



A View Within: Reproductive Ultrasound

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Since the early 1980s, the use of ultrasound has revolutionized our understanding of mare reproductive physiology, and with it stud management. Ultrasound allows the practitioner to diagnose problems and monitor reproductive activity with considerable accuracy. Additionally, the non-invasive nature of the technique reduces the risks of harming the mare or her fetus, and allows sequential monitoring. Today, ultrasound is regularly used to monitor ovarian activity, and to diagnose and monitor pregnancy and reproductive abnormalities.

The Principles of Ultrasound

There are many different types of ultrasonic scanners available that are tailored to specific uses, but they all work the same way. A transducer emits high-frequency sound waves and receives the low-level returning echoes bounced back by tissues. The intensity and location of reflected echoes of high-frequency sound waves transmitted into the body are recorded, then translated into an image that is displayed on a monitor.

The two-dimensional image produced is black and white with intermediate shades of gray. The strength of the sound echoes depends on the density of the tissues it hits; in general, high-density tissue (such as bone) appears white, and low-density tissue (such as fluid) appears black.

Examination Technique

In England, the mare is usually restrained in stocks for her safety and that of personnel. Ideally, most broodmares will be accustomed to the procedure and so shouldn't require additional restraint. In the United States, many mares are stood with their back half out of the stall door (with the door frame acting as stocks), and are handled and twitched from inside the stall.

The mare's tail should be bandaged or wrapped and held out of the way. The mare's rectum must be emptied of feces so that rectal palpation can be performed to ensure familiarization of the position

and nature of the reproductive tract prior to scanning. A plastic examination sleeve is placed over the examiner's arm and another over the transducer for protection and to ease cleaning. Water-soluble lubricant is then applied liberally to the practitioner's hand and the transducer.

The transducer is carefully introduced into the rectum; undue pressure should be avoided as this might rupture the rectum wall. Once in place, the transducer is positioned above the reproductive tract in such a manner that the sound waves are emitted through the rectum wall into the reproductive tract below.

The transducer can then be angled toward various structures to be examined. It is important that the whole tract is scanned in a methodical manner, normally from one side to the other.

The transducer can be placed externally on the mare's abdomen to examine her in late pregnancy; however, this is a less common practice.

Predicting Ovulation

The dynamics of follicular and luteal changes in the mare's ovaries can be monitored very effectively using ultrasound. The stage of the mare's cycle can be ascertained and the timing of ovulation predicted. Follicles are non-echogenic (as fluid-filled structures they absorb rather than reflect sound waves, and so appear black on a scanning image). They are characteristically spherical in shape and easy to identify and differentiate from other ovarian structures (Figure 1).

The absence of such follicles is indicative of no ovarian activity, which means the mare is in anestrus (period of sexual inactivity). The presence of multiple large follicles might indicate that she is in the transitional phase just prior to the first ovulation of the season. The presence of a single large follicle might indicate a pre-ovulatory follicle just prior to ovulation (Figure 2).

The changing characteristics of this preovulatory follicle are important in predicting the time of ovulation and hence when the mare should be covered or artificially inseminated. Such monitoring can be achieved by ultrasound.

Various characteristics can be used in combination to indicate ovulation is imminent. Traditionally, follicle size was used to make this determination. Follicles of diameters greater than or equal to 1.2 inches (3 cm) were considered to be highly likely to ovulate in the next 48 hours. Also, softening of the follicle indicates more imminent ovulation, usually within 24 hours. "Softening" appears on ultrasound as the clearly spherical follicle appearing to collapse and become more flattened as ovulation approaches (Figure 2).

Size is now known to be only a guide and other aspects are also considered, among them follicle shape. As follicles approach ovulation, their clear spherical shape is lost and their margins become irregular. The follicle might appear pear-shaped as it orientates toward the ova fossa. The ova fossa is in the

center of the concave side of the bean-shaped ovary, and is the spot through which the ovum is released and passed into the fallopian tube. This softening indicates a loss of follicular fluid pressure associated with fluid loss, thus signifying the initial stages of ovulation.

Changes within the follicle wall are also used as a guide; the wall appears more defined as it thickens prior to ovulation (Figure 2). Increased echogenicity of the follicular fluid might be observed as granulosa cells are increasingly shed into the fluid, appearing as white/gray stippling of the image rather than the uniform black of less mature follicles.

Ovulation can be observed as a break or tear in the follicle wall that allows follicular fluid and the ovum (egg) to pass through the ova fossa. This loss of fluid is evident as follicular collapse, which might occur suddenly or more gradually. The space left by the evacuated follicular fluid is replaced by the corpus luteum (CL; Figure 3). For the first few days, the CL is more strictly termed a corpus hemorrhagicum (CH) and is largely made up of blood surrounded by luteal tissue. Blood, being fluid, is non-echogenic and appears as black/gray on a scanning image. The luteal tissue is echogenic and appears white.

The relative amounts of blood and luteal tissue affect the scanning picture. In many CH, where blood predominates, they might appear as a central black area or areas surrounded by white luteal tissue. As the CL ages and the luteal tissue predominates, the whole CL becomes echogenic and so appears as a white near-spherical solid structure within the ovary (Figure 3).

The CL can be monitored throughout its lifespan to evaluate its activity and identify such conditions as CL failure or prolonged maintenance, which can then be followed by appropriate treatment.

Uterine Edema

More recently the value of ultrasonic observation of uterine edema (tissue swelling) in association with imminent ovulation has been appreciated. Ultrasonic images of the uterus prior to ovulation show fluid collected between the endometrial folds, resulting in an image somewhat like a cartwheel--the white "spokes" are the cores of the endometrial folds and the dark spaces between are fluid. Uterine edema occurs in the absence of progesterone and in the presence of estrogen. It is a good indicator of estrus activity and hence imminent ovulation.

Pregnancy Detection

The early equine conceptus enters the uterus as a blastocyst at five or six days of age. Until Day 17, it is highly mobile, moving freely throughout the uterine body and horns for the first 10 days.

By Day 15 mobility decreases, and it ceases altogether by Day 17, when the conceptus becomes fixed at the junction of one of the uterine horns and the uterine body. Throughout this time the equine conceptus is spherical in shape, making ultrasonic identification relatively easy.

Ultrasonic detection of the equine conceptus is first possible at Day 10, although the accuracy of detection increases with age. Scanning from Days 10-12 isn't advised due to the increased possibility of early embryonic death (EED) at this time. Most people scan at 14 to 15 days to detect twins. By this time the period of highest risk of EED has passed; after fixation the pregnancy has a high chance of being maintained. Very few veterinarians wait until 18-20 days as one can't reduce twins after 17 days if both embryos are in one horn.

The high incidence of multiple pregnancies in some breeds, such as the Thoroughbred, does justify early scanning at Days 14-15 and again at Days 18-20. The initial scan can give the first indication of multiple pregnancies and allows early treatment, normally by manually eliminating the smaller, less viable conceptus. The second scan ensures that any other multiple pregnancies are picked up and allows more accurate determination of pregnancies that have a good chance of proceeding to term.

Ultrasound examination of early conceptuses also allows their development to be monitored and embryos can be accurately aged. (The mean vesicle size of early conceptuses is given in "Embryo Size and Age" on page 102.) Conceptuses that are small for their age are unlikely to survive. This fact is used in the manual elimination of twin conceptuses, where the best success rates are obtained if the smaller embryo is eliminated.

Past Day 17, the conceptus can be easily located at the junction of one uterine horn and the body, but at this stage it begins to lose its spherical shape. During later pregnancy, embryonic and fetal development can be monitored in detail using ultrasound. This is particularly useful if abnormalities are suspected or if there is a risk of embryonic/fetal death. There is little that can be done to prevent early embryonic death, but ultrasound examination allows it to be identified, and the mare monitored and managed appropriately, increasing the chance of returning her to the stallion. Historically, such mares would not have been identified and would have lost the opportunity to foal with the added risk of uterine infection.

Ultrasound can also help ascertain whether natural reduction, or elimination of a twin, has occurred. If this is not evident by Day 30, treatment should be considered to terminate the pregnancy. More recent work has involved the use of ultrasound to guide administration of a lethal injection of potassium chloride to one of the twin conceptuses in later pregnancy (Days 60-160).

Ultrasound can be used to measure the fetal heartbeat, an indication of viability. It can be measured as early as Day 24 of pregnancy. Fetal sexing can be conducted between Days 64-70. Additionally, ultrasound is used transrectally to determine if there is ascending placentitis. Appropriate action or precautions can then be taken.

Identifying Abnormalities

Ultrasound is very successfully used to detect ovarian abnormalities such as:

Multiple ovulations--Multiple large, preovulatory follicles warn of multiple ovulation and the likelihood of multiple pregnancies--which is good for an embryo transfer donor and increases the chances that a mare will get pregnant. However, one of the twins should be crushed if two eggs are fertilized.

Anovulatory hemorrhagic follicles-- These result from preovulatory follicles that fail to ovulate, but continue to grow to unusually large sizes (2.8-4.0 inches, or 7-10 cm, in diameter). These invariably fill with blood, then gradually recede. They can be distinguished from corpus hemorrhagica by their size and different echogenic properties, since they are composed of mainly blood rather than echogenic luteal tissue. Obviously the failure to release the ovum means that there is no ovum available for fertilization, so continuing to breed the mare during that heat is useless.

Other ovarian abnormalities--These can include neoplasms (cysts and abnormal tissue growths), and also can be detected by ultrasound. Identifying problem mares allows them to be treated prior to covering or even removed from the breeding program, saving time, money, and effort. The main uterine abnormalities identified via ultrasound are:

Intrauterine fluid--Uterine fluid is graded according to volume and echogenicity. In the U.S., fluid is graded from 1-4 with 4 being the worst (strong echogenicity, white on ultrasound, caused by thick, creamy fluid indicative of severe infection and/or uterine debris) and 1 having minimal changes (from black), indicative of clear intrauterine fluid. In other countries, the grading might be the opposite with 4 denoting clear fluid and 1 denoting serious problems. Determination and classification of intrauterine fluid is particularly important in postpartum (after foaling) breeding. The industry demands that mares are put back in foal as soon as possible; however, fertilization rates are poor and early embryonic death rates high when mating on the foal heat. Excessive intrauterine fluid (and other factors) is thought to be responsible. Ultrasonography allows such mares to be identified and treated prior to covering, or suggests delaying mating until she has recovered.

Intrauterine cysts--These might be endometrial or lymphatic in origin. They appear as fluid-filled structures attached to the inner wall of the uterus. They present a potential problem with regard to pregnancy diagnosis using ultrasound due to their similarity to early embryonic vesicles. However, their failure to grow, static nature (they don't move), and spherical shape even at large sizes allow differentiation, especially with sequential scanning. Ultrasound is the only method of identifying such cysts in the live animal.

A few small cysts present minimal problems to reproduction; however, in greater numbers and size they can impede embryonic mobility and, therefore, maternal recognition of pregnancy and so cause early embryonic death. Alternatively, they can reduce the area of uterus available for placental attachment and thus restrict placental size, causing abortion later in pregnancy. Their presence might also be indicative of general uterine disease or senility (physical infirmity of old age). (For more information, see article #3937 online.)

Highly echogenic areas--Ultrasound allows the detection of air within the uterus as highly echogenic areas. The presence of air is indicative of incompetent vulval, vaginal, and cervical seals that allow air (and with it bacteria) to enter the upper reproductive tract. Endometritis is often a consequence.

Other abnormalities such as endometritis, debris, hematomas (bruising), adhesions, and abscesses also might be observed.

Ultrasound has revolutionized equine breeding management over the last 20 years by providing accurate, non-invasive, visual information on the mare's reproductive tract. Its use in the future is guaranteed and is likely to increase with the advent of further techniques such as ultrasound-guided transvaginal follicular aspiration (recovering embryos for transfer), allantoic fluid aspiration (for testing for genetic abnormalities), biopsy, and more.

FURTHER READING

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READING AN ULTRASOUND	
STRUCTURE	COLOR/APPEARANCE
Bone	White
Fibrous tissue	Moderate white
Fat tissue	Generally white/gray with white in areas of focal fat deposition

Soft tissue	Stippled (speckled) gray
Complex fluid	Black/gray stippled with white
Homogeneous fluid	Black

EMBRYO SIZE AND AGE	
The average size of the embryonic vesicle is fairly consistent during early pregnancy in the mare, and can be used to determine the age of the embryo or evaluate its health if age is known.	
DAYS POST-CONCEPTION	SIZE OF EMBRYONIC VESICLE (MM)
15	15-20
20	30-40
30	40-50
40	65
50	80
60	100-130
SOURCE--DR. DAVIES-MOREL	


FOLLICLES ON ULTRASOUND

Figure 1: A scanning image of a large preovulatory follicle. Note the clear spherical shape indicating that ovulation is not that imminent.

Figure 2: A large pre-ovulatory follicle just prior to ovulation. Note the loss of a clear spherical shape, and the thickening of the follicle wall.

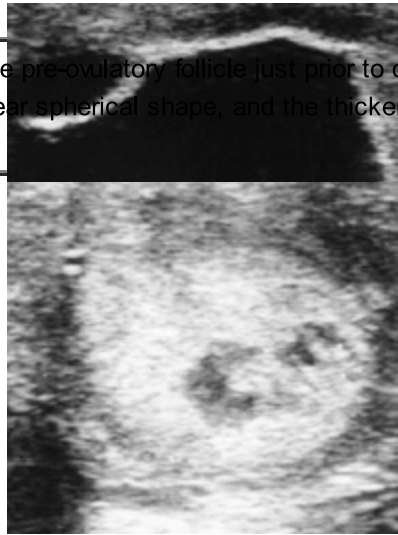


Figure 3: A corpus luteum three days after ovulation. Note the uniform echogenic nature of the structure.

IMAGES COURTESY DR. JOHN NEWCOMBE

Seek the advice of a qualified veterinarian before proceeding with any diagnosis, treatment, or therapy.

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