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CASE REPORT

First Report of Acephaly in a Lamb from the Cajamarca Region, Peru

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ABSTRACT

This case report aimed to describe the first documented case of acephaly in an aborted lamb fetus from the Cajamarca region. The case involved a fetus lamb of the Creole breed from an extensive farming system, which underwent a postmortem macroscopic examination complemented by ultrasonographic and radiographic evaluations. The evaluation confirmed a severe congenital cranial malformation, characterized by the absence of definable craniofacial skeletal structures. Radiographic analysis was decisive in confirming this finding, revealing only a small residual bony structure at the cranial extremity of the cervical column, articulated with a normally developed vertebral column. An additional external malformation was observed, consisting of the fusion of the auricles and the absence of auditory canals in the cervical region. It is concluded that this report constitutes a precedent for local veterinary medicine, contributing to the documentation of the congenital cranial malformations in lambs.

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INTRODUCTION

Congenital anomalies are defined as developmental abnormalities present at birth that alter the structure, function, or molecular organization of the organism (Sadler, 2019). In mammals, the central nervous system (CNS) is highly susceptible to the development of malformations, most of which are incompatible with extrauterine life (Balseiro *et al.*, 2022). Within this category, neural tube defects represent a group of serious anomalies resulting from abnormal or incomplete closure of the neural tube during the critical stages of embryonic neurulation (Zarzycki *et al.*, 2021). The most frequently reported neural tube defects (NTDs) include spina bifida, encephalocele and anencephaly, among others (Isaković *et al.*, 2022; Long, 2006). Within the spectrum of NTDs, it is necessary to distinguish between acephaly and anencephaly, terms that are sometimes used interchangeably. Anencephaly is characterized by the absence of cerebral hemispheres and cranial arch, but with craniofacial structures usually present (Salari *et al.,* 2022). On the other hand, acephaly refers to the congenital absence of the head, including the craniofacial structures (Shirazi *et al.*, 2009).

In sheep, these types of anomalies have rarely been reported. According to data from 4417 lambs necropsied over a three-year period, only 1.5% of the cases examined presented some type of CNS anomaly (Dennis, 1975). Of this group, acephaly is an extremely rare developmental anomaly characterized by the congenital absence of the head. Although several types of congenital CNS malformations have been previously documented in various domestic species (Moussaoui *et al.*, 2021), reports describing acephaly in sheep are very limited, with the study by Shirazi *et al.* (2009) being the most prominent report of acephaly, which resulted from an in vitro embryo transfer procedure. However, similar cranial malformations have been described in other regions, including a case of anencephaly and other associated malformations in a bovine fetus in India (Lakkawar *et al.*, 2002), and a more recent study described a case of anencephaly with concomitant congenital malformations in a lamb in Turkey (Dörtbudak *et al.*, 2024).

The etiology of congenital malformations is usually multifactorial. Genetic components include heredity, recurrence risks, inbreeding, and racial predispositions; environmental factors include exposure to chemical

agents, trauma, radiation, and even episodes of hyperthermia during pregnancy. Likewise, critical nutritional deficiencies, especially those related to folic acid metabolism, have been associated with developmental abnormalities of the central nervous system, while genetic and environmental interactions may also contribute to their development (Zarzycki *et al.,* 2021; Isaković *et al.*, 2022).

The objective of this study was to describe a case of acephaly in an aborted lamb fetus from the Cajamarca region of Peru.

Case description:

Case history: An aborted female lamb fetus of the Creole breed, weighing 3.2kg, was donated by the owner to the Veterinary Pathology Laboratory of the Universidad Nacional de Cajamarca for diagnostic examination. The animal came from an extensive breeding system located in the province of San Marcos. Genealogical information was not available. The predominant macroscopic anomaly was the absence of the head (acephaly).

External and macroscopic examination: A detailed clinical examination of the lamb was performed following standard veterinary pathological examination procedures (Maxie, 2015). This evaluation focused on the identification and description of visible congenital abnormalities. A zoometric analysis was also performed using a graduated measuring tape to record the linear dimensions of the specimen. A complete postmortem examination and internal organ sampling were not performed, as the diagnostic evaluation was complemented with imaging techniques (radiography and ultrasonography). This constitutes a limitation since a detailed necropsy and histopathological examination were not performed, which could have been useful in confirming the precise anatomical extent of the malformation.

X-ray: The imaging study was performed using a clinical-grade X-ray generator (Tingle X-Ray Products, TXR 325, USA) equipped with a collimator (Dongmun Co., Ltd, MD-LB50, South Korea), following standard veterinary radiographic diagnostic procedures (Thrall, 2018). For comprehensive anatomical visualization, the lamb was positioned in laterolateral (LL) and ventrodorsal (VD) projections. Radiographs were obtained to evaluate the cervical region and the axial and appendicular skeleton.

Radiographic exposure settings were adjusted according to the anatomical region of interest. The recorded tube voltage and tube current were as follows: cervical region 60–65kV/6–8mA; thoracic cavity: 65–75kV/5–7mA; limbs: 50–60kV/6–8mA; and entire skeleton: 65–75kV/6–10mA.

Ultrasound: The abdominal and thoracic cavities were examined using a portable veterinary ultrasonography system (Medsinglong Global Group, model MSLFP06, China) according to established standard procedures (Nyland and Mattoon, 2015). The scan was performed by placing the transducer in longitudinal and transverse planes for comprehensive visualization of hepatic and renal parenchyma, intestinal loops, and intrathoracic structures. High-frequency transducers (7.5–12MHz) were used, with medium-to-high gain and optimized focus, and a depth of field of 4–8cm.

Complete blood count: Blood samples were collected via post-mortem cardiac puncture using a vacuum extraction system (Vacutainer®) and tubes with anticoagulant (K_3 EDTA) and without additives for serum collection. Biochemical analyses were performed using an EC9004 VET veterinary biochemical analyzer (Excbio, Guangzhou Excbio Technology Co., Ltd., Guangdong Province, P.R. China). Hematocrit was determined using a KHT-410E hematocrit centrifuge (Kubota, Taiwan). Leukocyte, erythrocyte, and platelet counts, as well as differential leukocyte counts, were performed manually by light microscopy (ZEISS, Primo Star, Germany). The analysis focused on determining basic cellular and biochemical components, following standardized protocols for sheep (Brooks *et al.*, 2022).

It should be noted that the post-mortem interval between death and blood sampling was not accurately recorded, which is a limitation, since post-mortem autolysis and hemolysis may have influenced the measured values.

Diagnosis:

Macroscopic findings: The lamb's estimated gestational age was approximately 4 months, which was estimated using craniocaudal length and body measurements, data that were compared with reference fetal growth curves for the species (Sivachelvan *et al.,* 1996). External examination revealed the most severe anomaly: absence of the head with no externally definable cranial structures. No cranial structures were identified. The malformation profoundly affected the facial architecture, with agenesis of nasal and ocular structures (arrhinia and anophthalmia). However, an atypical anatomical arrangement was identified in the cranial termination of the cervical region: the presence of both ear pinnae positioned contiguously, associated with a total absence of external auditory canals (Fig. 1A).

The results of the zoometric measurements confirmed that, despite the acephaly, the development of the axial and appendicular skeleton was proportional to the estimated gestational age. The lamb had a body length of 27cm, a height at the withers of 28.5cm, and a thoracic circumference of 33cm, with a dorso-sternal diameter of 15cm. Croup length and croup height were 8cm and 27.5cm, respectively. The cannon bone circumference, in both fore and hind limbs, was 8.4cm. A cervical length of 14cm and a chest width of 10.5cm were determined (Fig. 1B).

Radiographic results: Imaging of the specimen confirmed the integrity of the axial and appendicular skeleton, in contrast to the acephaly. A vertebral column with normal anatomical development was identified, along with the absence of definable craniofacial skeletal structures, consistent with acephaly. No skull or mandibular bones were identified. However, a small, irregular residual bony structure was observed at the cranial end of the cervical spine. The atlas (C1) and the remaining cervical vertebrae showed normal anatomical morphology (Fig. 2A). The ventrodorsal projection corroborated the symmetrical and normal formation of the

forelimbs and hindlimbs. Furthermore, both the rib structure and the alignment of the vertebral column were within the standard anatomical parameters for the species (Dyce *et al.*, 2010) (Fig. 2B).

Ultrasound result: Ultrasound examination of the abdominal and thoracic cavities revealed no primary morphological abnormalities or congenital malformations in the organs examined; however, postmortem ultrasonography cannot exclude microscopic lesions or systemic pathology. The images allowed for the identification of the main visceral structures, whose echogenic characteristics were consistent with postmortem degenerative changes. On sagittal view, the left kidney measured 3.29cm in longitudinal axis, with a cortical thickness of 0.58cm. Abdominal ascites was detected in the peritoneal cavity. The changes observed in the tissue architecture were considered compatible with autolysis and other postmortem phenomena, which may have limited the ultrasonographic assessment (Fig. 3).

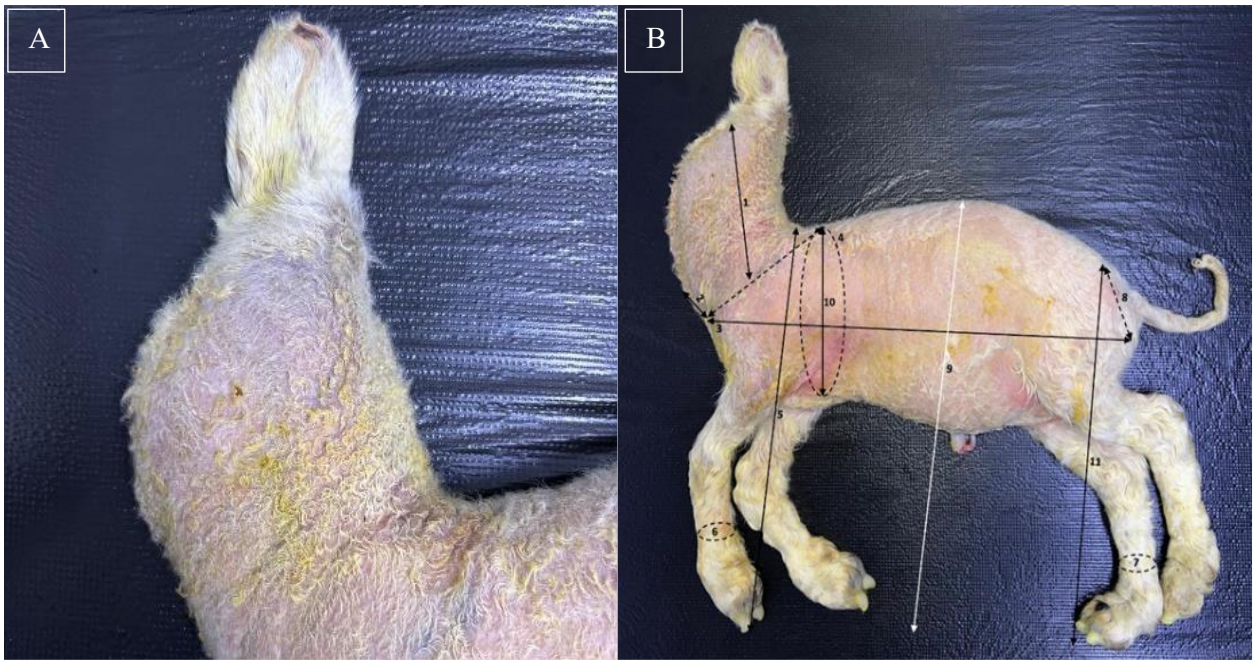


Fig. 1: Macroscopic and zoometric evaluation of the lamb with acephaly. **(**A) Macroscopic findings of the lamb with acephaly**:** close-up of the cranial termination of the cervical region showing absence of the head; noteworthy is the atypical and contiguous implantation of both ear pinnae in the upper cervical region, associated with the absence of external auditory canals. (B) Zoometric measurements in lamb. 1: Neck length, 2: Chest width, 3: Body length, 4: Thoracic circumference, 5: Height at the withers, 6: Cannon bone circumference of the forelimb, 7: Cannon bone circumference of the hindlimb, 8: Croup length, 9: Rump width, 10: Dorso-sternal diameter, 11: Croup height.

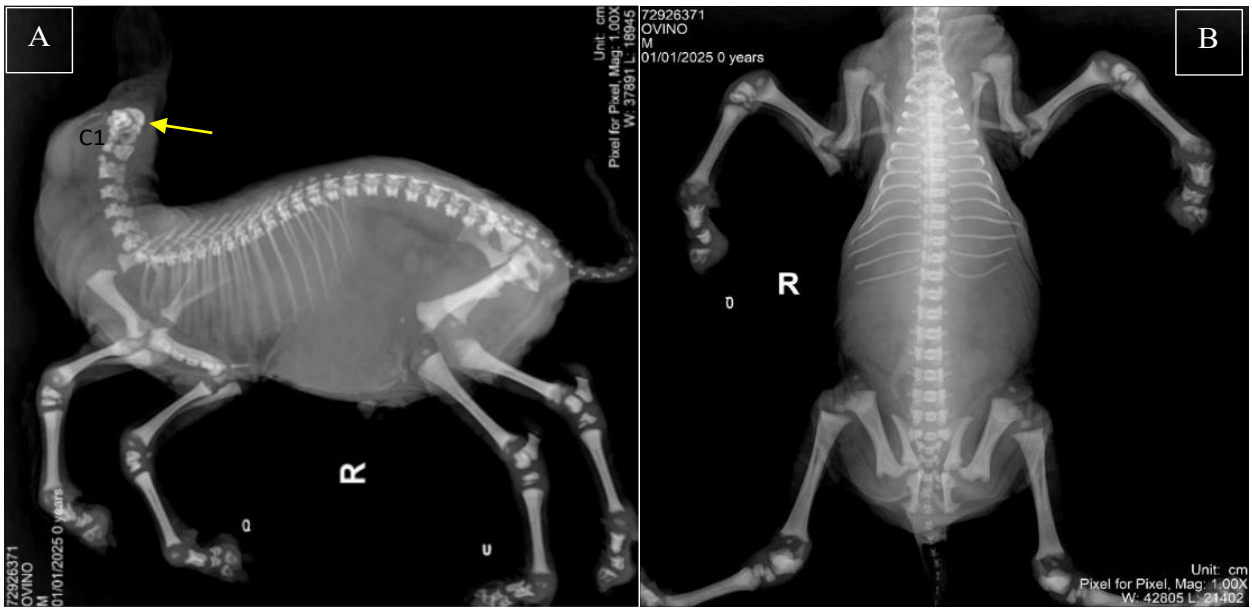


Fig. 2: Radiographic evaluation of a lamb diagnosed with acephaly. (A) Right lateral projection: absence of definable craniofacial skeletal structures (skull, mandible), with the cervical column curving dorsally and terminating in a small, irregular residual bone structure (arrow). The atlas (C1) is identified immediately caudal to the cranial termination of the cervical column. The cervical segment and the rest of the vertebral column do not show structural abnormalities. (B) Ventrodorsal projection: The normal anatomical development of the vertebral column is confirmed along its entire length, as well as the symmetry and complete formation of the forelimbs and hindlimbs, ribs, and pelvis.

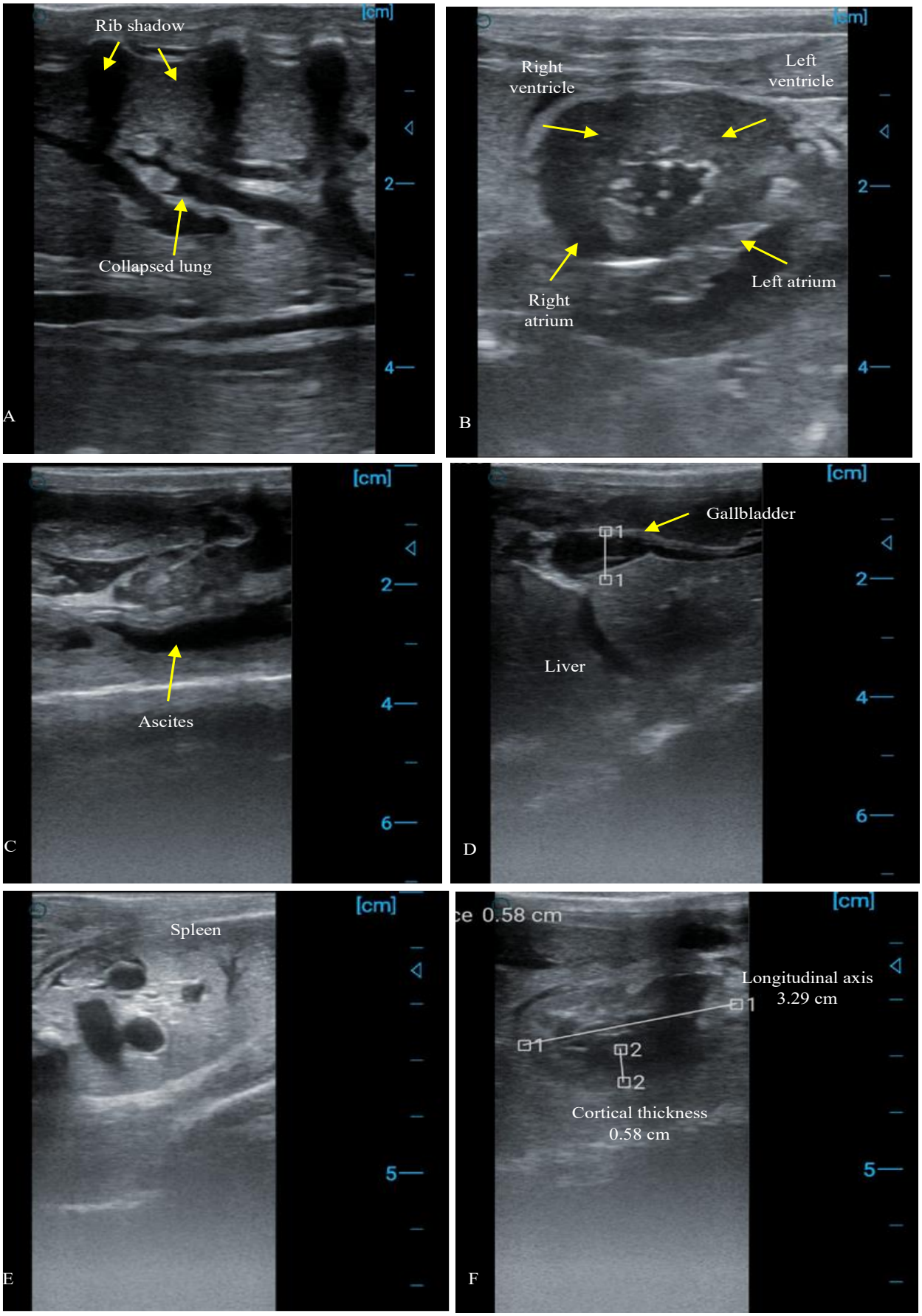


Fig. 3: Post-mortem ultrasonographic evaluation of thoracic and abdominal organs in the malformed aborted fetus lamb. (A) Thoracic ultrasound: rib acoustic shadows and collapsed lung parenchyma. (B) Cardiac transverse plane: grossly preserved ventricular morphology. (C) Evaluation of fluids and digestive tract: Marked abdominal ascites. (D) Longitudinal view of the liver with homogeneous parenchyma and gallbladder. (E) Splenic parenchyma with apparently preserved echotexture. (F) Sagittal view of the left kidney: ultrasonographic measurements of 3.29cm for the longitudinal axis and a cortical thickness of 0.58cm were recorded.

Hematological and biochemical results of the lamb: Severe leukocytosis (51.2 x 10^9/L) was identified, markedly exceeding the reference range (5.1–15.9x10^9/L), attributable to marked lymphocytosis (46.1x10^9/L). Monocyte count (1.44x10^9/L) and eosinophil count (0.27x10^9/L) were mildly increased above their respective reference ranges (0.1–0.8x10^9/L and 0–0.2x10^9/L), consistent with monocytosis and mild eosinophilia, while the remaining leukocyte parameters (neutrophils and basophils) remained within normal limits. Similarly, the total erythrocyte count (8.5x10^{12}/L) was slightly below the reference range, whereas hemoglobin concentration (13.3g/dL), remained within both the adult and neonatal reference ranges. Hematocrit (44.6%) exceeded the adult reference range (31.5–41.1%) but fell within the neonatal reference range (27–45%). However, the erythrocyte indices MCV (52.5fL) and MCH (15.6pg) were above the reference values, whereas MCHC (29.8g/dL) was below the reference value. In addition, the red blood cell distribution width (RDW-CV) showed an increase (22.4%). The total platelet count was below the reference range (213x10^9/L; reference 426–1142x10^9/L). However, the mean platelet volume (MPV) was decreased (6.2fL) (Table 1).

Table 1: Hematological and biochemical parameters of the lamb with acephaly from the Cajamarca region, Peru.

Parameter	Absolute value	Unit	Reference range(**)	Reference range(***)
Complete blood count (CBC), leukocyte differential and platelets				
Leukocytes	51.2	x 10^9/L	5.1 – 15.9	
Lymphocytes	46.1	x 10^9/L	2.1 – 10.2	
Neutrophils	3.3	x 10^9/L	0.8 – 6.3	
Monocytes	1.44	x 10^9/L	0.1 – 0.8	
Eosinophils	0.27	x 10^9/L	0 – 0.2	
Basophils	0.09	x 10^9/L	0 – 0.2	
Erythrocytes	8.5	x 10^{12}/L	9.2 – 13	
Hemoglobin	13.3	g/dL	10.5 –13.7	9.0 –15.0
Hematocrit	44.6	%	31.5 – 41.1	27 – 45
MCV(*)	52.5	fL	28 – 35	
MCH(*)	15.6	pg	10 – 13	
MCHC(*)	29.8	g/dL	32.2 – 39.2	
RDW-CV(*)	22.4	%	10.6 – 14.3	
Platelets	213	x 10^9/L	426 - 1142	
MPV(*)	6.2	fL	8.4 – 14.1	
Serum biochemistry profile				
ALT	75	U/L	26 – 34	-
AST	260	U/L	60 – 280	37 – 353.5
GGT	300	U/L	20 – 52	31- 154
ALP	480	U/L	68 - 387	58 – 727.7
Glucose	45	mg/dL	50 - 80	33 – 98.1
Urea	60	mg/dL	17.18 – 42.88	12.8 - 100
Creatinine	2.2	mg/dL	1.2 – 1.9	0.40 – 1.80
Total proteins	5.4	g/dL	6.0 – 7.9	3.10 – 11.4
Albumin	2.1	g/dL	2.4 – 3.0	-
Globulins	3.3	g/dL	3.5 – 5.7	-

(*) MCV: Mean corpuscular volume; MCH: Mean corpuscular hemoglobin; MCHC: Mean corpuscular hemoglobin concentration; RDW-CV: Red cell distribution width (coefficient of variation); MPV: Mean platelet volume. (**) Hematological reference ranges for adult sheep were obtained from Stayt (2022), while serum biochemical reference ranges were obtained from Kaneko *et al.* (2008) and Ferreira de Souza *et al.* (2020). (***) Hematological reference ranges for neonatal lambs obtained from published references when available, Lester *et al.* (2009). Serum biochemical reference ranges for lambs from birth to 1 year of age were obtained from Varanis *et al.* (2021).

Biochemical analysis of the lamb with acephaly revealed mild alterations in biomarkers of hepatic function. Elevated activity of alanine aminotransferase (ALT: 75U/L) was detected, whereas aspartate aminotransferase (AST: 260U/L) remained within the reference ranges reported for both adult and young lambs. These findings, together with the presence of hypoalbuminemia (2.1g/dL), may be compatible with impaired hepatic synthetic capacity and structural changes in the hepatic parenchyma, which could be related to the ascites observed on ultrasound examination. Urea concentration (60mg/dL) exceeded the adult reference range but remained within the reference range reported for lambs from birth to one year of age (Varanis *et al.*, 2021). Similarly, glucose concentration (45mg/dL) was below the adult reference range but within the reference range reported for young lambs (33–98.1mg/dL).

DISCUSSION

We report a case of an ovine fetus with acephaly, in which a small residual bony structure was identified cranial to the atlas, a finding rarely reported in sheep. Previous documentation of this anomaly in sheep is very scarce, with most of it limited to isolated cases (Shirazi *et al.*, 2009). The CNS is considered to have one of the highest rates of malformations compared to other organ systems because the development of nervous tissue is one of the earliest and most complex events in organogenesis. Therefore, any alteration or complication during these initial stages can irreversibly modify the architecture of the developing brain (Di Muro *et al.*, 2020).

Radiographic evaluation of the lamb revealed structural integrity of the vertebral column, contrasting with absence of cranial structures consistent with acephaly. The concordance between the findings of this study and international case reports is notable. Shirazi *et al.* (2009) documented a case where the only anomaly was a compact bony structure at the base of the skull that maintained its connection with the first cervical vertebra. This anatomical feature is important because the presence of this residual bony structure suggests that the axial skeleton developed normally despite the absence of the cranial structures. Although its precise anatomical identity cannot be confirmed without histological examination its presence indicates that a rudimentary, incompletely ossified cranial remnant may persist despite total aplasia of the cranial vault (neurocranium) (Maxie, 2015). Furthermore, the clinical observation of this case, characterized by the fusion of the auricles in the neck region and the absence of external auditory canals, closely resembles the clinical examination described by Shirazi *et al.* (2009). These morphological similarities suggest that a common pathogenic mechanism might be involved in both cases. However, further histopathological investigation is needed to confirm this hypothesis.

On the other hand, the use of ultrasound proved crucial for the non-invasive inspection of the coelomic cavity. The most relevant clinical finding was the detection of abdominal ascites. This finding suggests an alteration in fetal fluid dynamics, a common finding in cases of severe congenital anomalies. Furthermore, the sonographically homogeneous parenchyma of the liver, kidneys, and spleen (as assessed ultrasonographically) is consistent with a localized nature of the neural tube defect, although microscopic or systemic pathology cannot be excluded (Maxie, 2015).

Regarding the specimen's physical constitution, the body weight recorded in the present case (3.2kg) and the data collected through zoometric measurements (e.g., body length 27cm, thoracic circumference 33cm) were comparable to those reported by Sivachelvan *et al.* (1996), who described a mean fetal body weight of 2111±612.0g and a crown-rump length of 43.67±2.87cm at 18–20 weeks of gestation. This suggests that the allometric growth of extracranial structures was not compromised. Despite the severe cranial malformation and neurocranium, the somatic development of the rest of the body (trunk and limbs) remained within the standard growth parameters for the ovine species.

The hematological and biochemical profile of the acephalic lamb, with its marked lymphocytosis ($46.1x10^9$/L) accompanied by mild monocytosis ($1.44x10^9$/L) and thrombocytopenia ($213x10^9$/L), represents an atypical clinical finding that warrants consideration. In ovine fetal physiology, alterations in the lymphoid series may be associated with antigenic or inflammatory stimuli during gestation, although this finding is nonspecific and cannot establish an infectious etiology. Lymphocytosis has been described in association with infectious and inflammatory processes, including conditions affecting the developing central nervous system (Kirkland, 2015). Taken together, these findings could support the hypothesis that acephaly in lambs may be related to an infectious or inflammatory process occurring during the first triggmester of gestation, a critical period for neural tube closure (Vandevelde *et al.*, 2012). However, these findings do not establish the cause of the acephaly, as further genetic or molecular assessments would be required to evaluate these possibilities.

The absence of higher brain structures may have influenced the hypothalamic-pituitary axis, a key regulator of fetal metabolism; however, the glucose concentration recorded (45mg/dL), although below the adult reference range, fell within the reference range reported for young lambs (33–98.1mg/dL) and therefore does not support a diagnosis of hypoglycemia in this case. Other fetal or peripartum factors may also have contributed to variations in glucose homeostasis (Fowden and Forhead, 2007). Furthermore, the identified hypoalbuminemia (2.1g/dL) could have contributed to the abdominal ascites detected by ultrasound. Albumin is responsible for maintaining vascular oncotic pressure, and its decrease promotes fluid extravasation into the peritoneal space (Maxie, 2015). Additionally, the urea concentration (60mg/dL), although above the adult reference range, remained within the range reported for young lambs, and the observed biochemical alterations may be compatible with chronic fetal hypoxia and renal hypoperfusion; however, these associations remain speculative without further diagnostic confirmation.

Finally, it is important to highlight some limitations of this case study. A complete necropsy was not performed, which would have allowed for the dissection of cranial nerve endings, the spinal cord, and internal organs. In addition, it should be noted that the majority of reference intervals used to interpret hematological and biochemical results are based on adult sheep, due to a lack of specific literature, which complicates interpretation and necessitates a cautious approach. This also limited the histopathological analysis that, in previous studies such as that by Dörtbudak *et al.* (2024), has proven useful for a better characterization of associated congenital anomalies. Consequently, for this report, the diagnosis is based solely on external examination and imaging findings, including radiographs and ultrasounds, which are recognized as not being able to completely replace a detailed necropsy and histopathological analysis.

Conclusions: This report describes a case of acephaly in a Creole lamb from Cajamarca. The findings in this report through macroscopic examination, radiographs, hematology, and biochemistry are consistent with a severe congenital malformation, including leukocytosis with lymphocytosis and hypoalbuminemia. These findings could be associated with the reported congenital malformation and the peripartum condition of the fetus. This case contributes to the documentation of congenital cranial malformations in lambs from the Cajamarca region.

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Data Availability Statement: Data are available within the article.

Ethics Statement: Ethical approval was not required for this clinical case, as the animal was an aborted fetus and no experimental procedures were performed. The owner donated the carcass and authorized its use for diagnostic evaluation and scientific publication.

Author's Contributions: Conceptualization, L.Z and F.T.Q.; formal analysis, J.P.T and M.C.R.; investigation, F.T.Q and G.R.C.; resources, C.Q.P and W.A.; data curation, F.T.Q and G.R.C.; writing-original draft preparation J.P.T and L.Z.; writing-review and editing, M.C.R and F.T.Q.; visualization, C.Q.P and W.A. All authors have read and agreed to the published version of the manuscript.

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