Introduction
In both beef and dairy cattle, pregnancy diagnosis is an important tool to measure the success of a reproductive management, to allow for early detection of problems and to achieve resynchronization of nonpregnant cows. Here are some examples of how pregnancy diagnosis can affect the performance of beef and dairy cattle operations:

- **Beef Herds**
  - Beef herds commonly use a breeding season (i.e., cows are bred during only 2 to 4 months of the year). After the end of breeding season, in the absence of pregnancy diagnosis, nonpregnant cows that will not produce a calf will remain in the herd and increase costs to the producer. If nonpregnant cows are detected after the end of the breeding season, they may be culled from the herd to reduce these costs.

- **Dairy Herd**
  - Consider the following hypothetical example: Pregnancy diagnosis is performed routinely on a dairy farm. It is observed that monthly pregnancy rates went from 40% in the previous month to 5% in the present month. The significant decrease in pregnancy rates prompts the manager to review the dairy’s artificial insemination program. It is observed that the frozen semen is not being stored properly. When semen is replaced and storage conditions changed, pregnancy rates return to normal.

- **Estrus Synchronization**
  - Consider a second hypothetical example: A dairy herd is using estrus synchronization for the first service after calving. Pregnancy rates are acceptable to this synchronized estrus. However, estrus detection is not being done properly after the first service. The manager decides to conduct monthly pregnancy diagnosis and to re-synchronize estrus of all cows diagnosed nonpregnant. As a consequence, estrus detection after the first service improves and the cumulative rate at which cows become pregnant also increases.

Therefore, it is easy to appreciate why the decision to perform pregnancy diagnosis may significantly increase the profitability of cattle operations. In the laboratory, different pregnancy diagnosis methods will be presented and their advantages and disadvantages discussed.

Estrus Detection
Cows are commonly said to cows show estrus approximately every 21 days (20 days for a heifer). The actual average length in lactating dairy cows is about 23 days. After insemination, cows can either conceive or fail to conceive to that service. If a cow is pregnant after insemination, the corpus luteum will not regress, progesterone concentrations will
Pregnancy Diagnosis

remain high, and the cow will not return to estrus. Alternatively, if a cow is not pregnant after insemination, the corpus luteum will regress, plasma progesterone concentrations will decrease, and the cow will return to estrus approximately 20-23 days after insemination. Therefore, if a cow is observed in estrus after insemination, it can be concluded that she is nonpregnant.

The use of estrus detections methods in addition to visual observation (such as tail paint or chalk, pedometers, pressure sensors, etc, as discussed previously) can potentially increase the efficacy of estrus detection and the identification of nonpregnant cows.

The use of estrus detection following insemination is useful to detect nonpregnant cows and allow for re-synchronization of such a group of cows. Relying on estrus detection to diagnose pregnant cows is very unreliable, however. Listed below are some of the reasons this is so:

- If estrus detection is not properly performed, many nonpregnant cows will not be observed in estrus and will erroneously be considered pregnant. About 50% of dairy cows are not detected in estrus when they are actually in estrus. This number increases during heat stress.
- Cows with reproductive problems such as ovarian cysts, uterine infections or anestrus also fail to return to estrus and may be misdiagnosed as pregnant cows.
- Pregnant cows can be mistakenly detected in estrus and either re-inseminated (which would probably cause an abortion) or culled as a nonpregnant cow.

Use of Milk or Plasma Progesterone

As pointed out above, the corpus luteum regresses in cows that do not become pregnant and the cow returns to estrus within approximately 20-23 days after insemination. Therefore, it is possible to diagnose cows for pregnancy based on milk or plasma progesterone concentrations at about 21 days after insemination.

The main advantage of using milk or plasma progesterone concentrations to diagnose pregnancy is that this method allows detection of nonpregnant cows soon after insemination. In particular, cows can be diagnosed not pregnant as early as 21 days after insemination.

There are several disadvantages of using milk or plasma progesterone to diagnose pregnancy. Milk and plasma progesterone samples need to be collected at least two times after insemination. Usually, samples are collected at 21 and 24 days after insemination. If either one of those samples is considered to have low progesterone concentrations, cows are diagnosed as not pregnant. Thus, the use of progesterone tests is a good diagnosis tool to detect nonpregnant cows. If both samples have high progesterone concentrations, however, there is still a certain probability that the cow is not actually pregnant. The reliability of progesterone tests to detect pregnant cows is estimated to be only around 80%. That means that 20% of cows diagnosed pregnant by progesterone are, in reality, not pregnant. Here are some reasons why:

- Cows do not show estrus exactly every 21 days. A nonpregnant cow can have showed estrus less than 21 days after insemination. This cow may ovulate and form a corpus luteum, which will increase progesterone concentrations. Thus, this nonpregnant cow may be erroneously diagnosed as pregnant.
- Cows with reproductive problems such as ovarian cysts or uterine infections also may have two consecutive high progesterone concentrations and be diagnosed as pregnant.
- Cows may be pregnant at 21 days after insemination but lose that pregnancy in the next 30 to 40 days. Pregnancy losses in that period may reach up to 30% according to recent estimates.
Sometimes cows are inseminated when not really in estrus. At 21 days after insemination, these cows will often be in the luteal phase and have high progesterone concentrations, even if not pregnant.

**Ultrasonography**

In the 1980s, real time ultrasonography was developed for use in domestic animals. An ultrasound machine resembles a radar device. A probe is inserted through the rectum and positioned above the uterus. This probe generates pulses of ultrasound that are transmitted to adjacent tissues. These pulses are then reflected back to the probe from different tissue surfaces. The reflected pulses to the probe produce an electrical signal that is processed by a scan converter and displayed on a video monitor. On the video, the intensity of the ultrasound pulses returned to the probe is converted to different shades of gray, as compared to black and white. Structures that contain fluid (such as the fluid-filled placenta) absorb most of the ultrasound pulses and the result is a black image on the video screen. On the other hand, more dense structures (such as an embryo) are more ecogenic (i.e., have greater reflectivity) and result in a light gray or white image on the screen.

The main advantages of the use of ultrasound for pregnancy diagnosis are 1) the high reliability of the results that are generated and 2) the fact that pregnancy diagnosis may be conducted relatively early after insemination (i.e., as early as 25 days after insemination).

The main disadvantages of the use of ultrasonography are related to cost and time involved with the use of this technique. Ultrasound machines are expensive and it takes more time to perform a pregnancy diagnosis with an ultrasound machine than by rectal palpation.

**Rectal Palpation**

Palpation of the uterine contents rectally is probably the most commonly used method for pregnancy diagnosis. Pregnancy diagnosis after insemination can be conducted as early as 30 days in heifers and 35 days in cows, although much practice is necessary to be able to determine pregnancy at that stage.

Several palpable structures are indicative of pregnancy. Due to accumulation of fluids within the pregnant uterine horn, one of the initial signs of pregnancy is a difference in size of uterine horns (i.e., uterine asymmetry). Also, it is possible to feel the slipping of the chorioallantoic membrane (fetal membrane) along the greater curvature within the uterus (i.e., membrane slip).

As pregnancy progresses, it becomes possible to feel the presence of the fetus within the pregnant horn. After about day 150, the fetus is too far forward in the body cavity to palpate the entire fetus although fetal structures can be palpated.

There is a rule of thumb that is quite useful in estimating fetal age based on the size of the fetus in relationship to the size of some well known animals. This rule of thumb is detailed in the following table:

<p>| Fetal size at various stages of pregnancy in relation to the size of some commonly-known animals |
|-----------------------------------------------|----------------------------------|</p>
<table>
<thead>
<tr>
<th>Stage of pregnancy</th>
<th>Animal</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 months</td>
<td>mouse</td>
</tr>
<tr>
<td>3 months</td>
<td>rat</td>
</tr>
<tr>
<td>4 months</td>
<td>small cat</td>
</tr>
<tr>
<td>5 months</td>
<td>large cat</td>
</tr>
<tr>
<td>6 months</td>
<td>beagle dog</td>
</tr>
</tbody>
</table>

Beginning at about day 90, it becomes possible to feel the placentomes. Placentomes are the structures formed by the union of maternal caruncles and fetal cotyledons by which the placenta is attached to the uterus.
Beginning at day 120 of pregnancy, one can palpate the vibrations and pulsing in the uterine artery. This phenomenon is called fremitis and is caused by the large increase in blood flow through the uterine artery. Diameter of the uterine artery increases from about 3-4 mm in a non-pregnant cow to about 1-1.5 cm (i.e., 1000-1500 mm) in a late-pregnant cow.

A table with the uterine position and diameter, as well as structures felt at palpation according to stage of pregnancy is presented below:

<table>
<thead>
<tr>
<th>Stage of pregnancy (days of gestation)</th>
<th>Uterine position</th>
<th>Uterine diameter</th>
<th>Palpable Structures</th>
</tr>
</thead>
<tbody>
<tr>
<td>35-40</td>
<td>Pelvic floor</td>
<td>Slightly enlarged</td>
<td>Uterine asymmetry/fetal slip</td>
</tr>
<tr>
<td>45-50</td>
<td>Pelvic floor</td>
<td>5.0 - 6.5 cm</td>
<td>Uterine asymmetry/fetal slip</td>
</tr>
<tr>
<td>60</td>
<td>Pelvis/abdomen</td>
<td>6.5 - 7.0 cm</td>
<td>Membrane slip</td>
</tr>
<tr>
<td>90</td>
<td>Abdomen</td>
<td>8.0 - 10.0 cm</td>
<td>Small placentomes/fetus (10-15 cm long)</td>
</tr>
<tr>
<td>120</td>
<td>Abdomen</td>
<td>12 cm</td>
<td>Placentomes/fetus (25-30 cm long)/fremitis</td>
</tr>
<tr>
<td>150</td>
<td>Abdomen</td>
<td>18 cm</td>
<td>Placentomes/fetus (35-40 cm long)/fremitis</td>
</tr>
</tbody>
</table>

The following list describes when specific structures can first be palpated:

- Membrane Slip - 30-35 days
- Amniotic vesicle - 35-60 days
- Fetus - 65 days
- Placentomes - 90 days
- Fremitis - 120 days

Rectal palpation has the advantage of being an accurate, fast, relatively cheap method that is less labor intensive as compared to the previous methods. Nonetheless, training is necessary and the exam should be conducted by a veterinarian or by an experienced herdsman.

The main disadvantage of rectal palpation is that it cannot be performed until later in gestation than some other methods. Some veterinarians are able to determine pregnancy by palpation as early as 35 days after insemination, but usually rectal examinations take place between 45 and 60 days after insemination to increase the accuracy of the exam.

Website maintained by Peter J. Hansen  Modified 10-20-2003

Photos (from left to right) are from the PJ Hansen's collection (heifers in estrus), Paolete Soto and PJ Hansen (bovine embryo stained for TUNEL), Roslin Institute (Dolly and her first lamb, Bonnie and a Meishan x Large White sow and piglets), and John Parrish (stallion with lip curl).

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