geassocieerd worden met de aanwezigheid van ongebalanceerde chromosomale defecten. In deze casuïstiek wordt een geval van abdominale ectopia cordis beschreven bij een geaborteerd kalf in afwezigheid van ongebalanceerde structurele chromosomale varianten.

INTRODUCTION

Ectopia cordis is an uncommon congenital heart disease (Gabriel et al., 2014) described in several animal species such as cattle, pigs (Gruys et al., 1978), rabbits (De Sesso, 1979) and hamsters (Willhite, 1983). In these last two species, it is regularly associated with the exposure to teratogenic compounds during gestation (De Sesso, 1979; Willhite, 1983). In cattle and humans, no environmental causes have been described for this condition. In the literature, it has often been suggested that in humans, ectopia cordis may be the result of unbalanced chromosomal defects, such as mosaic trisomy 16, trisomy 18 and sex chromosome abnormalities (King, 1980; Shaw et al., 2006; Arnaoutoglou et al., 2010; Diaz-Serani and Sepulveda, 2020), although this is not always the case (King, 1980). In calves, only a limited number of ectopia cordis cases have been genetically investigated and were shown to be associated with the presence of extra chromosome material represented under the form of a marker chromosome (Windberger et al., 1992).

Ectopia cordis is postulated to start early in the embryonic period. There are three theories regarding the pathogenesis of this congenital defect: 1) primary failure of descent and midline fusion of the lateral body folds; 2) failure of midline fusion due to early rupture of chorion and/or yolk sac; 3) amniotic band syndrome (Shirian et al., 2010). In 1958, Cantrell et al. described a pentalogy of malformations in humans, often associated with ectopia cordis. Cantrell’s pental-
ogy consists of (1) midline supraumbilical abdominal wall defect; (2) defect of the caudal part of the sternum; (3) deficiency of the thoracic diaphragm; (4) defect in diaphragmatic pericardium; (5) congenital intracardiac defects. Although the pentalogy of Cantrell has been used interchangeably with ectopia cordis, in their original description, the authors were careful to distinguish between these two anomalies (Cantrell et al., 1958). Ectopia cordis occurs when the heart is displaced outside the thoracic cavity. The displacement of the heart can be cervical, cervicothoracic, thoracic, thoracoabdominal or abdominal. In humans, the most common types are thoracoabdominal and abdominal (Alphonso et al., 2003). However, in cattle, the cervical type is the most common (82 %) and the abdominal the least common (3%) (Hiraga and Abe, 1986).

CASE HISTORY AND FINDINGS

An aborted female Holstein-Friesian calf (30 kg) measuring approximately 80 cm in length (crown-rump length) was presented for necropsy in the abortion program follow-up organized in collaboration with the Federal Agency for the Safety of the Food Chain (FASFC). It originated from a 400 heads meat producing herd. During necropsy, several anomalies were detected, such as hydrocephalus internus, torticollis, arthrogryposis of the left foreleg, underdeveloped lungs which were partially ventilated and an abdominally located heart. The heart was embedded in the liver (normal hepatic structure), which adapted to the cardiac shape. The heart was cranially delineated by an intact diaphragm (Figures 1 and 2). The muscular portion of the septation between the thoracic and abdominal cavity was rather thin and underdeveloped at the periphery with formation of both crura. There was a central opening in the diaphragm for the esophagus (esophageal hiatus) and the truncus pulmonalis to ensure blood flow to the lungs. Therefore, a ‘would-be’ pericardium was excluded. There were no evident macroscopic abnormalities at the level of the greater vessels, namely the aorta, the cranial vena cava, the truncus pulmonalis and the vena pulmonalis. Additionally, there was a rather big blood clot located at the level of the umbilical arteries. Based on the position of the heart and the absence of a diaphragmatic hernia, it was diagnosed as an abdominal ectopia cordis as a result of migratory failure and not due to abnormal ventral midline fusion.

Genetic analysis

DNA from the spleen was isolated using the DNeasy blood and tissue kit (Qiagen, Hilden, Germany) according to the manufacturer’s instructions. The concentration of the DNA was quantified with the Qubit dsDNA BR Assay kit (ThermoFisher Scientific, Aalst, Belgium). DNA, 312 ng in 50 μL 0.1x TE buffer, was first sheared to 200 bp using the Covaris Adaptive Focused Acoustics shearing procedure (Covaris, Woburn, Massachusetts) on the M220 Focused-ultrasonicator (Covaris, peak incident power = 50W, duty factor = 20%, cycles per burst = 200, temperature = 20°C, sample volume = 130 μL).

Library construction of tissue DNA was performed using the NEXTFlex Rapid DNA-seq-kit (Bioo Scientific, Austin, Texas) according to the manufacturer’s instructions. Next, libraries were equimolarly pooled to a pool concentration of 4-6 nM. Shallow depth WGS was performed using a Hiseq 3000 device (Illumina Inc., Essex, UK), single-end 50 cycles as previously described by Van Roy et al. (2017). Raw reads were mapped by Bowtie 21 (v2.3.2) onto cattle reference genome GRCv38, using the fastlocal flag. Bio-bambam’s bamsormadup2 (v2.0.87) was used to mark duplicate reads and to sort the resulting bam files. No additional quality filtering was applied. The files were indexed by SAMtools3 (v1.4.1). The novel Wisecon-
dorX (v1.1.2) was deployed to reliably deduce normalized genome-wide log2 ratios across 100 kb bins, representing the copy number (Raman et al., 2019).

Shallow whole genome sequencing of the calf spleen revealed a normal copy number profile, without gains or losses of chromosome material (not shown).

**DISCUSSION**

Ectopia cordis is a rare and striking congenital defect, which was first observed 5000 years ago (Taussing, 1982). The present case is not a ventral midline defect, but rather a problem in body cavity formation and subdivision. According to the position of the misplaced heart, ectopia cordis can be classified into five types. The present case seems to be belong to the abdominal type. The majority of human ectopia cordis cases have been associated with intracardiac defects. Interventricular septal defect, interatrial septal defect, tetralogy of Fallot, and diverticulum of the ventricle are the most commonly encountered heart lesions in humans (Leca et al., 1989; Amato et al., 1995). In cattle, there is frequently simultaneous occurrence of defects and serious valve disorders, resulting in a reduced life span of the affected animals (Hiraga and Abe, 1986). In the present case, no prominent cardiac defects could be noticed. The severity and complexity of the intracardiac defect(s) contribute largely to the poor prognosis associated with this malformation (Amato et al., 1995). Ectopia cordis has also been associated with other congenital anomalies, such as abdominal wall defects, cranial and facial malformations, cleft lip and palate, anencephaly, hydrocephaly, neural tube defects, pulmonary hypoplasia, genitourinary malformation, gastrointestinal defect and chromosomal abnormalities (Diaz, 1992; Hochberg et al., 1995; Hornberger et al., 1996).

With the advances in all aspects of human medicine, the number of infants who undergo successful surgical repair for ectopia cordis and survive, is steadily increasing (Kabbani et al., 2002). In cattle, however, surgery is rarely an option, so the prognosis stays poor and the anomaly is mainly seen after birth/abortion. In a survey of Hiraga and Abe (1986), in 118 calves with ectopia cordis, 82% accounted for the abdominal type. The majority of human ectopia cordis, 82% accounted for the abdominal type, 14% for the thoracic and only 3% for the cervical type. Forty of the forty-two calves that produced life span of the affected animals (Hiraga and Abe, 1986). In a more recent study, a long-time survival of a calf that grew normally and even became pregnant, has been reported (Onda et al., 2011).

In the present case, the rare type in cattle, namely abdominal type of ectopia cordis, is described as a result of migratory failure, associated with torticollis, crooked front limb and limited hydrocephalus internus, which resulted in abortion at the end stage of gestation. No cardiac nor unbalanced structural chromosomal variants were identified.

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**REFERENCES**


