Ultrasound Imaging of the Reproductive Tract of the Bitch

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Introduction

There have been considerable advances in the use of B-mode ultrasonography for imaging the reproductive tract in a number of species, including dogs. In the bitch, the early use of ultrasound was confined to the diagnosis of pregnancy. However, more recently the full value of ultrasound imaging in dogs has been documented, and the technology is now finding wide applicability in monitoring fetal development, in timing gestation and predicting parturition, in diagnosis and management of reproductive tract disease, and in supplementing breeding soundness examinations. The aim of this article is to review the uses of ultrasonography for the examination of the reproductive tract in the normal bitch and to illustrate how this appearance changes in disease states. The literature cited includes earlier research reports and reviews by the authors [1-16], ultrasound studies and observations of others [17-30], and selected reports and reviews documenting details of canine reproductive pathology and the chronology of canine pregnancy [31-33].

The sonogram images reproduced in this review were obtained using various ultrasound machines and 5, 7 or 8 MHz scanning heads as previously described [11-12,14-16]. In many instances the abdominal hair was clipped and oil or gel applied to the skin before scanning. Others, and as currently done, involved wiping and flattening the abdominal hair with alcohol, and then applying ultrasound gel before scanning. The ability to ultrasonographically detect or discriminate a developmental feature at one day versus another during gestation is highly dependent on the equipment used. This must be kept in mind when using ultrasound units and heads that provide less resolution or a different depth of focus than used by the authors and others cited in this review. Permission to reuse images should be obtained from both Dr. Yeager and from the publisher. Events are timed in days in relation to the day of the pre-ovulatory LH surge (Day 0) unless indicated otherwise. Times in days after the day of ovulation (Day 2) are also indicated in many cases.

This review covers the following topics:

1. **The Normal Non-Pregnant Uterus**
   - Imaging the Uterus

2. **Abnormalities of the Uterus** (including cystic endometrial hyperplasia; pyometra; stump pyometra and stump granuloma)

3. **Pregnancy** (including diagnosis of pregnancy; implantation and early gestation; mid and late gestation; accuracy of pregnancy diagnosis with ultrasound; estimating fetal numbers; determination of gestational age; predicting time of parturition)

4. **Gestational Abnormalities** (including embryonic resorption; underdevelopment of fetuses; fetal abortion; fetal abnormalities; fetal distress and fetal death)

5. **The Postpartum Uterus** (including normal postpartum changes; postpartum abnormalities)

6. **The Ovary** (including detection of ovulation; abnormalities of the ovary; ovarian cysts; ovarian neoplasia; absence of the ovaries)

7. **Conclusion**
is easily imaged with the bitch in the standing position, after clipping the ventral abdomen. Alternatively, and sometimes preferably, many bitches can be examined in dorsal or lateral recumbence with minimal restraint and without tranquilization. The abdominal hair can either be clipped (research dogs) or matted down with alcohol (clinical cases) before applying ultrasound gel (Fig. 1). The uterus is positioned dorsal to the bladder and therefore in conventional imaging appears below the bladder on the ultrasound machine screen and sonogram images. The uterus may indent the normal margins of the bladder. In most cases the uterine body is found in the midline position, but this is not always the case; the uterus may be positioned lateral to the bladder, or may lie obliquely over the bladder. Often the cervix and uterine body are found in the midline just dorsal to the caudal portion of the bladder and the neck of the bladder (Fig. 2a). During anestrus, the body of the canine uterus is frequently about 5 - 8 mm in diameter, but it may be smaller than this in prepubertal animals and nonparous young adults.

**Appearance of the Uterus**

Imaging of the uterus in the transverse plane demonstrates a circular cross-sectional image (Fig. 2b), whilst the uterus appears rectangular when imaged in the longitudinal plane (Fig. 2c).

The uterus viewed in sonograms is sometimes composed of two distinct layers; a central homogeneous relatively hypoechoic region surrounded by a peripheral hyperechoic layer [9]. These two distinct layers are likely to be 1) the inner hypoechoic region representing the central endometrium and adjacent layers of circular and longitudinal myometrium, and 2) the hyperechoic surrounding uterine serosa, which appears as an hyperechoic interface. In some instances the lumen may appear, in long-section, as a central line that is less echogenic than endometrial tissue, its visibility and size depending on the amount of fluid that may be present. By tracing the uterine body cranially it is possible to detect the bifurcation and the two uterine horns in approximately one-half of bitches. In these cases, the uterine horns have a similar appearance to the uterine body but are frequently smaller in diameter. In other cases, the uterine horns are not identified because of the presence of small intestine that interferes with the ultrasound image.

**Changes in the Uterus**

The ultrasonographic appearance of the uterus changes according to the stage of the estrus cycle. During proestrus and estrus, the uterus becomes increasingly hypoechoic but can develop central radiating hyperechoic lines that are characteristic of uterine edema or faint concentric layers which might represent hypertrophy. In the longitudinal view, the shape of the uterus occasionally changes from rectangular to coiled or serpentine. The changes in uterine echo texture are associated with an increase in uterine diameter, and a small volume of central uterine fluid may be identified as an anechoic line that designates the lumen in mated bitches [9]. During the ultrasound examination of bitches in estrus after the
collection of a vaginal cytological sample, there is frequently an echogenic line visible centrally within the uterine lumen. This is indicative of aspirated air. The diameter of the uterus continues to increase several mm during the early luteal phase, and therefore is easier to identify during estrus and the first week or two of the metestrus (diestrus) stage of the cycle, compared to other stages of the cycle. However, with the decline of plasma estrogen during estrus, the edematous appearance progressively decreases. There is no apparent additional increase in uterine diameter in pregnant compared with non-pregnant bitches in early metestrus (diestrus) until implantation around Day 22 after the LH surge. By that time there is swelling due to edema at implantation sites. The diagnosis of pregnancy is initially dependent upon the detection of anechoic central (i.e., lumenal), 2 mm vesicles of fluid (gestational sacs) at each presumptive implantation site at or around Day 17 to 20 after the LH surge. Whether these initial vesicles represent uterine fluid, expanded blastocysts, or the early developing yolk sac fluid is not known (see below).

**Ovariohysterectomized Bitches**

It is not possible to make a diagnosis of ovariohysterectomy based on the apparent absence of the uterus in an ultrasonographic examination. In some cases involving ovariohysterectomized bitches, the uterus is not entirely removed, potentially leading to a false negative diagnosis. However, an enlarged uterus or uterine segment would not be entirely consistent with complete removal of the ovaries, in the absence of a sex-steroid secreting tumor of the adrenal gland. In other cases, involving intact bitches, the uterus is present but cannot be identified because of its infantile size or due to technical imaging problems, potentially leading to a false positive diagnosis.

**2. Abnormalities of the Uterus**

Abnormalities of the uterus that can be examined and that are often diagnosed based on ultrasound examination include cystic endometrial hyperplasia, pyometra, stump pyometra, stump granulomas, and uterine neoplasm.

**Cystic Endometrial Hyperplasia**

Cystic endometrial hyperplasia is a common condition that is present in many middle aged-breeding bitches. The condition may contribute to infertility by increasing the incidence of early pregnancy failure. It can also result in mucometra and pyometra.

Endometrial hyperplasia is a normal response of the uterus to exposure to increased estrogen followed by increased progesterone during the estrus cycle. Hyperplasia may result in increased echogenicity of the endometrial part of the uterine wall. The hyperplasia, and accompanying glandular activity which is stimulated by progesterone, then slowly regresses at the end of the luteal phase. The abnormal condition of cystic endometrial hyperplasia, or CEH, presumably involves a greater than normal hyperplastic and glandular response to normal changes in estrogen and progesterone when it occurs spontaneously. The condition is also seen in bitches administered excessive amounts of progestins and in bitches administered estrogen during the luteal phase or following administration of a progestin. The natural incidence is not known but appears to be much higher than in other species. The spontaneous occurrence of CEH is typically not diagnosed unless it progresses to a more severe condition, and is usually an incidental finding. It may also involve a failure of the normally hyperplastic uterus of the cycle to undergo normal regression at the end of the luteal phase, such that a degree of hyperplasia persists, and ultimately the uterine wall becomes thickened and small cystic regions can be identified within the endometrium. Depending on the size of the cysts, CEH may or may not be detected ultrasonographically, with the fluid-filled cysts appearing as small multiple 1 to 4 mm diameter anechoic regions within the uterus (Fig. 3).

Figure 3. Abdominal sonogram of a 10 year-old Doberman Pinscher bitch with dilated cardiomyopathy and chronic liver disease, showing a segment of uterus in longitudinal section with CEH observed as an incidental finding. The serosal margin is indicated by the white arrows. There are multiple fluid-filled cysts appearing as small 2 to 3 mm diameter anechoic (black) regions within the uterus throughout most of the segment. The scale is marked in 0.5 cm units. - To view this image in full size go to the IVIS website at www.ivis.org . -

In some cases there is also a small volume of free fluid present within the lumen of the uterus. Potentially, the condition can persist from one cycle to another, and become the cause of conception failure or early embryonic loss. CEH is typically an incidental finding during a routine ultrasound exam for pregnancy diagnosis or for a non-reproductive condition. Frequently bitches are examined for pregnancy at one month after breeding. If the bitch is not pregnant, careful attention to the ultrasonographic appearance of the uterus may demonstrate a low-grade endometrial hyperplasia. In non-breeding bitches, the occurrence of cystic endometrial hyperplasia is typically not documented and the existence of a uterine problem is only recognized when the condition progresses into a pyometra.

**Mucometra and Pyometra**
When CEH has resulted in the retention of fluid that is observed to be significant in volume and mucoid in nature, either upon clinical examination or in a gross pathological examination, the condition is termed mucometra (Fig. 4). The term pyometra refers to pus in the uterus, as a result of inflammation, and usually involves a bacterial infection. Continued endometrial hyperplasia and the secretion of fluid into the uterine lumen, combined with bacterial colonization of the uterus at estrus is the likely etiology of pyometra. The condition of pyometra most often develops during the luteal phase when the cervix is closed by the action of progesterone, and the opportunity for uterine discharge is diminished.

In most cases, the condition is first suspected on the basis of other clinical findings and after systemic effects have occurred, and often not until late metestrus (diestrus) or even anestrus. At this stage, ultrasound examination reveals that the uterus is enlarged and filled with fluid. Most commonly this enlargement and fluid collection is throughout the uterus, but enlargement may be localized to one uterine horn or a portion therein, or it may be segmental in both horns. (Fig. 4 and Fig. 5).

The fluid is frequently anechoic, but the presence of echogenic particles is common, and these can be seen to move with gentle ballottement at the time of imaging. The serosa and muscular layers of the uterine wall are usually edematous and may reach 2 to 3 mm in thickness. The endometrium is also thickened by hyperplasia and edema. The entire uterine wall may reach 7 - 10 mm in thickness. However, in some cases where the uterine lumen is distended with a significant volume of secretion or pus, the uterine wall may be compressed and/or reduced by pressure atrophy, and not show the enlargement associated with the underlying CEH. It has been reported that the ovarian end of the uterus has a target-like appearance in these cases [24], although the authors have not observed this. However, in many cases it is difficult to identify the proximal and distal regions of the uterus, and several sections of the uterus may be observed in one image (Fig. 5).

Ultrasound exams can help assess the extent and severity of pyometra in bitches. Affected bitches not intended for breeding are typically treated surgically. Medical treatments in bitches intended for breeding often includes, in addition to any antibiotic or supportive therapy, administration of prostaglandin F2a or prostaglandin-F analogs alone or in combination with a dopamine agonist (cabergoline or bromocriptine) to promote opening of the cervix and expulsion of uterine contents. Efficacy of medical treatment can and should be monitored using ultrasound, as a means to determine extent and completeness of efficacy in term of emptying the uterus of fluid content. Sonographic measurements of the reduction in the diameter of the uterus may be useful for monitoring medical treatment of cases of pyometra [25]. Treatment is best continued until there is an absence of fluid in all regions of the uterus.

**Uterine Stump Pyometra and Stump Granuloma**

Stump pyometra may follow the surgical removal of a pyometra, or the administration of exogenous progesterone to an ovariohysterectomized bitch in which the uterus has not been entirely removed. In these cases, the ultrasonographic appearance is similar to that for pyometra, with the exception that the lesions are only located at the pelvic inlet. In cases of uterine stump granuloma, there is frequently a mixed echogenicity appearance of the uterine stump, with hypoechoic regions resulting from the accumulation of pus or edema, and hyperechoic regions representing fibrous tissue reaction.

**Uterine Neoplasm**

Uterine neoplasms are rare in the bitch but have been diagnosed ultrasonographically [20,24]. Although no ultrasonographic findings can provide a diagnosis as definitive as histological evaluation, the appearance, location and clinical findings are usually sufficiently suggestive of the condition. Characteristically uterine neoplasms are homogeneous mass lesions attached to the uterine wall and project into the lumen. They may produce uterine fluid accumulation [24]. They may also be echogenic or have a complex mixed echogenicity if they are necrotic or fibrotic [19].

**3. Pregnancy**
Ultrasound imaging has been widely accepted as a routine method of pregnancy evaluation in many species since it allows the confirmation of pregnancy, the documentation of embryonic viability, and the detection of pregnancy abnormalities. When considering the features of a pregnancy that can be detected with ultrasound and their timing, it is important to remember that pregnancy does not necessarily commence upon the day of mating in the bitch, and that events should be related to day of the pre-ovulatory LH surge or to the time of ovulation which occurs 2 days after the LH surge (Table 1). This is especially true in the bitch where a pregnancy may result from mating as early as 5 days before ovulation or as late as 5 days after ovulation. The day of the LH surge is considered to be Day 0 of the cycle or of pregnancy. If ultrasound examinations are timed in relation to the time of a mating instead of the time of ovulation erroneous results may occur. Parturition typically occurs 64 to 66 days after the LH surge. The timing of events of gestation has been reviewed in detail elsewhere (click here to see the chapter by P.W. Concannon) [2].

Table 1. The age at first appearance of ultrasonographic features of early pregnancy typically observed in the bitch, expressed as days after the pre-ovulatory LH surge and days after ovulation. The ultrasound status of embryos or fetuses can be used to estimate gestational age. The gestational age can be used to predict the day of parturition based on parturition occurring 65 + 1 day after the LH surge in the vast majority of bitches. Based on England et al., [7] and Yeager et al., [16].

<table>
<thead>
<tr>
<th>Ultrasonographic Parameters Observed with a 5.0 or 7.5 MHz Transducer</th>
<th>Days after Ovulation</th>
<th>Days after LH Surge</th>
</tr>
</thead>
<tbody>
<tr>
<td>Detection of the conceptus as a 1 - 2 mm uterine vesicle</td>
<td>17 - 18</td>
<td>19 - 20</td>
</tr>
<tr>
<td>Presence of the embryonic mass within vesicle, at periphery</td>
<td>21 - 22</td>
<td>23 - 24</td>
</tr>
<tr>
<td>heartbeat</td>
<td>22 - 23</td>
<td>24 - 25</td>
</tr>
<tr>
<td>Identification of the yolk sac membrane</td>
<td>23 - 25</td>
<td>25 - 27</td>
</tr>
<tr>
<td>Identification of the allantoic membrane</td>
<td>25 - 29</td>
<td>27 - 31</td>
</tr>
<tr>
<td>Placenta develops zonary shape</td>
<td>25 - 27</td>
<td>27 - 29</td>
</tr>
<tr>
<td>Bipolar embryo shape</td>
<td>24 - 26</td>
<td>26 - 28</td>
</tr>
<tr>
<td>Differentiation of the head with focal anechoic area</td>
<td>25 - 28</td>
<td>27 - 30</td>
</tr>
<tr>
<td>Chorionic cavity exceeds the size of the yolk sac</td>
<td>26</td>
<td>28</td>
</tr>
<tr>
<td>Collapsing of the elongated yolk sac</td>
<td>29 - 32</td>
<td>31 - 34</td>
</tr>
<tr>
<td>Detection of dorsal tubular spinal column</td>
<td>28 - 34</td>
<td>30 - 36</td>
</tr>
<tr>
<td>Detection of formation of limb buds</td>
<td>31 - 33</td>
<td>33 - 35</td>
</tr>
<tr>
<td>Detection of formation of the axial skeleton</td>
<td>31 - 32</td>
<td>33 - 34</td>
</tr>
<tr>
<td>Anechoic stomach and urinary bladder become visible</td>
<td>34 - 36</td>
<td>36 - 38</td>
</tr>
<tr>
<td>Hyperechoic lung distinct from liver</td>
<td>36 - 38</td>
<td>38 - 40</td>
</tr>
<tr>
<td>Trunk diameter exceeds diameter of head</td>
<td>36 - 38</td>
<td>38 - 40</td>
</tr>
<tr>
<td>Trunk diameter exceeds 50% of chorionic cavity diameter</td>
<td>36 - 40</td>
<td>38 - 42</td>
</tr>
<tr>
<td>Crown-rump length exceeds length of placenta</td>
<td>36 - 40</td>
<td>38 - 42</td>
</tr>
<tr>
<td>Detection of kidneys</td>
<td>38 - 44</td>
<td>40 - 46</td>
</tr>
<tr>
<td>Detection of eyes</td>
<td>38 - 44</td>
<td>40 - 46</td>
</tr>
<tr>
<td>Detection of individual cardiac chambers</td>
<td>40</td>
<td>42</td>
</tr>
<tr>
<td>Trunk diameter exceeds 50% of uterine outside diameter</td>
<td>44 - 46</td>
<td>46 - 48</td>
</tr>
<tr>
<td>Detection of intestines</td>
<td>56 - 60</td>
<td>58 - 62</td>
</tr>
</tbody>
</table>

Diagnosis of Pregnancy
The uterine implantation vesicles, gestational sacs or embryonic yolk sacs can be first imaged approximately 17 to 18 days after ovulation, and thus 18 to 19 days after the LH surge, however, they are not seen distinctly or routinely until 18 days after ovulation, i.e., Day 20 after the LH surge [9,11]. The vesicles at this time appear as spherical anechoic structures within the lumen of the uterus. They are frequently 1 to 2 mm in diameter when first detected (Fig. 6a) and tend to increase in size at a specific rate (Fig. 6b, Table 2).

![Figure 6a. Day 20 uterine vesicle (2 mm gestational sac). Sonogram of a pregnant bitch showing the anechoic fluid within a gestational sac in the uterine lumen, in longitudinal plane, at Day 20 after the estimated day of the LH surge. The black arrows mark the outer diameter (serosal surface) of the uterine horn. The diameter of the gestational sac is 2 mm. The ruler at the left side of the image is marked in 2 mm increments. At the level of the embryonic vesicle, the uterine wall appears to be about 3 mm thick. Note the bright specular echoes present dorsally and ventrally to the anechoic sac. - To view this image in full size go to the IVIS website at www.ivis.org. -]

![Figure 6b. Day 22 uterine vesicle (gestational sac). Sonogram of a pregnant bitch at Day 21 after the estimated day of the LH surge. The anechoic spot in the upper left quadrant is the fluid within a gestational sac in the uterine lumen, in cross-section plane. No embryonic mass is visible. The diameter of the gestational sac is 4 mm, as shown in this view. The ruler at the left side of the image is marked in 2 mm increments. The uterine wall appears to be 3 mm thick. Note the bright specular echoes present dorsally and ventrally to the anechoic vesicle. - To view this image in full size go to the IVIS website at www.ivis.org. -]

| Table 2. Summary of uterine and fetal measurements useful for estimating canine gestational age and obtained from Beagle bitches studied sonographically at known days of gestation, with Day 0 being the day of the pre-ovulatory LH surge. Adapted from Yeager et al., [16] |
|------------------|------------------|------------------|------------------|------------------|
| Day of Gestation | Chorionic Cavity Diameter (mm) | Trunk Diameter (cm) | Crown-Rump Length (cm) | Bi-parietal Head Diameter (cm) |
| 20 | 2 | - | - | - |
| 22 | 4 | - | - | - |
| 24 | 6 | 0.1 | 0.2 | - |
| 26 | 10 | 0.3 | 0.8 | - |
| 28 | 12 | 0.5 | 1.0 | - |
| 30 | 15 | 0.6 | 1.2 | - |
| 32 | 17 | 0.7 | 1.6 | - |
| 34 | 20 | 0.9 | 2.0 | - |
| 36 | 25 | 1.1 | 2.8 | 1.0 |
| 38 | 28 | 1.3 | 4.6 | 1.1 |
| 40 | 33 | 1.5 | 5.0 | 1.3 |
| 42 | 34 | 1.8 | 6.0 | 1.4 |
| 44 | 34 | 2.1 | 6.8 | 1.5 |
| 46 | 34 | 2.5 | 8.0 | 1.7 |
| 48 | - | 3.0 | 9.1 | 1.8 |
| 50 | - | 3.6 | - | 1.9 |
| 52 | - | 3.7 | - | 2.0 |
| 54 | - | 3.8 | - | 2.1 |
Table 2. Summary of uterine and fetal measurements useful for estimating canine gestational age and obtained from Beagle bitches studied sonographically at known days of gestation, with Day 0 being the day of the pre-ovulatory LH surge. Adapted from Yeager et al., [16]

<table>
<thead>
<tr>
<th>Day of Gestation</th>
<th>Chorionic Cavity Diameter (mm)</th>
<th>Trunk Diameter (cm)</th>
<th>Crown-Rump Length (cm)</th>
<th>Bi-parietal Head Diameter (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>56</td>
<td>-</td>
<td>3.9</td>
<td>-</td>
<td>2.2</td>
</tr>
<tr>
<td>58</td>
<td>-</td>
<td>4.3</td>
<td>-</td>
<td>2.4</td>
</tr>
<tr>
<td>60</td>
<td>-</td>
<td>4.7</td>
<td>-</td>
<td>2.6</td>
</tr>
</tbody>
</table>

Figure 6c. Day 23 uterine vesicle (gestational sac) with embryo. Sonogram of a pregnant bitch showing (upper right quadrant) the anechoic fluid within an elongated gestational sac in the uterine lumen, in longitudinal plane, at Day 23 after the estimated day of the LH surge. The body of the embryo is visible within the fluid at the periphery of the sac as a small, 1 mm mass, consistent with the estimated gestational age. The apparent diameter of the gestational sac is 6 mm, the length 18 mm. The ruler at the left side of the image is marked in 2 mm increments. The uterine wall appears to be 3 - 4 mm thick. - To view this image in full size go to the IVIS website at www.ivis.org.

The uterine wall between adjacent conceptuses is generally less echogenic than the uterine wall surrounding each conceptus. The physical basis of these uterine vesicles at this stage, is not clear. They may represent pre-implantation uterine reactions and fluid accumulation, expanded blastocysts, or perhaps early developing embryonic yolk sacs. Over the next few days these vesicles or gestational sacs become elongated, filled with anechoic fluid, and may be absent any evidence of the developing embryo mass for 1 or 2 days. Careful examination of the uterine vesicle image at this stage often shows bright specular echoes present above and below the anechoic sphere (Fig. 6c).

Implantation and Early Gestation

At Day 20 - 21 after ovulation, i.e., Day 22 - 23 after the LH surge, the conceptus is approximately 7 mm in diameter and 15 mm in length and the embryo may be identified within the fluid filled vesicle [9,11]. However, elongated vesicles without visible embryos may be seen as late as Day 23 after the LH surge, and 21 days after ovulation (Fig. 6d and Fig. 6e).

Figure 6d. Day 24 uterine vesicle (gestational sac) with embryo. Sonogram of a pregnant bitch showing (upper left quadrant) the anechoic fluid within a gestational sac in the uterine lumen cross-section plane, at Day 24 after the estimated day of the LH surge. The body of the embryo is visible as a 1 - 2 mm mass at within the fluid at the periphery of the sac, consistent with the estimated gestational age. The diameter of the gestational sac is 6 to 8 mm in this view. The ruler at the left side of the image is marked in 2 mm increments. The uterine wall appears to be 3 - 4 mm thick. - To view this image in full size go to the IVIS website at www.ivis.org.

Figure 6e. Day 23 vesicle with no visible embryo, in longitudinal plane. This sonogram of a pregnant bitch shows an elongated, ellipsoid embryonic vesicle which is 5 mm in diameter at the center (black arrows). White arrows mark the segments of uterus between embryonic vesicles. The uterine walls between vesicles is less echoic that the uterine wall surrounding the vesicle. The scale at the right is marked in 5 mm units. - To view this image in full size go to the IVIS website at www.ivis.org.

It is around this time that implantation is reported to occur. Apposition of the embryonic membrane and the uterine mucosa, and expansion and lengthening of the vesicle occurs over the previous several days, but by around Day 22 after the LH surge, the embryonic membranes invade the uterine mucosa providing firm attachment [3]. At this time, the embryo itself often appears as a 2 to 3 mm homogeneous oblong-shaped hyperechoic structure, which becomes lifted away from the uterine wall and protrudes into the anechoic yolk sac. The embryo may be readily identified as a distinct mass within the fluid filled vesicle [9,11] by Day 22 to 23, and usually by Day 24 (22 days after ovulation). The embryonic heartbeat has been detected as a bright echogenic flicker as early as 21 days after ovulation, and 23 days after the LH surge (Fig. 7a and Fig. 7b).

Figure 7a. Day 24 vesicle and embryo. Sonogram of a pregnant bitch showing the anechoic fluid within
a gestational sac in the uterine lumen, in cross-section plane, at Day 24 after the estimated day of the LH surge. The embryonic mass is visible within the vesicle attached to the periphery, and is measured as 4 mm long by the white electronic calipers (x - x). In the real-time images, the embryonic heartbeat was visible as an hyperechoic flickering element within the embryo. The diameter of the gestational sac is 8 - 9 mm. The ruler for the sonogram is not shown. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 7b. Day 24 vesicle and embryo. Sonogram of another pregnant bitch showing the anechoic (dark) fluid within a gestational sac in the uterine lumen, in cross-section plane, at Day 24 after the estimated day of the LH surge. The embryo is 4 mm long and located at the periphery of the anechoic fluid-filled sac. The embryonic heartbeat was detectable as an hyperechoic flickering element within the embryo. The diameter of the gestational sac is 7 to 9 mm. The scale at the right is marked in 5 mm units. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 7c. Day 24 vesicle and embryo. Sonogram of a third Day 24 pregnant bitch showing cross section of embryonic mass at the periphery of the gestational sac. In this image, the uterine horn and vesicle are located dorsal to, and thus below, the large anechoic urinary bladder. - To view this image in full size go to the IVIS website at www.ivis.org . -

However, it is usually not detectable until Day 24 - 25 after the LH surge, at 22 to 23 days after ovulation, using 5 MHz or 7 MHz ultrasound heads. The heartbeat is usually first seen on the day that the embryonic mass is first seen (i.e., Day 24), or on the following day (i.e., Day 25 after the LH surge). Usually, by two more days, at 26 to 27 days after the LH surge, it is possible to identify a second fluid filled sac adjacent to the embryo. This is the developing allantois. As the allantois increases in volume it becomes positioned between the chorion and the yolk sac and it can be clearly identified as a separate structure. After about 25 days after ovulation and Day 27 after the LH surge, the yolk sac does not increase further in size, whilst the allantois continues to enlarge. It exceeds the yolk sac in volume from 26 days after ovulation and Day 28 after the LH surge (Fig. 7d).

Figure 7d. Day 28 embryo and embryonic membranes. Abdominal sonogram of a pregnant bitch 28 days after the LH surge, showing an anechoic (black), chorionic-fluid filled vesicle within a uterine horn. The uterine horn appears round in this transverse section, and is located in the right half of the image. The embryo appears as an echogenic bipolar (dumbbell shaped) mass. The embryo is 1 cm long. The scale is marked in 5 mm units. In this section, the yolk sac membrane appears as an obvious echoic irregular line that extends from one side of the embryo to the opposite side of the vesicle and returns to the embryo. Segments of the developing allantoic membrane appear as an interrupted faint thin line closer to the periphery and enclosing the yolk sac as well as the embryo. The scale at the right is marked in 5 mm units. - To view this image in full size go to the IVIS website at www.ivis.org . -

The two structures can usually be distinguished because the yolk sac membrane is more echogenic and becomes markedly folded and ultimately surrounded by the allantois at approximately Day 28 - 30 after ovulation and Day 30 - 32 after the LH surge. The yolk sac remnant may be distinctly visible within the amniotic cavity throughout mid-gestation (Fig. 7e). A further fluid-filled structure, the amnion, which closely surrounds the embryo, is identifiable from approximately 24 days after ovulation (Fig. 7f).

The time at which the heartbeat can be first detected varies with the instrumentation, the thickness of the body wall, and other factors. It is usually very easy to detect by Day 28 after the LH surge. Thus, 4 weeks after the estimated time of the LH surge, 26 days after ovulation, or one month after mating are appropriate times to schedule an initial ultrasound examination for pregnancy evaluation.

Figure 7e. Day 36 yolk sac and placenta. Sonogram showing an uterine gestational sac in longitudinal plane at Day 36 of gestation. In this image the long, echogenic remnant of the tubular yolk sac can be seen to extend the full length of the chorionic sac, being attached at each end to the chorionic poles. The chorionic sac containing the dark, anechoic chorionic fluid extends at each end beyond the margins of the placental girdle. Sections through the placental girdle appear as echogenic bands above and below the yolk sac in this image. The marginal hematomas of the placenta appear as small thickenings at each end of the sections through the placenta. No parts of the embryo proper appear in this sonogram. The electronic ruler at the right hand
edge of the sonogram is marked in cm. - To view this image in full size go to the IVIS website at www.ivis.org. -

**Mid Gestation**

The embryo has a bipolar shape by Day 28 of pregnancy, or 26 days after ovulation. The head region is identifiable as containing an anechoic area by Day 30, or 28 days after ovulation. The limb buds are usually identifiable from Day 32 to 34, i.e., 30 to 32 days after ovulation, onwards. The crown-rump length of the embryo remains less than the length of the placental girdle until at least until Day 36 (Fig. 8a). The fetal skeleton is evident by Day 34 of pregnancy, or 32 days after ovulation, and thereafter, when fetal bone appears hyperechoic and casts acoustical shadows (Fig. 8b). The bones of the head appear first, followed by those of the lower body. At this stage the hyperechoic heart valves can be imaged and are seen to be moving, and the great vessels can be traced cranially and caudally. In general, from Day 38 to 40 onwards, i.e., from 36 to 38 days after ovulation, the trunk diameter exceeds that of the head (Fig. 8c and Fig. 9b).

![Figure 8a. Bipolar Day 35 embryo. Sonogram showing a segment of uterus in sagittal section at a placental site at approximately 35 days after the LH peak, with the echogenic embryo shown in longitudinal section. The electronic calipers (+ - +) and dotted line indicate the measurement of the crown-rump length. The embryo is positioned with its dorsal side opposed to the dorsal aspect (bottom of image) of the placental girdle. The brighter, heteroechoic lines immediately ventral to (above) the embryo probably reflect the remnant of the yolk sac which extends toward the two poles of the chorionic vesicle. The enlargements of the placental girdle at each end of the sections through the placenta and which protrude into the vesicle and away from the uterine surface are the marginal hematomas. Other sonic sections through the embryo revealed distinct limb buds. The embryo is decidedly shorter than the placental girdle at this stage. - To view this image in full size go to the IVIS website at www.ivis.org. -](image)

![Figure 8b. Day 32 embryo. Section similar to that in Figure 8a, obtained in a pregnant bitch at Day 32 post LH surge, showing the fetus in sagittal section. The crown-rump length of 1.6 cm is approximated by the electronic calipers. The head shows evidence of the shape of the snout. The bright hyperechoic dot-like elements represent bone formation in the head and thorax. Limb buds were evident in real-time scanning but are not evident in this image. - To view this image in full size go to the IVIS website at www.ivis.org. -](image)

![Figure 8c. Sonogram of a pregnant bitch 37 days after the LH surge, showing the fetus in dorsal section from the head (left) to caudal abdomen (right). The neck does not appear in this section and an anechoic space is seen between the head and the top of the torso and the thoracic limb buds. The fetus crown-rump length is less than the length of the placental girdle, suggesting the fetal age is slightly less than 38 - 40 days after the LH surge. The placenta appears in section as two echogenic bands, above and below the fetus. The marginal hematomas at each end of the placenta appear as pairs of inwardly curved ends of the longitudinal sections through the placental girdle. In this view, developing bones in the cranium and mandible appear as hyperechoic, bright white elements in the head. The mandible has a "V" shape. The developing long bones in the fore-limb buds are visible as bright spots in cranio-lateral aspects of the fetal torso. The amniotic membrane of the amniotic sac can be seen as a thin echo genic line surrounding the fetus. The thick yolk sac membrane is seen near the chorionic pole extending to the left of the placental hematoma near the head. The scale on the right is marked in 5 mm increments. - To view this image in full size go to the IVIS website at www.ivis.org. -](image)

**Placenta**

The zonary, circumferential placenta wraps around the central portion of the conceptus like a waistband (Fig. 9a), and is observed ultrasonographically between the fetus and the uterine walls in all planes. When imaged in the longitudinal plane, the placenta appears as two thick bands one on either side of the fetus, between the fetus and the uterine wall (Fig. 8c and Fig. 9b). In this plane, the curved and thickened edge of the placenta is seen, and represents the region of the marginal hematoma at each end of the placental girdle (Fig. 7e and Fig. 8c). From Day 38 to 40 after ovulation, onward, the crown-rump length of the fetus exceeds the length of the placenta (Fig. 9b). When viewed in cross section or transverse section the placenta appears as a thick band which surrounds the embryo, extra embryonic fluid and embryonic membranes (Fig. 9c).

![Figure 9a. A Day 44 Beagle fetus shown within an intact chorionic membrane and surrounded by](image)
allantoic fluid. The head, thorax, abdomen and forelimbs are visible above the placenta. The zonary, circumferential placenta (placental girdle) has an irregular, mottled outer surface, whereas the inner surface is smooth and glistening (not shown). The irregular tan and red surface is the result of the detachment of the placenta from the endometrial (uterine) component of the placenta and presumably has tags of uterine tissue embedded in it. The marginal hematomas are visible as green margins on each end of the placental girdle. A fetal crown-rump length longer than the placental girdle is consistent with the age of the fetus. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 9b. Day 42 fetus with early skeleton and placental girdle in longitudinal plane. Sonogram of a uterine segment of a Day 42 pregnant Beagle bitch, showing a fetus in dorsal plane. The hyperechoic (bright white) double-line visible along the dorsal midline of the fetus is the developing vertebral column running from the head (left) to the rump (right). The crown-rump (CR length) is clearly greater than the 4 cm long placental girdle, that encircles the fetus. The girdle is seen in longitudinal view as two thickened echogenic bands immediately lateral to the fetus, and located immediately above and below the fetus in this view. The placental sections show slight evidence of the formation of thickened marginal hematomas, appearing as a hyperechoic thickening at each end of the placental section below the fetus. The hyperechoic elements in the head and neck represent bone formation in the skull and cervical vertebrae. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 9c. Early spinal column. Sonogram showing a Day 34 embryo in dorsal section at the level of the developing spinal column which appears as two parallel hyperechoic lines. - To view this image in full size go to the IVIS website at www.ivis.org . -

At about 34 to 36 days after ovulation, i.e., after Day 36 - 38 after the LH surge, it is possible to identify the fluid-filled fetal stomach caudal to the liver in more than 90% of fetuses (Fig. 10a). A day or so later the fetal bladder is identifiable in the caudal abdomen and with careful examination the urachus may be imaged (Fig. 10b). These changes are obvious by Day 40 - 45 after the LH surge (Fig. 10b, Fig. 10c, Fig. 10d and Fig. 10e)

Figure 10a. Transverse section of Day 45 fetus and fetal stomach. Abdominal sonogram of pregnant bitch around 45 days after the LH, with an oblique section through a gestational sac near the center of the image. The sonic section passes through the 2.25 cm diameter cranial abdomen of the fetus. This sectional view passes through the very echogenic (bright grey-white) placental girdle (marked +) on one side of the fetus, but not the other because the section imaged is near the edge of the circumferential placental girdle. The anechoic area around the fetus and between the fetus and placenta is the allantoic fluid which fills the gestational sac. The sonic section through the fetal abdomen shows the vertebral column as an echogenic (bright) spot near the periphery of the body, shows the liver as homogenous echogenic (grey) tissue, and shows the stomach as an acentric, anechoic (black) spot of 5 mm diameter to the left of the vertebral column. The scale at the right is marked 0.5 cm units. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 10b. Day 43 fetal neck and trunk including skeletal elements, heart and bladder. Sonogram of a pregnant bitch approximately 43 days after the LH surge. The fetus is slightly longer than the length of the placental girdle. The dorsal section of the fetus shows the base of the skull and the vertebræ of the neck extending to the right, and the two hind limbs at the left. At least one hind limb shows hyperechoic evidence of bone. There are hyperechoic (white) sonic sections through the ribs on either side of the thorax above and below the anechoic (black) chambers of the heart and great vessels. The heart is surrounded by lung which is hyperechoic (brighter) compared to the liver. The round anechoic (black) mass in the caudal abdomen is urine in the fetal urinary bladder. The fetal stomach is not present in this section. The scale at the right is marked 0.5 cm units. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 10c. Day 43 fetal head. Sonogram providing a frontal/coronal section through the head of the
same fetus of a Day 43 pregnant bitch shown in figure 10b. The sonic section is longitudinally through
the uterus; the longitudinal sections through the placental girdle are well to the left of the fetal head; the
fetal head lies within the anechoic (dark) allantoic cavity which projects far beyond the margins of the
placenta at each end of the placental site. The c-shaped parietal bones of the skull enclose the brain. The hyperechoic bones
(bright white) projecting to the left of the cranium represent developing bones in the base of the skull and/or mandibular
bones. The biparental diameter is about 1.3 cm; the scale on the right is marked in 0.5 cm units. - To view this image in full
size go to the IVIS website at www.ivis.org . -

Figure 10d. Day 47 fetal head and neck. Sonogram of a Day 47 pregnant bitch showing a fetal head
(right) and neck (left) in frontal section. The hyperechoic bones (bright white) of the calvarium outline
the skull. The bones of the vertebrae are likewise hyperechoic. The bright echogenic area below the
head is an acoustical shadow caused by the density of the head bones. The scale on the left is marked in
1.0 cm increments. - To view this image in full size go to the IVIS website at www.ivis.org . -

Late Gestation
The skeleton becomes more obvious in late pregnancy and the skull, spinal column and ribs are easily identifiable (Fig. 11a,
Fig. 11b).

Figure 11a. Day 46 fetus. Sonogram of a uterine segment of a Day 46 pregnant Beagle bitch, showing a
fetus with a crown-rump length exceeding the length of the placental girdle. The image shows the
echogenic round head within anechoic fluid oriented to the left, and the body in the center of the image
is aligned horizontally almost entirely within the placental girdle. The placental girdle is shown in
sections above the fetus and below the fetus. These sections through the placenta have at their left and
right margins a more echogenic round section which is probably a section through the marginal hematoma at that periphery
of the placental girdle. The head of the fetus (left) has two hyperechoic (bright white) spots, which are sonic sections
through the mandible. Between the head and body, the neck tissue is not visible in this plane, and the region is composed of
anechoic (black) embryonic fluid and a section through the amniotic membrane. The section through the thorax shows the
chambers of the heart (and major vessels) as two large anechoic fluid-filled cavities with extensions. The echoic lungs are
seen on either side of the heart. Caudal to the lungs and heart is the less echoic liver. Caudal to the liver, the anechoic spot
within a triangle of echoic tissue in the abdomen is the fluid in the fetal urinary bladder. The hyperechoic spots lateral to the
lungs at the top of the image are sonic sections through ribs. - To view this image in full size go to the IVIS website at
www.ivis.org . -

Figure 11b. Day 40 fetal skull and thorax. Sonogram of canine fetus approximately Day 40 of
gestation, sectioned in dorsal plane though the head (right), neck, thorax, and upper abdomen (left).
The fetus is surrounded by anechoic (black) allantoic fluid. Sections of the placental girdle are above
and below the thorax. The fetal head shows the hyperechoic (bright white) parietal bones lateral on the
either side of the skull. On either side of the neck, the membrane of the amnion is faintly seen in
section, with some of the small amount of anechoic amniotic fluid which surrounds the fetus on the interior of the amniotic
membrane visible between the membrane and the neck. The sonic section of the thorax intersects multiple hyperechoic
(bright white) ribs, anechoic major vessels at the level of the heart, and the echoic lungs on either side of the heart. - To
view this image in full size go to the IVIS website at www.ivis.org . -

In the last 20 days of pregnancy, the kidneys can be imaged (Table 1) and they are frequently more echogenic than
observed in the adult animal (Fig. 11c).

Figure 11c. Day 47 fetal stomach and kidneys. Sonogram of a bitch 47 days pregnant, containing a
dorsal sonic section through the fetus from the upper thorax (right) to the lower abdomen (left). The
two rows of hyperechoic (bright white) spots are sonic sections through the ribs on either side of the
lungs and the anechoic (dark) chambers of the heart. The stomach (S) appears and an anechoic
structure just caudal to the ribs. The left kidney (K) and right kidney appear as oval, bilaterally
symmetrical, echoic structures with central anechoic urine filled renal pelvis. - To view this image in full size go to the
IVIS website at www.ivis.org . -

Fetal vasculature becomes obvious, and the umbilical vessels may be traced from the liver to the umbilicus. In late
pregnancy, the intestine (Fig. 11d) may be detected after Day 58 [16]. In cross section view, the vertebrae appear as hyperechoic elements dorsal to the aorta, and the liver and stomach can be identified as echogenic and anechoic elements respectively (Fig. 11e).

Figure 11d. Day 52 fetal head and neck in dorsal plane. Sonogram of a canine fetus at Day 52 of gestation, with the cranial portion of the fetus shown as a sonic dorsal section. The parietal bones, which appear as white curved lines (marked by white arrows), and the vertebrae (V) are substantially mineralized based on the very hyperechoic image (white color) and the casting of acoustic shadows (dark areas in the image below these skeletal structures). Mineralization of the fetal skeleton becomes obvious in the last third of gestation, after Day 42. The bi-parietal diameter of 2.0 cm shown by the electronic calipers (+ --- +) can be used to estimate gestational age. The ruler at the right side of the image is marked in 0.5 cm increments. - To view this image in full size go to the IVIS website at www.ivis.org.

Figure 11e. Day 52 fetal abdomen in transverse section. Sonogram of fetus in transverse plan at the level of the abdomen in a 52 days pregnant bitch, estimated to be 13 days prior to parturition. The fetal diameter is 3.3 cm. The fetus is located within the zonary placenta, which appears as a 0.9 cm thick ring of echoic tissue surrounding the fetus. At this time, during late gestation, there is normally little or no appears fluid detected between the fetus and the placenta. The aorta (a) and individual viscera are distinct, including the liver (l) and stomach (s). An hyperechoic vertebra is seen just dorsal to (and thus below in this view) the aorta. The mean body diameter is used to estimate gestational age in conjunction with the bi-parietal skull diameter (Fig. 11f). The ruler at the right side of the image is marked in 0.5 cm increments. - To view this image in full size go to the IVIS website at www.ivis.org.

**Accuracy of Pregnancy Diagnosis with Ultrasound**

Ultrasound examination in the first month after mating may lead to an inaccurate pregnancy diagnosis if the variation between mating time and ovulation time (see above) is not considered, and the examination is scheduled too early in relation to the time of ovulation. In the worse case, a pregnant bitch may be examined earlier than when a pregnancy diagnosis can be performed leading to a false diagnosis of non-pregnancy. When looking for fetal heart movements as the criterion of pregnancy, the ultrasound exam should be scheduled about 26 to 30 days after the estimated day of ovulation (i.e., Day 28 to 32 after the LH surge), in bitches that have been monitored carefully to estimate the day of ovulation. If timing is based on observed or suspected breeding alone, then the exam should be conducted 31 - 33 days after that breeding, since bitches may become pregnant following a breeding that occurs as early as 5 days before ovulation and 3 days before the LH surge.

False negative diagnoses may also be produced by overlooking a conceptus, or due to acoustic artifacts produced by gas or fecal material hiding a conceptus. False positive diagnoses [26] may be the result of the confusion of empty loops of small intestine with early pregnancy. However, the intestine can be shown to be tubular by imaging in two planes, whereas early pregnancy involves the segmental occurrence of individual vesicles in the uterine lumen. Resorption of an entire litter may be missed if the initial ultrasound exam is delayed much beyond 30 days, and inaccurately reported as a failure to conceive.

**Estimation of Fetal Numbers**

The accuracy of detecting absolute fetal numbers is poor [6,27]. England [4] suggested that for the bitch the greatest accuracy was before Day 30 after the LH surge when 38% of examinations were successful in predicting fetal numbers [4]. Generally, the numbers of fetuses are underestimated, with the error being associated with overlooking fetuses, mistaking them as already counted or due to acoustic artifacts. The accuracy was reduced for larger litters, and bitches were classified as having either five pups or more, or four pups or less, and the efficiency of prediction on that basis was 97% [4]. The accuracy of predicting the actual fetal number is low later in pregnancy. For examinations between 30 and 50 days of pregnancy, the accuracy was 18%, and after 50 days of pregnancy it was 8% [4]. Fetal resorption may also produce a disparity between the number of conceptuses imaged and the number of offspring born.

**Determination of Gestational Age**

A number of studies have examined the size of various components of the fetus in relation to the gestational age. These studies are useful within specific breeds for the calculation of gestational age, and may also indicate whether pregnancies are developing normally (see below). The timing of the first ultrasonographic appearance of certain organs may be even more useful for the estimation of gestational age and prediction of parturition. For example the kidneys are usually only visible within the last 20 days of gestation when using a 5.0 MHz transducer. The timing of the first appearance of anatomical landmarks or relationships during pregnancy are useful since they appear to be constant between breeds (Table 1).
In contrast, the application of exact fetal measurements as criteria to describe the stage of pregnancy is less useful, as these can vary among bitches of different size breeds, especially during the phase of rapid fetal growth after Day 30. Nevertheless, a reasonable degree of accuracy can be obtained using fetal measurements (see below).

**Prediction of Parturition using Ultrasound**

The majority of pregnant bitches give birth 64 to 66 days after the LH surge (click here to see chapter by Concannon) [2]. Accurate estimation of the day of the LH surge is the best means to predict the date of parturition. However, the sonographic appearance of the embryos or fetuses can often indicate their gestational age accurately enough to allow a prediction of the time of parturition with an accuracy of plus or minus 2 days.

The gestational age of the embryos determined at the time of an ultrasound exam conducted to confirm or diagnose pregnancy, typically performed 4 to 5 weeks after mating, can be estimated by measuring 3 or more of the following parameters for each of 2 or more fetuses:

1. Gestational sac (or chorionic cavity) diameter (Fig. 12a) in early pregnancy
2. Crown-rump length (Fig. 12b)
3. Body (abdominal) diameter at the level of the liver and stomach (Fig. 12c and Fig. 12d)
4. Biparietal diameter (Fig. 12e) after mid-gestation

All 3 fetal measurements were shown to be correlated to gestational age. From days 20 to 37, chorionic cavity (or gestational sac) diameter was the best predictor of gestational age. From days 38 to 60, head (bi-parietal) diameter was the most accurate predictor [16]. Such a method of predicting the date of parturition is particularly useful in pregnancies in which the day of the LH surge was not estimated with any degree of accuracy.

In one set of studies the ability to determine the day of parturition within 2 days was about 80 to 85% accurate in medium and large breeds of bitches studied at 28 to 32 days after the estimated day of the LH surge, comparing published values with those obtained for 3 or more of the parameters listed above [34,35]. Accuracy of predicting the whelping date was independent of litter size. The method was less accurate for toy, miniature and giant breeds. In applying correction factors to account for breed differences in overall size, to obtain a prediction accuracy of +2 days in 80% or more of the cases, one
day needed to be subtracted from the estimated day of whelping for toy and miniature breeds, and 2 days had to be added for giant breeds. The data on which the estimates were made are those summarized in Table 2. Using these parameters in the last 3 weeks of gestation was less accurate (about 50%). The use of bi-parietal skull diameters (Table 2) together with body diameters was preferred at that stage.

Another parameter that is useful to evaluate when ultrasound is conducted shortly after mid gestation is crown-rump length in relation to the length of the placental girdle. The crown-rump length is typically less than the length of the placenta before Day 38 of pregnancy, and then becomes longer than the placenta so as to very clearly exceed the length of the placenta from Day 40 - 42 onward (Fig. 9c and Fig. 9d).

4. Gestational Abnormalities

Embryonic Resorption

Should embryonic death occur before 35 days after ovulation, there is usually complete resorption of the conceptus. This can occur without vaginal discharge as late as Day 30. The incidence of resorption is not known, although Andersen and Simpson [31] reported the frequency in bitches to be approximately 11% in relation to the number of corpora lutea. Resorption of multiple conceptuses with continuation of a pregnancy in the absence of clinical disease has been reported [36-38]. England [4] reported that 5% of bitches suffered isolated spontaneous embryonic resorption with continuation of the pregnancy. A second study demonstrated an incidence of 13% [22]. In some instances, entire litters of 3 to 8 fetuses have been observed to resorb over a 2 to 5 day period without clinical signs in apparently healthy bitches (Yeager and Concannon, unpublished observations).

The sonographic aspects of a resorption are generally a reduction in the volume of the conceptus, an increased echogenicity of the embryonic fluid (sometimes particles may be identified free-floating within the allantoic fluid), an absence of the embryonic heartbeat, disintegration of the embryonic mass and ultimately collapse of the conceptus with inward bulging of the uterine wall (Fig. 13a and Fig. 13b). The uterus often remains slightly enlarged in this region, and there may be a small volume of free lumenal fluid; the uterine wall often appears moderately hyperechoic.

**Figure 13a.** Embryo undergoing resorption at mid gestation. Sonogram of a gestational sac of a horn in transverse view, at the site of resorption and containing remnants of a dead embryo (*), with the serosal margin marked by white arrows. The embryo (*) had no heartbeat at the time of examination. Other abnormalities include echoic (dark gray) embryonic fluid (F) and irregular shape of the gestational sac. In a viable pregnancy, the fluid is normally anechoic (black) throughout, similar to that shown in one compartment here (f), and in transverse view the shape is normally circular and the contour smooth. - To view this image in full size go to the IVIS website at www.ivis.org . -

**Figure 13b.** Resorption site at mid gestation. Sonogram of gestational sac of another resorption site in the same animal, shown in transverse view, with embryonic fluid (F) but no embryo, and with the gestational sac having a greatly reduced size and a thicker placenta compared to the resorption site in Fig. 13a. In contrast, in normal pregnancy, gestational sacs are of equal size. The gestational sac fluid (F) here is also abnormally echogenic (grey instead of black) and the shape is abnormally irregular. - To view this image in full size go to the IVIS website at www.ivis.org . -

**Figure 13c.** Day 55 M-mode examination of fetal heart rate. Sonogram of a Day 55 canine fetus (top image) and M-mode display of the heartbeat (bottom image). The sonogram is centered on the fetal heart, the chambers and great vessels of which appear as anechoic (dark) areas within the fetal thorax. The thorax is delineated by the two lines of hyperechoic (bright white) ribs, above and below the heart in this view. - To view this image in full size go to the IVIS website at www.ivis.org . -

Fetuses Underdeveloped for their Gestational Age

Conceptuses which are small or otherwise underdeveloped and do not exhibit the expected features for a specific gestational age may be abnormal. They frequently fail to develop and are ultimately resorbed. In some cases a single
conceptus is identified as being underdeveloped by comparison with adjacent echographically normal conceptuses. In one set of studies (England, unpublished observations), at least 50% of conceptuses were lost when the day of first detecting an embryo was more than 2 days later than expected in cases of accurately timed ovulation (i.e., the embryo mass was identified later than Day 24, or 22 days after ovulation). A similar outcome was noted if the embryo was identified by Day 22 - 24, but the heartbeat was not present until more than 2 days later than expected (i.e., the heartbeat was first seen on Day 26 or later, rather than Day 24 or 25 of pregnancy; or at 24 days after ovulation, or later, rather than 22 or 23 days after ovulation).

**Fetal Abortion**

Abortion and vaginal discharge usually follows the deaths of fetuses occurring after Day 35 of pregnancy and is associated with expulsion of fetal material and fluid. The owner does not always observe this, and it may be a surprise that the pregnancy has been lost. Many bitches will immediately eat the discharged material and lick the vulva of any evidence. The early features of fetal abortion are an increased echogenicity of the allantoic and amniotic fluid often with echogenic particles, followed by an absence of the fetal heartbeat and sometimes a thickening of the uterine wall. After expulsion, the uterus assumes an appearance that is similar to that observed in the postpartum bitch (see below).

**Fetal Abnormalities**

It is uncommon to detect fetal abnormalities in the bitch, since there are usually multiple fetuses and it is difficult to fully examine each. However, a number of striking abnormalities have been detected, some of which have necessitated delivery of the litter by cesarean operation. Such abnormalities include hydrocephalus, fetal anasarca, herniation of the ventral abdominal wall and fetal monsters [1,24]. In each case, the diagnosis was made by ultrasonographic identification of a change in the normal anatomical appearance of the fetus.

**Fetal Distress and Fetal Death**

At the time of a prolonged or difficult parturition, fetuses may become hypoxic. The fetal response is to become bradycardic. Fetal heart rate can be measured using B or M mode ultrasonography [28], and it is widely suggested that a decline in fetal heart rate to less than twice the maternal heart rate is indicative of fetal distress (hypoxia). Measurement of fetal heart rate may therefore be useful in decision-making when presented with a bitch with apparent dystocia, a bitch with a history of perinatal puppy death, or a bitch in which the gestational age has not been determined accurately. In one study, using both conventional and Doppler ultrasonography, normal fetal heart rates were usually > 220 BPM and measured more consistently when studied in Doppler mode [30]. An example of M-Mode ultrasound determination of fetal heart rate is shown in Fig. 13c. Fetal distress was considered severe with values < 180 BPM at Day 58 - 62 of pregnancy. In most cases, bowel movements were associated with fetal distress and were observed in all fetuses with heart rates under 180 BPM. Whether bowel movements at term were associated with pathological distress or with the physiological stress of parturition was not clear. The study also considered Day 58 to term fetuses to be growth-retarded if the bi-parietal head diameter was less than half the abdominal diameter, as these animals also tended to be born with much lower body weights.

Signs of fetal death detected by ultrasound include the following: absence of heartbeat, lack of fetal movement, reduced volume and increased echogenicity of fetal fluid, and accumulation of gas within the fetal stomach, other fetal cavities or the uterus. The latter should not be confused with artifacts produced by overlying intestine.

5. The Postpartum Uterus

**Normal Postpartum Changes**

Immediately after parturition the uterine body and horns are easily imaged. The horns remain enlarged and fluid-filled for a variable time after parturition. Central lumenal fluid is not invariably anechoic and may have echogenic regions within it. Uterine diameter decreases during the first two days after parturition and assumes a characteristic ultrasonographic appearance (Fig. 14a).

![Figure 14a. Postpartum 1 week. Abdominal sonogram of a bitch 1 week postpartum. The uterine horn is shown in longitudinal plane, with a placental site in the left half of the image. The uterine diameters (black arrows) at the placental site (P) versus that of the inter-placental site (j) were 1.6 cm versus 1.0 cm. The scale on the left has divisions every 2 mm. The still-enlarged uterine wall shows the circular and longitudinal myometrial layers as ecogenically distinct from one another. The endometrium at the inter-placental site (i) is hyperechoic (bright) compared to the more peripheral tissues and compared to the soft tissues at the placental site (P). - To view this image in full size go to the IVIS website at www.ivis.org. -](image_url)

In the first week the horns are composed of multiple layers of various echogenicity and have multiple discrete enlargements with hypoechoic centers at placental sites (Fig. 14b) [12]. Large variations are noted in uterine diameter between placental
sites and interplacental zones [23] (Fig. 14c and Fig. 14d). The uterus may return to the size noted during anestrus within 4 to 6 weeks; however, uterine involution is not complete ultrasonographically until 15 weeks postpartum.

Figure 14b. Postpartum 3 weeks, normal uterine involution. Abdominal sonogram of a pregnant bitch at 3 weeks after parturition. The uterine horn is shown in longitudinal plane, with a placental site (p) in the right half of the image. The uterine diameters (black arrows) at the placental site (p) versus interplacental site (j) were 1.2 cm versus 0.9 cm. The diameters are greatly reduced from those in the first week postpartum. The scale on the left has divisions every 2 mm. - To view this image in full size go to the IVIS website at www.ivis.org. -

Figure 14c. Postpartum 5 weeks, normal involution. Abdominal sonogram of a pregnant bitch 5 weeks after parturition. The uterine horn is shown in longitudinal plane, with a placental site in the right half of the image. The uterine diameters (black arrows) at the placental site (p) versus interplacental site (j) were 0.7 versus 0.5 cm, and greatly reduced from those at 1 week and 3 weeks postpartum. The scale on the left has divisions every 2 mm. - To view this image in full size go to the IVIS website at www.ivis.org. -

Figure 14d. Postpartum 15 weeks uterine involution. Abdominal sonogram of a pregnant bitch at 15 weeks after parturition. The uterine horn is shown in longitudinal plane. The uterine diameter (black arrows) was 0.5 cm throughout. The appearance of the uterus is indistinguishable from that of anestrus, i.e., the uterus is small, has no obvious layers, and placental versus interplacental sites are no longer distinguishable. The scale on the left has divisions every 2 mm. - To view this image in full size go to the IVIS website at www.ivis.org. -

Postpartum Abnormalities
The presence of retained fetuses may be suspected clinically by the an animal's behavior and a persistent vaginal discharge. Fetuses may be readily diagnosed with ultrasound by the presence of echogenic skeletons. Fetal viability may be assessed as previously discussed.

Placental retention is rare and is most commonly diagnosed on the basis of a persistent green-colored vulval discharge, with signs of malaise. The diagnosis of placental retention with ultrasound has not been reported, however, this may be complicated since soft tissue debris or blood clots may persist normally within the uterus after parturition [23], and may be indistinguishable from remnants of placental tissue. Sub-involution of placental sites (SIPS) can occur in the bitch. The echographic appearance of SIPS has not been reported.

6. The Ovary
The ovaries of the bitch are difficult to examine with ultrasound due to their small size, the fact that they are surrounded by a bursa, which may be fatty in older bitches, and their rather superficial location. The ovaries are typically located adjacent to the caudal pole of the kidney, level with the fifth lumbar vertebrae. They are positioned mid-abdominally in the standing bitch close to the lateral abdominal wall. Imaging of the ovary is therefore best achieved with the bitch either in the standing position after clipping the hair from the lateral abdominal wall, or with the bitch in dorso-lateral recumbence, with the transducer placed over the ventro-lateral abdomen. During estrus the position of the ovaries may vary slightly and they are often located more caudally and ventrally [6].

Follicles and Detection of Ovulation
Ovarian follicular development has been reported in the bitch, and a recent study documented presumptive follicular waves [8]. During late anestrus the hypoechoic ovaries tend to increase in size and small 1 - 2 mm fluid-filled follicles may be detected. This situation may be maintained for 30 or more days until the onset of proestrus when the number and size of follicles increases. The changes in ovarian sonographic appearance around the time of ovulation have been reported [13,15]. Follicles are quite obvious 5 days before ovulation, at which time the diameter of large follicles increases and reaches a maximum approximately 1 day after the LH surge. At this time the internal diameter of the largest follicles may be 8 or 9 mm, and there is frequently a change in outline to less spherical. Follicles tend to protrude above the margin of the ovarian stroma at this time (Fig. 15a). Several or all of the anechoic follicles in an ovary are typically replaced by hypoechoic or echoic structures at the time of ovulation, and seem to disappear (Fig. 15b).

In figure 15a, figure 15b and figure 15c, the right ovary of a Beagle bitch is shown shortly before ovulation (a), shortly after ovulation (b), and at 4 days after ovulation (c). The images were obtained at 1.5 days after the LH surge and before
ovulation (a); 2.5 days after the LH surge and after ovulation (b); and 6 days after the LH surge (c).

Figure 15a. The right ovary of a Beagle bitch examined at 1 day after the LH surge. The ovary appears in longitudinal plane. Three large pre-ovulatory follicles appear as anechoic spheres with thin walls. On of the follicles is measured at it largest diameter by the electronic calipers (x - x) as being 6 mm internal diameter, one day before ovulation. The scale on the left has divisions every 2 mm. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 15b. Ovary after ovulation. Sonogram showing the right ovary (longitudinal plane) of a Beagle bitch obtained at 2.5 days after the LH surge and shortly after ovulation which occurs at 2 days after the LH surge. This ovary at 0.5 day after ovulation had no distinct anechoic distinct follicles visible. The size of this ovary is somewhat decreased compared to one day before ovulation. The scale on the left side of the images is marked at 2 mm intervals. - To view this image in full size go to the IVIS website at www.ivis.org . -

This change, which is obvious for 1 - 2 days, can be detected as an acute event if the ovaries are examined daily by ultrasound. Whether this change in appearance involves a change in follicular fluid echogenicity, collapse of the follicle, bleeding into the follicle, or the rapid early proliferation of luteal tissue into the follicle antrum is not known. However, this echogenic appearance is only transient as the corpora lutea rapidly cavitate and again appear to be fluid filled and have an anechoic center 24 to 48 h later (Fig. 15c). Luteal phase ovaries can vary considerably in appearance because the echogenicity of the corpora lutea may not be greatly different from that of the ovarian stroma (Fig. 15c, Fig. 15d and Fig. 15e).

Figure 15c. Day 6 ovary with new corpora lutea. Sonogram showing in longitudinal plane the right ovary of a Beagle bitch obtained at 6 days after the LH surge and 4 days after ovulation. The ovary contained in total 2 large, 2 medium and 2 small anechoic structures with variable wall thickness. The small anechoic structure at the bottom of the image measured 4 mm at its maximal diameter using electronic calipers (x - x) and was considered a non-ovulating follicle. The other anechoic structures (two large and one small-sized) anechoic structures in this section were fluid filled cavities within rapidly developing corpora lutea from the recent ovulations, based on the pregnancy outcome. At this stage corpora lutea may be indistinguishable from pre-ovulatory and non-ovulatory follicles. Notice that the size of this ovary four days after ovulation is somewhat larger than an ovary containing pre-ovulatory follicles (Fig. 15 a). The centimeter ruler on the left side of the images is marked with lines at 2 mm intervals. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 15d. Mid-gestation luteal phase ovary. Sonogram of canine ovary in longitudinal view, marked by electronic calipers (x - x) as a 1.0 x 1.6 cm structure with four solid-appearing, round corpora lutea with diameters of approximately 0.6 cm. The image was obtained at a routine ultrasound pregnancy check in which this bitch was positive at Day 30 after the LH surge. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 15e. Mid-luteal phase ovary. Sonogram of a canine ovary in longitudinal view. The image was obtained during a routine pregnancy exam at approximately 30 days after the LH surge. The exam was negative. Note the several round, hypoechoic, 5 mm corpora lutea. The scale at the right is marked in 5 mm increments. - To view this image in full size go to the IVIS website at www.ivis.org . -

There may be variation among follicles within an ovary as to the extent of observed echogenicity of the follicle at the time of ovulation, and in the size and hypoechogenicity of the corpora lutea a few days after ovulation. Whilst the corpora lutea have thicker walls than pre-ovulatory follicles, they are quite difficult to interpret and for the inexperienced ultrasonographer it is not possible in a single examination to determine that ovulation has occurred until several days after ovulation when the luteal wall progressively thickens. Currently, the detection of ovulation using ultrasound has limited use for the detection of the optimal breeding time, especially when compared with the more simple method of measurement of plasma progesterone concentration as it increases immediately before ovulation and remains elevated.

Abnormalities of the Ovary
A small number of ovarian abnormalities have been reported in the bitch. In many cases the clinical history provides
presumptive evidence for the underlying condition, and ultrasound provides only confirmative information rather than being the primary means of diagnosis. Ovarian cysts and ovarian neoplasia have been described ultrasonographically, and ultrasonic confirmation of ovariectomy has been attempted.

**Ovarian Cysts**

It is important to remember that the most frequently observed ovarian cysts are cysts of the ovarian bursa. These cysts are endocrinologically inactive; they usually are thin-walled and are not clinically significant. They are not connected to the ovary per se and care should be taken to make sure that they are not interpreted as being clinically significant. True ovarian cysts originate from the ovarian stroma or follicles. They are usually multiloculated, with frequent irregular septae. Bilateral disease is not uncommon. Unfortunately in some cases the ultrasonographic appearance is bizarre. It is not possible to distinguish these structures from ovarian neoplasms. True ovarian cysts have been identified in up to 10% of bitches [33]. Follicular cysts may be associated with persistent or prolonged estrus, whilst luteal cysts are more commonly found in older bitches and may be associated with a persistent hemorrhagic vulval discharge. Luteal cysts may also be identified in bitches with pyometra. Should there be doubt over the significance of ovarian cystic lesions, ultrasound guided needle aspiration may be attempted, allowing measurement of hormone concentration within the cyst fluid, but this has not been reported, and the expected hormone concentrations in particular situations have not been reported either. The occurrence of polycystic ovaries has been documented in the dog using ultrasound. The significance of these lesions is uncertain, although they are more commonly found associated with the first estrus, or after an estrus-induction regime using exogenous gonadotrophins (Fig. 16a, Fig. 16b, Fig. 16c and Fig. 16d).

**Ovarian Neoplasia**

Ovarian neoplasia is not common in the bitch. The most common neoplasm is the granulosa cell tumor that may become large and cystic. It may be difficult to distinguish this structure from cystic ovarian disease, although with neoplasia there is more commonly the presence of a moderate volume of ascitic fluid (Fig. 17a, Fig. 17b and Fig. 17c).

The neoplastic ovaries are frequently markedly enlarged and consist of mixed echogenicity tissue. This is often
characterized by hyperechoic fibrous tissue, heterogeneous neoplastic tissue and anechoic cystic regions [29]. Bilateral ovarian adenocarcinoma was reported in the bitch [21], and was characterized by large ovaries containing multiple irregular cystic structures. The clinical signs of these tumors may include persistent estrus or cystic endometrial hyperplasia if they secrete estrogen or progesterone, respectively, however, clinical signs may not be noticed until the tumor is advanced.

Figure 17b. Ovarian carcinoma (presumptive). Sonogram showing right ovary of same animal. Ovary is shown in longitudinal view, marked by electronic calipers ( + - + and x - x) as a large structure, 2.0 x 2.8 cm in size, and has slightly irregular contours and consisting mostly of a heteroechoic, solid-appearing, soft-tissue mass, consistent with ovarian neoplasia. - To view this image in full size go to the IVIS website at www.ivis.org . -

Figure 17c. Ovarian carcinoma (presumptive), sonogram of left cranial abdomen of same animal in longitudinal view. The spleen (SPL) is surrounded by a large volume of echoic (dark gray) peritoneal fluid, increasing the suspicion of an ovarian carcinoma. Echos in the peritoneal fluid indicate that it is not a pure transudate. Analysis of the peritoneal fluid sample revealed that it was hemorrhagic and contained clusters of malignant epithelial cells. - To view this image in full size go to the IVIS website at www.ivis.org . -

Absence of Ovaries
It has been postulated that ultrasound examination may be useful for the detection of bitches with an unknown history that have had an ovariectomy or an ovariohysterectomy. This would negate the requirement for unnecessary laparotomy. Whilst Boyd et al., [18] found that this was possible in a number of cases, the precision of the technique is not sufficiently high, and the development of endocrinological testing methods means that this method is not widely employed.

7. Conclusion
Ultrasonography is a rapidly advancing diagnostic technology that has much to offer to the theriogenologists and practitioners specializing in canine reproduction. Ultrasound imaging has a major role in the documentation of normal physiological events as well as the diagnosis and staging of pregnancy. In addition, it is particularly suitable for the detection of pathological changes within the reproductive tract that may be demonstrated by changes in the size, shape, margination or internal architecture of the reproductive organs.

References


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