

## ULTRASONOGRAPHIC DETECTION OF ABNORMAL ALLANTOIC FLUID ACCUMULATION SECONDARY TO EMBRYONIC DEATH IN A CROSSBRED COW

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### SUMMARY

Embryonic mortality (EM) remains a major constraint in improvement of reproductive efficiency and profitability in livestock enterprises. Usually embryonic death (ED) is followed by the resorption of embryo and gestational fluids and the cow returns to oestrus eventually. The present communication reports a rare case of abnormal allantoic fluid accumulation secondary to ED in a 13 year old crossbred dairy cow and its earliest detection by transrectal ultrasonography (TRUS).

**Keywords:** Allantoic fluid, Bovine, Embryonic mortality, Ultrasonography

Embryonic mortality (EM) has been ascribed to be the prime cause for reproductive failure especially in high producing dairy cows. It is generally defined as the loss of conceptus that occurs during the first 45 days of pregnancy. Approximately 30 per cent of pregnancies are lost in the form of EM causing an increase in the successive calving interval, thereby adversely affect the success of annual calf crop programme in dairy cows (Chaudhary and Purohit, 2012).

The prime consideration to reduce the inter-calving interval in dairy cow is early diagnosis of pregnancy so as to capture, treat and rebreed the animals on time that fail to conceive (Pieterse *et al.*, 1990). Embryonic mortality can be assessed by using a variety of techniques including measurement of milk progesterone concentration or pregnancy specific proteins, and TRUS (Bekele *et al.*, 2016). Ultrasonography (US) has an advantage over traditional method of rectal palpation in pregnancy diagnosis (PD) as the detection of foetal viability and assessment of embryonic loss during early stages of gestation is possible with US (Patel *et al.*, 2005). The introduction of US for early pregnancy diagnosis has allowed researchers to differentiate the timing and extent of late embryonic losses in cattle (Santos *et al.*, 2004). Following early embryonic death (EED), the embryonic tissues are usually resorbed and the animal returns to oestrus if there is no other conceptus in the uterus.

The total quantity of foetal fluid in cattle increases progressively throughout pregnancy. Sharp rises in the quantity of amniotic and allantoic fluid occurs between days 40 and 65 of gestation, between 3 to 4 months and again between 6.5 to 7.5 months (Roberts, 1986). The first and last of these are due to allantoic and the second is due amniotic fluid increment. The present study details about the use of US in bovine reproduction especially in early

pregnancy diagnosis, detection of embryonic loss and associated accumulation of sterile allantoic fluid in the uterus.

The study was carried out in a 13 year old crossbred dairy cow maintained at University Livestock Farm and Fodder Research Development Scheme, College of Veterinary and Animal Sciences, Kerala. The animal had calved one year back and had a history of repeat breeding. The animal was inseminated at the oestrus induced by double PG protocol (500 mcg prostaglandin intramuscularly at 11 days interval). Since the animal did not return to subsequent oestrus, TRUS was performed on day 28 post-service for confirming the pregnancy. Pregnancy was confirmed based on the presence of anechoic vesicle in the lumen of uterine horn representing the fluid filled allantoic cavity and the presence of echogenic embryo with heartbeat within (Fig. 1). The embryonic heart beats were considered as the main criteria to assess the viability of the embryo and the blood flow to heart was assessed by Doppler system. TRUS was done with a real time color Doppler ultrasound scanner (MyLab™ Gamma, EsaoteSpA, Italy) equipped with liner array, 5-10 MHz frequency transrectal transducer (SV3513, Esaote Europe B.V, Netherland).

On 60<sup>th</sup> day post AI, a routine rectal palpation revealed presence of clear disparity between the gravid and non gravid horn and TRUS revealed an abnormally small foetus/ foetal parts or embryonic remnants without heartbeat (Fig. 2.1). Cardiac and umbilical blood flow indicating foetal viability was not observed on Doppler ultrasonography, suggestive of late embryonic or early foetal loss. Functional corpus luteum (CL) verum was detected in the ovary ipsilateral to the gravid horn (Fig. 2.2). Though no caruncles were observed, amniotic and allantoic fluid was observed in the gravid horn. Diameter of the amniotic sac was 25.7 mm (Fig. 2.3). The allantoic measurements could

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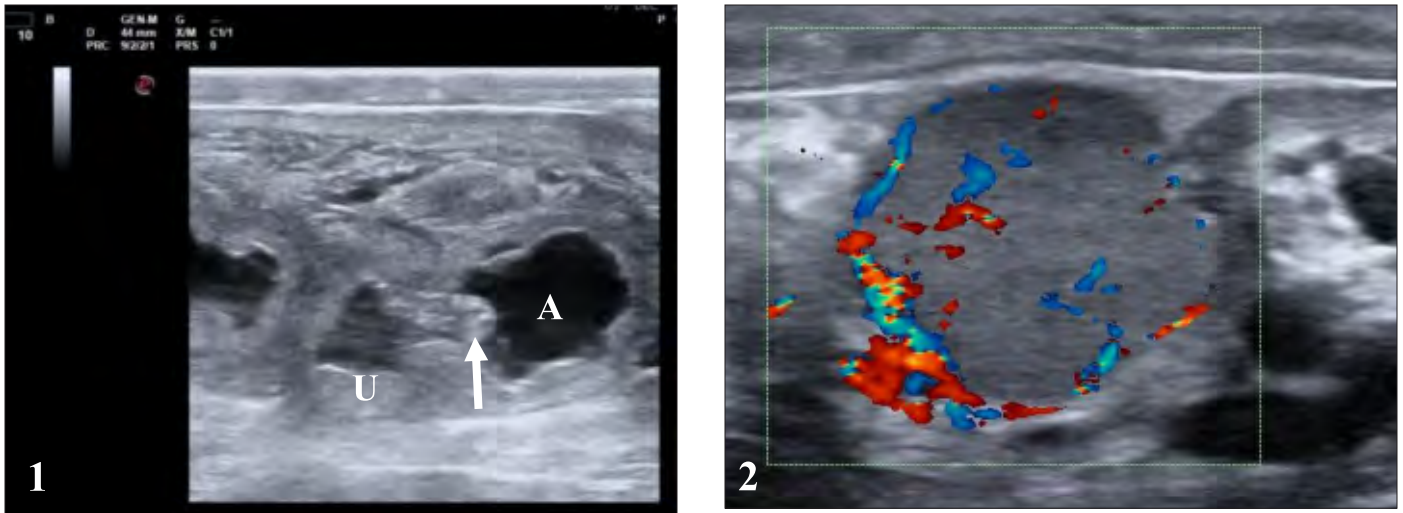


Fig. 1. Early pregnancy diagnosis on day 28 by transrectal ultrasonography: (1) A- Gestational sac; U- uterine wall; Arrow head- Embryo; (2) Corpus luteum verum with active blood flow

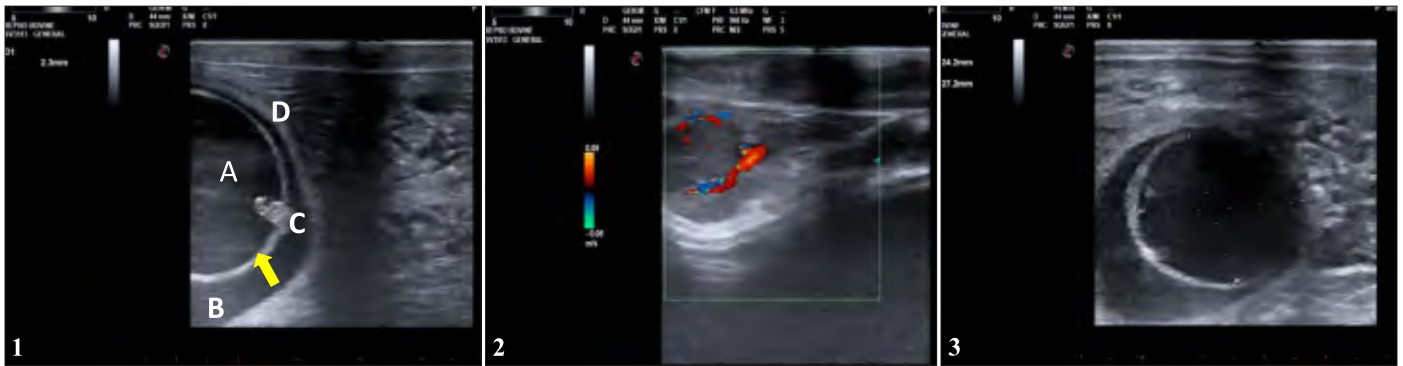


Fig. 2. TRUS on day 60: (1) A- amniotic fluid; B- allantoic fluid; C- remnant of dead embryo; D- Uterine wall; Arrow head- Thickening of amnion; (2) Corpus luteum is retained with active blood flow; (3) Diameter of the amniotic sac is 25.7 mm

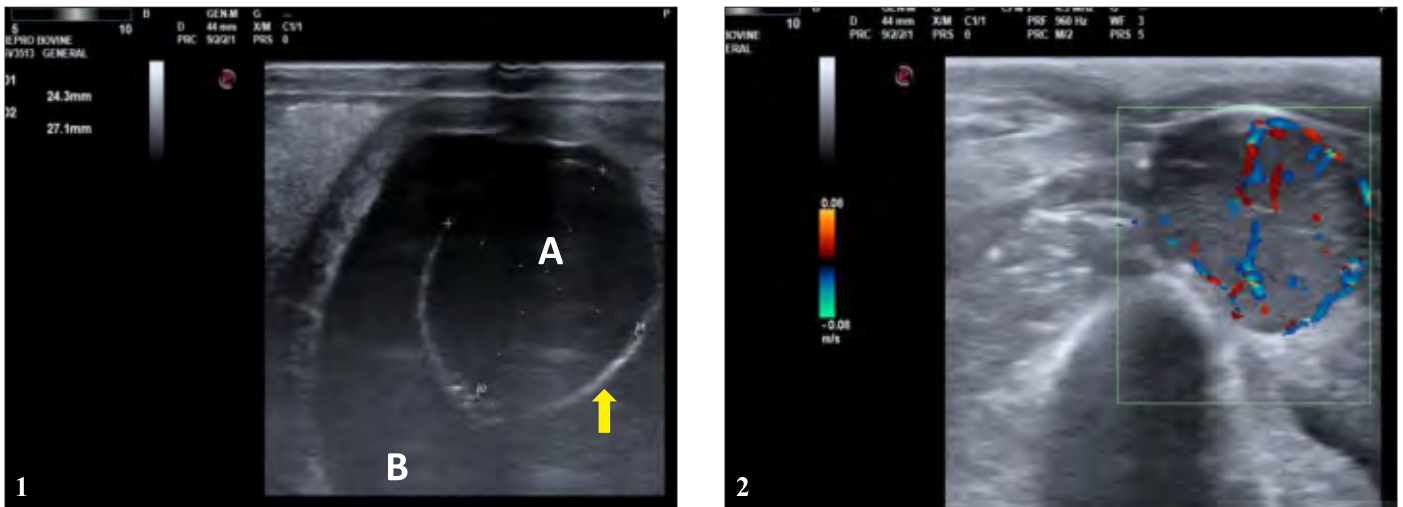


Fig. 3. TRUS on Day 75: An increase in allantoic fluid noticed on day 75 (1) A- amniotic fluid; B-allantoic fluid; arrow head-amniotic membrane; Diameter of the amniotic sac is 25.7 mm; (2) Corpus luteum is retained with active blood flow

not be taken as the whole of allantois could not be visualized in a single window with TRUS. The animal was active and alert and all the clinical parameters were in normal range.

A review after two weeks revealed almost complete

resorption of embryonic/foetal parts and there was no increase in the size of amniotic cavity, but a drastic increase in the allantoic fluid (Fig. 3.1). Doppler US revealed a functional CL (Fig. 3.2) and the serum progesterone level was determined to be 4.1 ng/ml. By 90<sup>th</sup>

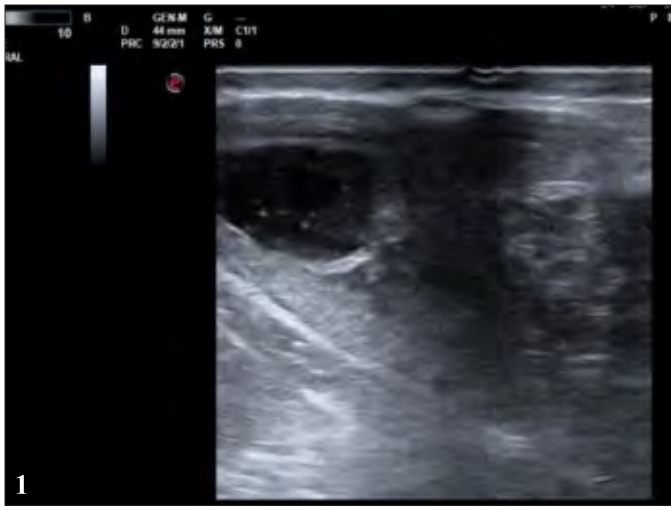


Fig. 4. TRUS after 36 h of PGF<sub>2</sub>α administration revealed evacuation of uterine contents (1) and complete elimination of uterine contents noticed after 48 h of PGF<sub>2</sub>α administration (2)

day of gestation, the fluid filled uterus became abnormally enlarged and descended into the abdominal cavity. The pregnancy was terminated with 500 mcg Cloprostenol (i/m), which was followed by evacuation of large quantity of foul smelling fluid 36 h later (Fig. 4). On culture of the uterine fluid, no organisms could be detected suggesting a probable non-infectious cause for the EM.

In the present study, the pregnancy was diagnosed early at day 28 post-AI wherein diagnosis of pregnancy by TRUS relied on the detection of anechoic allantoic fluid and the echogenic embryo proper. The viability of the embryo was confirmed with the detection of heart beat. The study conducted by Romano *et al.* (2006) had suggested that TRUS could be used for early PD in bovines with the maximum sensitivity between days 28 and 30 post-service. The animal under the present study exhibited embryonic loss between days 28 and 60 post AI, which was diagnosed by TRUS. Transrectal examination suggested the presence of uterine horn disparity suggestive of two months pregnancy but TRUS revealed the disappearance of majority of the foetal parts but adequate amniotic and allantoic fluid in the gravid horn. Late EM can be diagnosed when the embryo is detected without a heartbeat or when a previously observed embryo with a heartbeat is no longer visible during subsequent US examinations (Rani *et al.*, 2018). Silke *et al.* (2002), who used TRUS and recorded the embryonic losses in cows and heifers between days 28-42, 43-56, 57-70 and 71-84, ascribed the conceptus losses to be 3.2 - 1.52, 1.99 - 2.27, 0.66-1.51 and 1.34 - 0.76 per cent, respectively.

In this study, the animal which exhibited embryonic loss between days 28 and 60 post- service did not show any evidence of embryo expulsion nor was the fluids resorbed. Doppler US revealed retention of an active corpus luteum

with an active blood flow. Abnormal fluid accumulation stress on the failure of expulsion and failure of resorption but an increase in allantoic fluid, which was not expected. Contrarily to this finding, Kastelic *et al.* (1991) reported that ED is usually followed by either resorption or expulsion of the embryo and uterine contents through the cervix, which either goes unnoticed or appears as a vulval discharge of clear mucus.

Singh *et al.* (2020) reported that ultrasonography provided an immediate result and information about the fetal viability. This study also highlighted the importance of a subsequent examination to detect the embryonic loss in those animals diagnosed as pregnant early in gestation using US. Subsequent examination of pregnancy can identify cows that experience embryonic loss and can be subjected to early rebreeding post pregnancy loss. This is of utmost significance in the efforts to improve dairy profitability, since the reproductive efficiency is remarkably reduced among these animals by extending the calving interval (Fricke, 2002).

In conclusion, US has a great role not only in the diagnosis of ED in bovines, but is also highly beneficial for an early diagnosis of serious pathological conditions like drossy, which may have been left undiagnosed by conventional methods.

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