



Prediction of the ovulation time in estrus mares by different methods

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Abstract

The study aimed to evaluate the prediction of ovulation time by three different methods body temperature, Dramineski estrus detection apparatus, and observation of the mature follicle daily by ultrasonography in mare's estrus. Sixteen mature cycling mares were used during the breeding season in 15 March and 15 June of 2021, aged 5-6 years old, at the Nineveh Equestrian club in Mosul city, Iraq. Mares were examined daily by rectal palpation and ultrasound with a 5.5-7 MHz linear-array transducer probe to scan the ovaries and uterus. The mare's follicle diameter and body temperature were recorded daily in the Dramineski estrus detection apparatus to predict ovulation when the data was up to 400 degrees. The result showed that the body temperature was increased significantly from the 1st day to the third day of estrus at $P < 0.05$, Dramineski estrus exhibited a elevation on the fourth day showing high significant variation at the 1st day to the third day and fourth day of estrus at $P < 0.01$. The ovulation was higher in left than right ovary was recorded 62.5%, 37.5%, respectively. There was an increase in the follicular diameter scores during the third and fourth days of estrus at $P < 0.01$. In conclusion, the combination of Dramineski estrus detection and preovulatory follicle diameter recorded daily is the best method to predict the time of ovulation, mares with a follicle diameter larger than 40 mm predicted the ovulation it has been pregnancy rate up to 75% after natural breeding service for only one time.

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Introduction

Estrous behavior in the mare ordinarily about 4-7 days, varied according to different breeds, and the ovulation time usually occurs at the last 1-2 days (0-48 hours) before the ends of estrus (1). Ovulation is defined as a complex sequence of events. It has been included that cytological, biochemical, and hormonal changes happen as consequences of pre-ovulation. It is well known that the pre-ovulatory follicle will collapse and then release the ovum from the ovulatory follicle (2). The inefficient breeding in mares is associated with low foaling rates. It has been reported that failure to detect the predicted time of ovulation is regarded as one of the leading causes of decreased fertility (3). Prediction of the ovulation time could have substantial use in synchronizing with the time of natural service or artificial insemination breeding within the predictable time of

ovulation (4). Breeding at ovulation day leads to decreases the possibility of post-breeding endometritis in mares (5). Currently, the Veterinarian uses different methods for detecting the onset of ovulation in mares, including palpation per rectum and ultrasound-based on some parameters of the ovulation process, duration of estrus, and an indication of ovulation is close. Therefore, the examination should be repeated every 4-6 hours of the interval during the estrous phase. However, the continuous examination was not economically feasible and practically, estrous behavior ordinarily within the ends 24h after ovulation event, while some ovulated in mares when estrous signs were finishing. The soft pre-ovulatory follicle develops 12 hours before ovulation in 40% of the estrus mares (6). This method is still not suitable for predicting ovulation in some mares because these methods need skill and are possible for dangerous mares (3). Most estrus mares were found the poor tone of the

uterine by palpation per rectum; however, no particular changes at the ovulation time (4). Although during the 7th day, the size of the pre-ovulatory follicle increases linearly at an average rate of 2.7 mm every day (5,6); meanwhile (7) detects the stop of this increase in the diameter of follicle size within 1-2 days before ovulation time. Pre-ovulatory follicles show a distinct variation in shape from a spherical to appear shaped or conical structure in 84% of the pre-ovulatory phase. The lasting follicles were retained to a spherical shape (5). Moreover, decrease the amount of luminal fluid of endometrial edema earlier to ovulation time, although this changes not continuously in all cases (4). The specific ultrasound images could be informed that ovulation time was imminent by using several data such as the increased thickness of the follicular wall, increased follicular fluid in echogenicity, follicular shape become irregularity and detection of the texture ovary appear is a very soft follicle by palpation per rectum. Nevertheless, the daily examination of estrus mare predicts ovulation would be acceptable the mare becomes pregnant by one service (6). The temperatures of mares where fluctuations, therefore, could be used as a technique for prediction to time of ovulation; though, researchers showed to consider this method in mares have been to predict ovulation (3).

The study aimed to evaluate the prediction of ovulation time in Mares in different ways, including measurement of body temperature, Dramineski estrus detection apparatus, and observation of the mature follicle daily by ultrasonography at estrus Mares.

Materials and methods

The data were collected from sixteen mature cycling mares during the breeding season from March 15th to June 15th, 2021. Animals were aged 5-6 years old, good body condition; all mares were kept under the same management and nutrition at the Nineveh Equestrian club in Mosul City-Iraq.

Confirmation of normal reproductive estrus mares was checked daily by using signs of estrus behavior in the presence of the fertile stallion. Estrus signs were included increased interest in the stallion, winking of the vulvar lips, frequent urination, squatting, and tail raising. Mares were examined daily by rectal palpation and ultrasound with a 5.5-7 MHz linear-array transducer probe for scanning the ovaries and uterus daily detection for estrus (8).

Three different methods were used to evaluate ovulation time in mares, including; body temperatures of the mares were measured using a conventional digital thermometer in the rectum: Dramineski estrus detection and prediction of ovulation time apparatus when the data up to 400 degrees. Mares were determined for estrus daily, and their ovaries were scanned for assessment of pre-ovulatory follicle diameter during days of estrus. When a pre-ovulatory follicle of greater than 40 mm is detected, it means the presence of

an echogenic structure at the location of the previously shown follicle that was taken to indicate ovulation. The uterine scan was detected off the echogenicity uterine appearance and the intra-uterine fluid. The normal was a small amount of an intrauterine anechoic fluid while excluding the eight mares suffering from post-breeding endometritis was showed moderate or high intrauterine echogenic fluid recorded ultrasonography (9).

Statistical analysis

Data were evaluated using the statistical SPSS program v.23 software (SPSS In. Chicago, IL., USA). All results were expressed as mean \pm standard error of the mean. One-way ANOVA and LSD test evaluated the significance between groups, and $P < 0.05$ was considered significant.

Results

Evaluation of the predicted ovulation time in mares in different ways was presented in Table 1. The results showed that the body temperature was significantly different between the days of estrous cycles, Table 1. It should be noted that the body temperature from the first day of estrus to day 4th of the estrus cycle increased sharply by about 1 degree (Table1). Interestingly, detection of Dramineski estrus showed increased significantly at different days of estrus cycles (Table 1). The results of Dramineski estrus detection showed that day fourth recorded the highest values compared to day 1st, 2nd, and 3rd by about 138, 84, and 59 degrees, respectively (Figure 1).

The ovulation data was recorded more frequently ovulated from the left ovary than the right ovary 62.5 and 37.5%, respectively. The diameter of the follicles was increased during the 3rd and fourth days of estrus cycles (Table 1). However, there was no significant difference between the 2nd and first days of estrus. The average increase of follicular diameter growth during estrus days was 0.5, 0.6, and 0.68 cm/daily in the 2nd, 3rd and 4th days of estrus, respectively (Figure 2). The results showed that the volume of the uterine fluid increased at 1st day and second day while the volume declined during the third and fourth days.

Table 1: Shows different methods used for the prediction of ovulation time in mares

Parameters	Body Temperature (°C)	Dramineski estrus detection	Follicle diameter cm
1 st day	36.86 \pm 0.19 ^a	278.75 \pm 19.8 ^a	2.46 \pm 0.27 ^a
2 nd day	37.12 \pm 0.21 ^{a,b}	322.5 \pm 18.0 ^{a,b}	2.95 \pm 0.27 ^{a,b}
3 rd day	37.55 \pm 0.17 ^b	357.5 \pm 18.68 ^b	3.55 \pm 0.26 ^b
4 th day	37.62 \pm 0.19 ^b	416.25 \pm 6.25 ^c	4.28 \pm 0.06 ^c

The letters a, b, c, and d, indicate a statistically significant difference at $P < 0.05$ within each column.

The correlation between parameters revealed a strong positive correlation noticed among the days of estrus and body temperature, Dramineski estrus detection, and follicular diameter at r value 0.390 and p value 0.003, 0.000, 0.000, respectively. Moreover, the Dramineski estrus

detection was also showed a positive correlation with the diameter of the follicle (r value 0.390 and p value 0.027). Meanwhile, the body temperature does not reveal a correlation to Dramineski estrus detection (r value 0.234 and p value 0.197) (Table 2).



Figure 1: Show the value of Dramineski estrus detection. (A) 290 on 1st day, (B) 310 and (C) at 2nd day, (D) 400, (F) 430, and (G) 450 on the at 4th day of estrus in Mares.

Table 2: The values of positives correct and negatives correct between methods prediction of ovulation time in mares

Methods	Dramineski estrus detection	Follicle diameter cm	Days
Body Temperature (°C)	0.234	0.390*	0.507**
Significant (2-tailed)	0.197	0.027	0.003
Dramineski estrus detection		0.715**	0.749**
Significant (2-tailed)		0.000	0.000
Follicle diameter cm			0.725**
Significant (2-tailed)			0.000
Days			
Significant (2-tailed)			

* Correlation is significant at the 0.05 level (2-tailed).



Figure 2. Show the size of follicle diameter (cm) at the day of estrus by ultrasonography. (A) 2.3 cm at 1st day, (B) 3.1 cm at 2nd day, (C) 4.0 cm at 3rd day and (D) 4.2 cm, (E) 5cm at 4th day of estrus in mares.

Discussion

Predicting ovulation time at breeding farms is imperative to increase pregnancy rates and assist the veterinarians in limiting the optimal time for breeding an average estrus mare. The present study provides a method that becomes even more critical with used artificial insemination or when heavily booked stallions to reduce post-breeding endometritis by only one service per cycle is possible and reduce uterine contamination with delayed service or artificial insemination (10).

This study recorded that estrus signs occurring increase frequently within larger pre-ovulatory follicles (2). The results of the current study are in agreement with results (6). However, the body temperature increased significantly at $P < 0.05$ in the 3rd day and fourth day of estrus, Roig *et al.* (6) showed that the beginning of a temperature points its ability to accurately predict ovulation in the mares associated with measurement of LH surge due to the onset of temperature increase occurs when releasing of the LH surge, lead to the

ovulation event. So, body temperature was advanced significantly when a pre-ovulatory follicle was more significant than 35 mm. Therefore, body temperature change could be related to ovulation time in mares (3), this would be the period one would expect to see the difference. Because of the established diurnal effect, this change in body temperature may be a reflection of that particular fluctuation (11).

Dramineski estrus pattern was observed significant elevation on the 4th days compared to the first, second, and third day of estrus in mares. Estrus detection increased value associated with the impending ovulation time related to changes in the chemical and electrical resistance of the vaginal mucus and the LH hormone level. A decline of vaginal electrical resistance was shown during the pre-estrus phase, followed by a gradual increase until ovulation (9,12-14). In addition, it is the best method for predictable the beginning of ovulation in mares.

Ultrasonic evaluation of the ovary during estrous shows the follicular diameter tended to increase gradually during the third and fourth day prior to ovulation day's high

significantly, without differences between the first and second days before ovulation was between 3.55cm in the third day before ovulation and over 4.2 cm in fourth days before ovulation and then growth stopped. A recent study showed that quantifiable ultrasonic evaluation of the pre-ovulatory follicle is useful for successive assessment of the developmental progress until ovulation time (15). Therefore, many studies have been conducted on the concept that LH stimulation of ovarian follicles involves activating a local epidermal growth factor in the mare (7). The increase of LH level leads to ovulation of a pre-ovulatory follicle (6,16).

Published data support the measurement of follicular diameter as a good method for predicting ovulation of the estrous mare (6,17-19). In addition, Dolezel *et al.* (20) recorded a relation of ovulation time with increasing the size of pre-ovulatory follicles. The increase of follicular diameter growth prior to ovulation at estrus days in the current study agrees with Dolezel *et al.* (20) report that the follicle diameter increases linearly at an average rate of 2.7 mm / daily at estrus days prior to ovulation. The ovulation time in mares is typically measured by the size of a pre-ovulatory follicle and is related to the estrous behavior of the animal. However, when the dominant follicle reaches up to 35 mm, ovulation can occur within a few hours, associated with variation in individual mares (17). While Gastal (15) we reported that ovulation about 10 percent occurs after the end of estrus in mares. The amount of endometrial edema in the uterine lumen is a declining day prior to ovulation; the edema development and regression are related to estrogen concentrations in estrus mares (21-24).

The appearance and disappearance of endometrial edema are related to the onset of estrus and ovulation in mares. Therefore, according to this study, there was a change in Dramineski estrus concerning increased follicular diameter by ultrasound to these time increments prior to ovulation that could be utilized to help predict ovulation.

Conclusion

It can be concluded that this study provides methods used for prediction of the ovulation time in mares during estrus cycles, such as measurement of follicular diameter by ultrasound and Dramineski estrus, which were much higher successful than measurement of body temperature at estrus mare. It should be noted that the pregnancy rate at first service was about 75% in mares, where the above methods predicted their ovulation time.

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Conflict of interest

The author has no conflict of interest.

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التنبؤ بوقت الإباضة في الأفراس العاطف بطرق مختلفة

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الخلاصة

دفت الدراسة الحالية إلى تقييم التنبؤ بوقت الإباضة بثلاث طرق مختلفة قياس درجة حرارة الجسم، استخدام جهاز كشف العطف ومتابعة اليومية للجريب النامي باستخدام الأمواج فوق الصوتية في الأفراس العاطف. في ستة عشر فرسا ناضجا بعمر ٦-٥ سنوات أفراس الدراسة في موسم التناسل للفترة من ١٥ آذار إلى ١٥ حزيران من نادي الفروسية لمدينة الموصل -العراق. تم الفحص اليومي من خلال الجس عبر المستقيم والأمواج فوق الصوتية بالمجس المستقيمي بتردد ٥،٥-٧ ميغاهيرتز لفحص المبايض والرحم وقياس قطر الجريب وتسجيل درجة حرارة جسم الأفراس يوميا وقراءة قيم جهاز كشف العطف درامانسكي للتنبؤ بوقت الإباضة عندما تصل القيم فوق ٤٠٠. أظهرت النتائج زيادة في درجة حرارة جسم الأفراس بشكل معنوي عند مستوى معنوية مقارنة باليوم الأول، الثاني والثالث من العطف. كما وجد فرق معنوي عالي عند مستوى معنوية = ٠,٠١ في قيم جهاز كشف العطف باليوم الرابع مقارنة اليوم الأول مع اليوم الثالث والرابع من العطف. وسجل زيادة تكرار نسبة الإباضة من المبيض الأيسر مقارنة بالمبيض الأيمن ٦٢,٥ و ٣٧,٥% بالتعاقب. كما تبين وجود زيادة في قطر الجريب في اليوم الثالث والرابع من العطف وبفرق معنوي عالي عند مستوى معنوية = ٠,٠٥. نستنتج أن ارتباط تسجيل قيم جهاز كشف العطف وقطر الجريب بالفحص اليومي أفضل الطرق بالتنبؤ بوقت الإباضة. وان قياس قطر الجريب قبل الإباضة أكبر من ٤٠ ملم وبنسب حمل ٧٥ بالمئة بعد تسفيد الأفراس لمرة واحدة فقط.