

## Ultrasonographic investigation of early embryonic death in mares

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### Abstract

The aim of the study was to characterize clinical symptoms for an impending embryonic death using Real-Time ultrasound technique. The present study was conducted in two different farms, including (257) foaling mares (Arabian, Thoroughbred and Cross breed) in Al-laith equine herd and Equestrian club in Al-Ameria, 4-14 years old foaling mares which are situated in areas around Baghdad and extended one calendar from July 2011 to July 2012. These mares were examined for pregnancy diagnosis by using trans-rectal ultrasonography a 5MHz linear-array transducer between days 10 and 60 after breeding (last breeding day=day 0). The incidence of early embryonic death (EED) in this study occurred most frequently  $\leq$  day 40 (6/ 13, 46.2%), while low occurrence of early embryonic loss was during the period from 41-50 day and 51-60 day (2/13,15%), (5/13,38.45) respectively as the total number of mares suffered from EED were (13) mares. The results of this study showed a significant differences ( $P<0.05$ ) between the percentage of early embryonic death and gestation period through 10-60 days. Sonographic characteristics of an impending embryonic death were as follows: the size of conceptus was underdeveloped, the endometrium become heterogenic and estrous-like. A reduced volume of conceptual fluid and disorganization of the conceptual membrane were the most important criteria after embryonic death. In conclusion, that B-mode ultrasound is a useful and essential method to diagnose and predict embryonic mortality in mares.

**Key words:** Ultrasonography, ultrasound, early embryonic death, mare.

### استخدام الموجات فوت الصوتية في التقصي عن هلاك الجنين المبكر في الأفراس

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### الخلاصة

تهدف الدراسة الحالية الى تقييم هلاك الجنين المبكر في الأفراس باستخدام تقنية الموجات فوت الصوتية عبر المستقيم. شملت الدراسة على (257) فرسا من سلالات مختلفة (العربية، والهجينة، والثوروبريد) وتراوحت اعمارهن بين (4-14) سنة. أجريت الدراسة في نادي الفروسية في العامرية ومزرعة الليث لتربيته الخيل الخاصة، الواقعتان في أطراف بغداد. تم تشخيص الحمل في الأفراس باستخدام تقنية الموجات فوت الصوتية عبر المستقيم مع مجس مستقيمي ذو تردد 5MHz خلال الفترة (10 - 60) يوم بعد التسفيد واعتبر يوم آخر تسفيد هو يوم الصفر. وبينت الدراسة أن أغلب حالات هلاك الجنين المبكر (6/ 13، 46.2%) حدثت قبل اليوم الرابع من الحمل مع فارق معنوي منخفض ( $P<0.05$ ) بين هلاك الجنين المبكر وفترة الحمل مقارنة مع الفترات الأخرى (2/13، 15%) و (5/13، 38.45%) يوم و (51-60) يوم بعد التسفيد حيث كانت (13، 15، 2%)، (5، 13، 38.45%) على التوالي. في ثلاثة عشر فرسا شخص فيها هلاك الجنين المبكر بالموجات فوت الصوتية. بين التصوير بالموجات فوت الصوتية ان اهم علامات حصول هلاك الجنين المبكر هي: القصور في تطور وحجم الحويصلة الجنينية مع قلة السوائل لها، بطانة الرحم تعطي مظهرا متجانسا وذات شكل يماثل حالة العطاف، اضافة الى عدم انتظام اغشية الحويصلة الجنينية المبكرة، نستنتج من الدراسة الحالية أن الموجات فوت الصوتية تعد طريقة مفيدة وأساسية في تشخيص وتوقع حدوث هلاك الجنين المبكر في الأفراس.  
الكلمات المفتاحية: الموجات فوت الصوتية، هلاك الجنين المبكر، الأفراس.

## Introduction

Early embryonic death in the mare is one of the main causes of infertility in mares (1). A rate of embryonic death ranging from 5 to 24% between 11 and 50 days post-ovulation (2, 3). Pseudo pregnancy occurred more frequently following embryonic loss after day 20 (100%) (4). Various causes and factors responsible for EED in mares, a part from presence of twins (5). Lactating mares and mares bred during foal heat have been reported to have a higher incidence of EED than non-lactating mares, embryo-loss rate (Days 12 to 39) was greater in old than in young mares (6, 7). The condition of mares, such as breeding condition and the uterine environment of the embryo or fetus are the major factors in relation to occurrence of early embryonic death (8). (9) Reported that the endometritis is considered an important cause of embryonic loss in mares. Pregnancy rates are reduced in species such as the equine due to post ovulatory aging of the oocytes which occurs when insemination is performed more than 12 hrs. after ovulation (10). Ultrasonic indications of impending loss at later stages included failure of fixation, an echogenic ring (vesicle) or mass floating in a collection of fluid, an echogenic area in the dead embryo, absence of heart beat, and a gradual decrease in volume of placental fluids with disorganization of the

placental membranes (4). Papa et.al (11) showed that the diameter of the embryonic vesicles after 16 days of ovulation were smaller in the mares with embryonic loss.

## Materials and methods

Two hundred and fifty seven mares are employed in this study. They were from different farms, including: AL-Laith equine herd 115 mares and equestrian club in AL-Ameria 142 mares. These mares were from different breeds (Arabian, Thoroughbred and Cross breed), and they were aged from 4-14 years which was estimated by dentition. Ultrasound examinations were done weekly and made according to (12), with a real time B-Mode scanner equipped with a 5MHz linear array rectal transducer, (SIUI-CTV-200V, China). Early embryonic death (EED): Beside appearance of estrus signs on some mares, EED was detecting by disappearance or abnormal development of the embryo, after their detection in previous examination.

## Statistical Analysis

The Statistical Analysis System- SAS (13) was used to effect on different factors in study parameters (percentage). The Qi-square ( $\chi^2$ ) test at the comparative between percentages in this study at 0.01 or 0.05 level of probability.

## Results

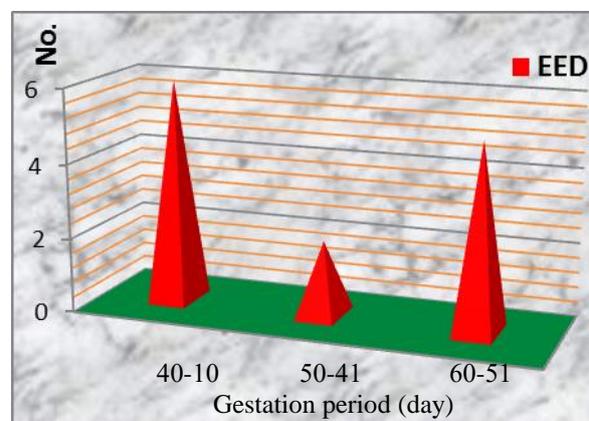
Results of the present study, showed a significant differences ( $P < 0.05$ ) between EED rate and gestation period through 10-60 days (Table 1 and Fig. 1).

**Table 1: The relation between the EED rate and gestation period. \* ( $P < 0.05$ ).**

Gestation period (days)	No. of EED (%)
10-----40	6(46.2)
41-----50	2(15.4)
51-----60	5(38.4)
Total	13(100)
Qi-square - $\chi^2$	5.371 *

Thereafter, the majority of pregnancy losses in mares in the present study occurred

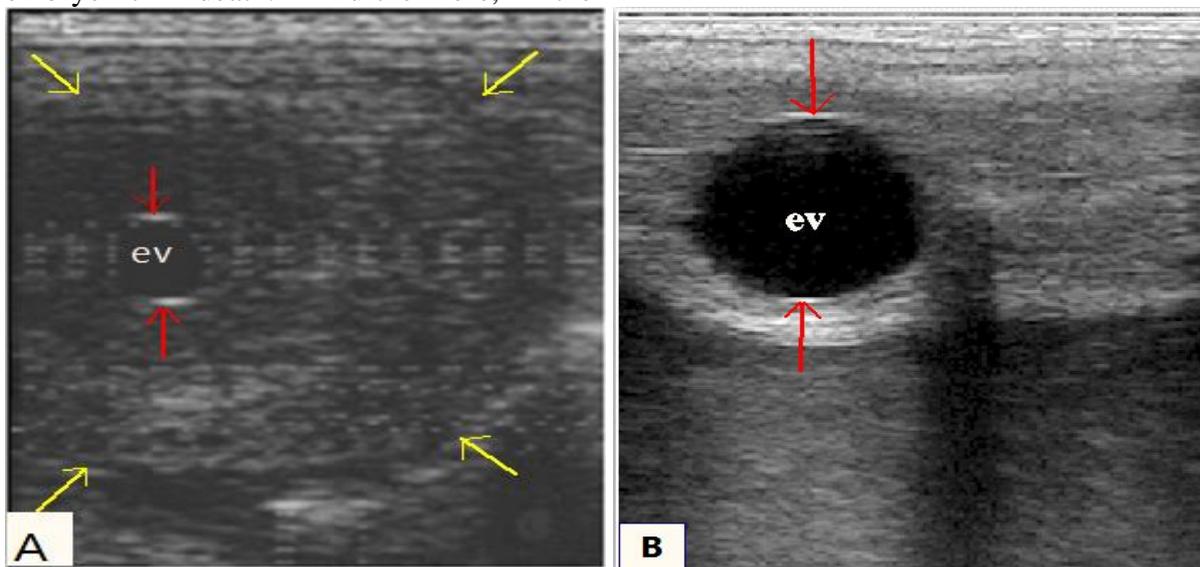
**Fig. 1: The relationship between the post-breeding interval (days) and EED through the period of 10-60 days.**



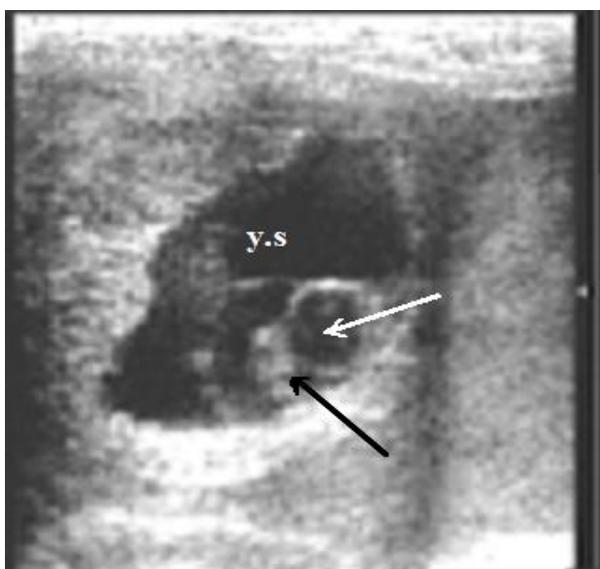
most frequently  $\leq$  day 40 (6/13, 46.2%), while low occurrence of EED was during the

period from 41-50 day and 51-60 day (2/13, 15%), (5/13, 38.45) respectively. Meanwhile, the ultrasonographic scanner in current study showed a smaller diameter of conceptus (Fig. 2), was the most important criteria of sonographic characteristics of an impending embryonic death. Furthermore, the

sonographic scanner show that the vesicle was underdeveloped with a small allantoic sac, absence of embryonic vesicle, abnormal shape and absence of corpus luteum associated with estrous behavior indicated early embryonic death (Fig. 3).



**Fig. 2: Ultrasonographic images of conceptus day 14th (A) and day 22nd (B). Undersize embryonic vesicle (ev). Periphery of cross-section of the uterine horn (yellow arrows), specular reflections (red arrows).**



**Fig. 3: Ultrasonographic image of embryonic vesicle at 30 day, the vesicle was underdeveloped with a small allantoic sac (white arrow), the embryo (black arrow), and yolk sac (y.s).**

## Discussion

The present study shows that early embryonic loss agree with the results of (14) (15), who reported that the majority of EE

loss occurred prior to fortieth day of gestation was 77.1%, 63%, respectively, because of a critical developmental processes during this period. Moreover, for the majority of this period, the conceptus is entirely dependent on progesterone produced by a primary CL for its continued survival (16) who reported a slight inflammatory reagents in the uterus initiate the production of prostaglandins which induces luteolysis and in turn ends pregnancy (17). While others found a maternal age beyond approximately 14 years is accompanied by a marked increase in the incidence of EE loss, because the maternal age factor is associated with deficiencies in the uterine environment and related with the degenerative changes in the endometrium, reducing its nutritive capacity for the developing conceptus (9,18,19). Moreover, the majority of EE loss prior to 40 days of gestation may be refer to the increased number of mares bred on foal heat. Similar results were reported by (20) who adverted identified significantly higher loss rates in mares bred at foal heat compared with those bred at subsequent heats.

Meanwhile, measurement of embryonic vesicle diameter by ultrasonography was showed coincide with observation of (11,21), whom reported that the diameter of the EV measured by ultrasonography was smaller in the mare with impending embryonic losses during days (11-20) of gestation, compared to the normal pregnancy (22). Additional important ultrasonic observation that indicate

an imminent embryonic death including floating membranes, abnormal shape of conceptus and underdeveloped phenomenon of embryonic vesicle as well as a reduced volume of conceptual fluids (10). In conclusion, the results of current study showed that a Real-time ultrasound is a valuable method to predict and diagnosis early embryonic mortality in mares.

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