

Fetal Sex Determination in Cattle by Ultrasonography

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Abstract: After explaining the possible economical interest of fetal sex determination, the authors describe the development of the external genitalia of the bovine fetus. They explain that sex determination can be performed during two different periods of the pregnancy : an early period (from day 55 to day 65) and a late period (from day 80 to day 100). The sex determination technique using ultrasonography is described in detail. This technique was applied to 107 cows under farm conditions. Difficulties and possible errors of diagnosis are also mentioned.

Key words: cattle, fetus, sexing, ultrasound

Introduction

The first fetal sex determination technique using ultrasonography was described by Müller and Wittkowski in 1986. The sex diagnosis was performed between day 70 and day 120 of the pregnancy and consisted of looking for the male scrotum or the female mammary teats. This technique is now considered as a late fetal sex diagnosis. Sex diagnosis consisting of a search for the genital tubercle between day 55 and day 65 of the pregnancy was described for the first time by Curran *et al.* in 1989. The genital tubercle will give the male penis or the female clitoris. Its bilobate and hyperechogenic picture makes it easily identifiable by ultrasonography. The genital tubercle migrates towards the umbilical cord for the male fetus whereas it migrates towards the base of the tail for the female fetus. This last technique is considered as an early fetal sex diagnosis. It can however only be used for the species in which the male genital tubercle migrates towards the umbilical cord. Those species are mainly cows and horses.

Usage of the fetal sex determination: Fetal gender determination is particularly useful in dairy farms where a female calf is much more valuable than a male one.

Knowing the sex of the fetus will help :

- * to determine whether it is worth keeping an old pregnant cow (as opposed to sending it immediately to the abattoir).
- * to increase the price of a pregnant cow
- * to determine whether a caesarean section is financially justified in case of dystocia calving.
- * to detect twins comprising one male and one female which could lead to the birth of a female freemartin calf.

Development of the external genitalia in bovine fetus: Fetal sex determination cannot be performed before day 50. Before day 45, the genital tubercle cannot be seen using ultrasonography. Between day 45 and day 50, the genital tubercle is very difficult to see between the rear limbs and will not start moving before day 50.

From day 50, the genital tubercle starts moving towards the umbilical cord in the male fetus and towards the tail in the female fetus. The move only becomes really clear from day 52. In practice, it is advised to wait until day 55 before performing the sex determination. On day 55, the genital tubercle is halfway between its initial position (between the rear limbs) and its final position (close to the umbilical cord or to the tail). The difference between a male fetus and a female fetus is by then very clear.

On day 60, the genital tubercle reaches its final position. In the male fetus, it is located at the back of the umbilical cord. In the female fetus, it is located under the tail. The genital tubercle is very easy to see between day 60 and day 65 : it looks like a bilobate and hyperechogenic structure. It is composed of two hyperechogenic dashes which make it look like an equal symbol (=). In fact, histology shows that the genital tubercle is unilobar. Bilobate images are produced by a specular reflection.

In the male fetus, the body of the penis can be seen from day 60 – this is the proximal part of the penis. The body of the penis is located in the sagittal plane and looks like an echogenic line from the anus to the umbilical cord. It can only be seen with difficulties on a frontal section.

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In the male fetus, scrotal swellings (which will become the scrotum in bull) can be seen just before day 60. Scrotal swellings look like two echogenic spots located between the rear limbs on both side of the penis.

From day 65, the fetus can only be seen with difficulty as it moves more deeply inside the pelvic cavity because of its weight. The sex determination is therefore rather difficult.

Between day 70 and day 75, the sex determination is particularly difficult for the male fetus : the genital tubercle loses its bilobate and hyperechogenic structure while turning into a penis. It becomes hypoechogenic and therefore difficult to identify using ultrasonography.

From day 75, the penis is fully formed. The extremity of the penis is the only part which can be seen by ultrasonography. It looks like a round hyperechogenic shape located at the back of the umbilical cord. At this stage, the scrotum can be seen between the rear limbs.

From day 80, the mammary teats become visible in the female fetus. They appear as four hyperechogenic points located in the inguinal area of the fetus. The four mammary teats are in a trapezoid shape. They can only be seen in a frontal section.

After day 100, the fetus is deep inside the abdominal cavity. The great size of the fetus prevents from obtaining favourable sections for the gender determination.

In the female fetus, the genital tubercle becomes a clitoris around day 100. The clitoris looks like a globulous and echogenic structure in the shape of a triangle which is located under the tail. It is however very rare to clearly see the typical triangle shape of the clitoris because the fetus is too deep inside the abdominal cavity.

In the male fetus, both vaginal cavities move into the scrotum between day 105 and day 110. They look like two ovoid and almost anechogenic structures. They are located in the proximal part of the scrotum. They are separated from each other by the scrotal septum. The testes cannot be seen inside the scrotum at this stage of the pregnancy.

Gender determination using this technique is impossible after day 120.

Sex determination periods:

Three periods can be identified:

- * an early stage : from day 55 to day 65
- * an intermediate stage : from day 65 to day 80
- * a late stage : from day 80 to day 100

In practice, sex determination should be preferably performed during the early or late stages. The intermediate stage should be avoided. (Fig. 1)

During the early stage, gender determination is based on the relative location of the genital tubercle. If the genital tubercle moves toward the umbilical cord, the fetus is a male. If the genital tubercle moves toward the tail, the fetus is a female. The scrotal swellings and the body of the penis of the male fetus can also be seen after day 60 using ultrasonography.

Between day 55 and day 60, the fetus must be examined on a frontal section.

Between day 60 and day 65, the fetus can either be examined on a frontal section or on a transversal section.

During the intermediate stage, gender determination is performed by looking for the presence of the genital tubercle, penis and scrotum of the male fetus or the genital tubercle of the female fetus.

Both frontal or transversal sections can be used but frontal sections should be preferably used.

Sex determination is difficult at this stage because the fetus has moved deeper into the abdominal cavity.

During the late stage, sex determination is based on searching for the scrotum and penis of the male fetus or the mammary teats and genital tubercle of the female fetus.

Transversal sections should first be used to identify an eventual scrotum. If no scrotum can be seen, frontal sections should be used for the research of the presence of mammary teats.

Sex diagnosis technique: The rectum must first be emptied. Any pressure on the uterus should be avoided during this process as they could move the fetus.

The original position of the fetus is usually favourable to gender determination (a frontal section is very likely). Pressure on the uterus could also make the fetus move deeply inside the pelvic cavity, preventing the gender determination.

For the same reason, holding both uterine horns in his hand (as usually performed during a classic examination of the genitalia) should be avoided as this operation could lead to a displacement of the fetus.

The operator can then introduce the probe inside the rectum. The probe shall be resting on the rectum floor in order

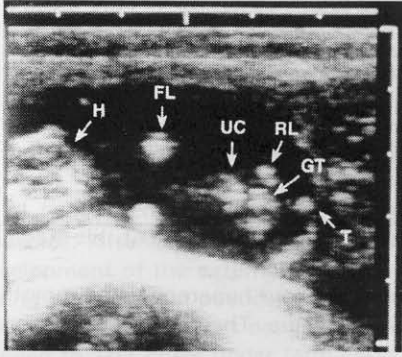


Photo 1: A male fetus in frontal section on day 52

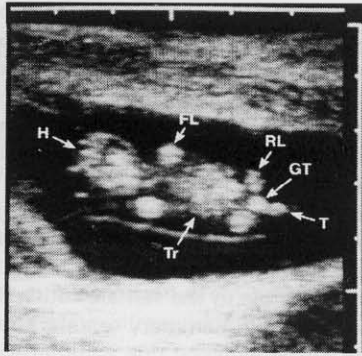


Photo 2: A female fetus in frontal section, on Day 52

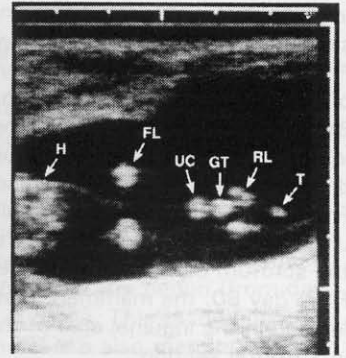


Photo 3: A male fetus in frontal section on Day 55

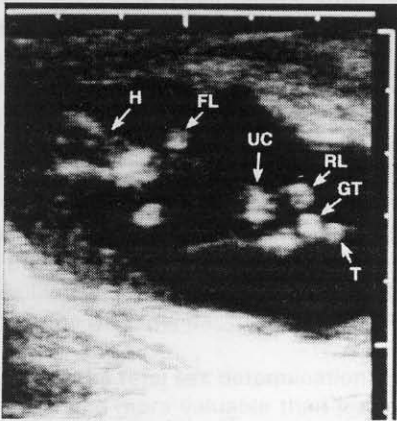


Photo 4: A female fetus in frontal section on Day 55

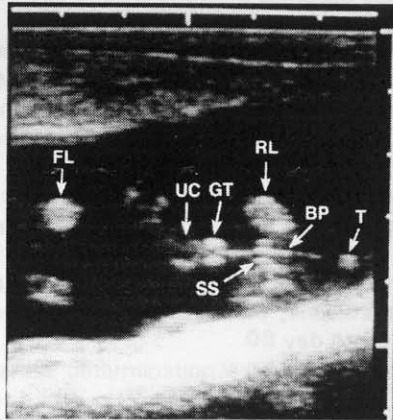


Photo 5: A male fetus in frontal section on Day 61

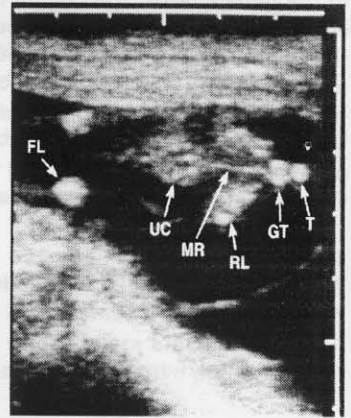


Photo 6: A female fetus in frontal section on Day 63

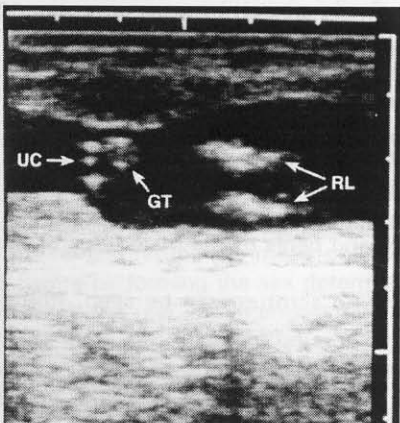


Photo 7: A male fetus in frontal section on Day 70

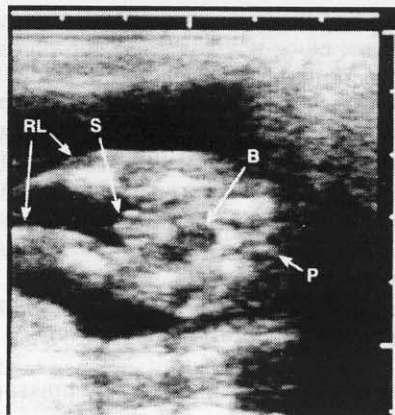


Photo 8: A male fetus in transversal section on Day 72.

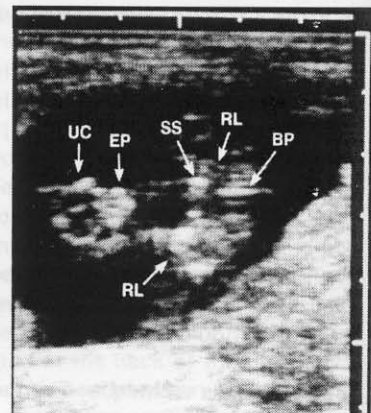


Photo 9: A male fetus in frontal section on Day 78

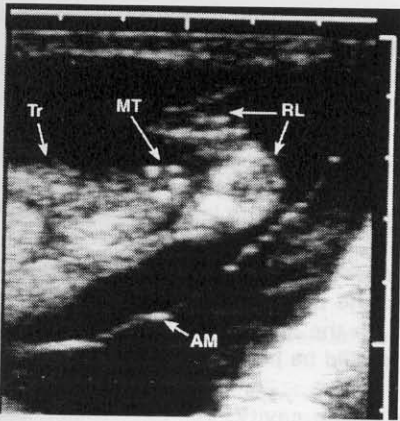


Photo 10: A female fetus in frontal section on Day 78

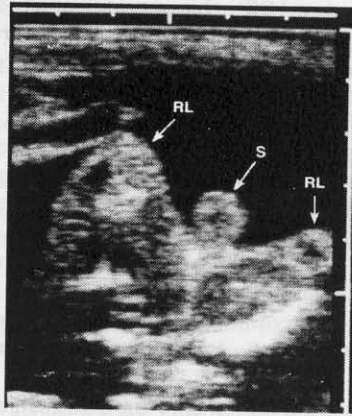


Photo 11: A male fetus in transversal section on Day 100

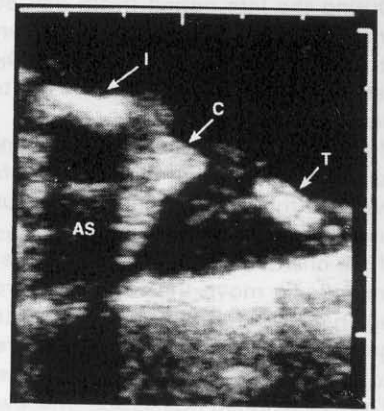


Photo 12: A female fetus in frontal section on Day 120

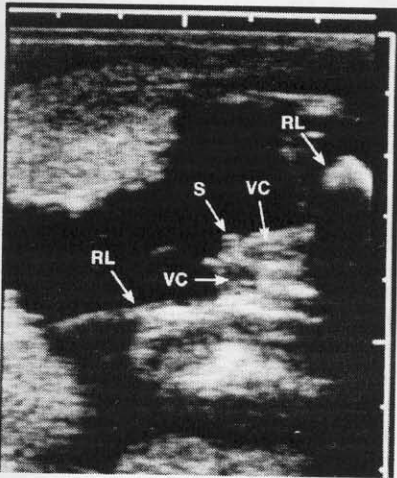


Photo 13: A male fetus in transversal Section on Day 107

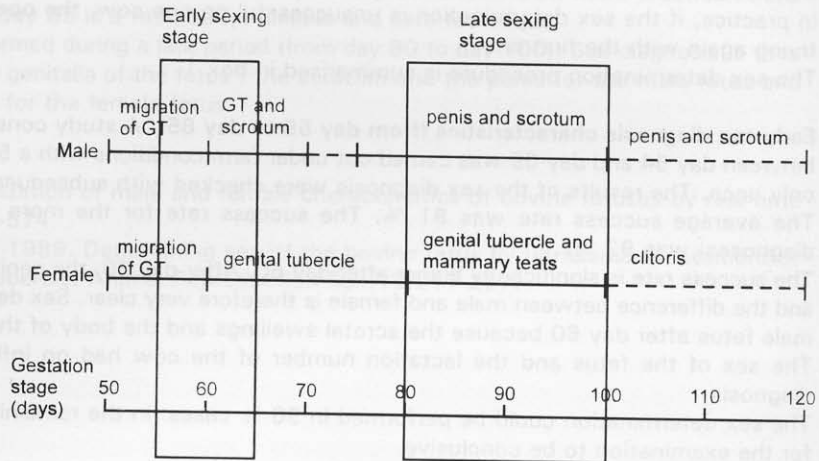


Fig. 1: The two main sex determination periods

to get a picture of the fetus.

The main parts of the fetus (head, trunk, limbs, etc,...) shall be first identified in order to know the orientation of the fetus. Once the orientation of the fetus is known, the probe can be easily moved to get a favourable section for the gender determination.

During the early period (from day 55 to day 65), a frontal section is the best section for gender determination. The genital tubercle and the surrounding structure (rear limbs, umbilical cord, tail, scrotal swellings, etc,...) can be seen at the same time on such a section. Wrong diagnostics are therefore very unlikely. A frontal section is however difficult to obtain.

If a frontal section cannot be found, the sex determination will have to be performed with a transversal section. Transversal sections are easier to obtain : first, the head of the fetus must be identified (which is usually a very easy task). The probe shall then be caudally moved from the head towards the umbilical cord. The operator shall then check for the presence of a male genital tubercle behind the umbilical cord. If the genital tubercle cannot be seen, the operator shall not immediately conclude that the fetus is female. This conclusion should only be reached if the female genital tubercle can be identified at the base of the tail.

During the late period (from day 80 to day 100), the operator shall look for the presence of the scrotum on a transversal section or for the presence of the mammary teats on a frontal section.

It may be useful to slightly push the probe against the uterus, so as to bring the probe closer to the fetus. The fetus will then be displayed at the top of the screen which usually has a higher accuracy than the bottom of the screen.

If the fetus is deep inside the abdominal cavity, the probe shall be moved round the uterus with a lateral movement as an attempt to get closer to the fetus. When moving the probe laterally, the empty uterine horn may be in the way. As the empty uterine horn is usually on the right, it is advised to move the probe to the left-hand side of the uterus.

If the orientation of the fetus makes the examination of the extern genitalia impossible, the fetus should be slightly moved. To move the fetus, the probe shall be placed laterally to the uterus and pushed gently and repeatedly against the uterus. The movements will be communicated to the fetus through the amniotic fluid. As the fetus gently moves, it usually takes a different orientation. Warning : this operation should be performed with great care to prevent any damage to the fetus.

The pressure of the probe will eventually make the fetus move inside the pelvic cavity. For this reason, the examination should not last more than 10 minutes.

If the sex could not be determined within 5 to 10 minutes, the examination should be interrupted for about 10 minutes. The fetus will meanwhile return naturally to its initial position, near the rectum. This position is usually a much better position for sex determination.

In practice, if the sex determination is unsuccessful on one cow, the operator will examine another cow before trying again with the first one.

The sex determination procedure is summarised in box 1.

Early sex diagnosis characteristics (from day 55 to day 65): A study consisting of 107 sex diagnoses performed between day 54 and day 69 was carried out under farm conditions with a 5 MHz scanner. Each cow was examined only once. The results of the sex diagnosis were checked with subsequent calving.

The average success rate was 81 %. The success rate for the more certain cases (about two third of the diagnoses) was 92 %.

The success rate is significantly higher after day 60. After day 60, the genital tubercle has reached its final position and the difference between male and female is therefore very clear. Sex determination is particularly easier for the male fetus after day 60 because the scrotal swellings and the body of the penis can be seen.

The sex of the fetus and the lactation number of the cow had no influence on the success rate of the sex diagnosis.

The sex determination could be performed in 96 % cases. In the remaining 4 %, the fetus was too inaccessible for the examination to be conclusive.

Abortion occurred for 2 % of the cows. This rate corresponds to the usual abortion rate of 2 or 3 % in dairy cattle. The sex diagnosis is therefore a safe technique which does not endanger the pregnancy of the cow.

The origins of most common mistakes were also identified during this study. In the male fetus, images produced by the tail can be mistaken for the female genital tubercle. In the female fetus, it is the umbilical cord – and in particular the umbilical arteries – which can be mistaken for the male tubercle.

An incorrect determination of the orientation of the fetus was also a common source of errors.

Early sex diagnosis characteristics are summarised in box 2.

- The rectum shall be emptied without applying any pressure on the uterus.
- The uterine horns should not be held in one hand.
- The probe shall be pressed against the uterus gently.
- The sex diagnosis should normally take between 5 and 10 minutes.
- If the fetus is difficult to reach, the probe shall be moved round the uterus with a lateral movement. (As the empty horn is usually on the right, the probe shall be first moved to the left-hand side.)
- If the sex diagnosis is unsuccessful, it should not be reattempted for at least 10 minutes.

Box 1 : Sex diagnosis tips.

- Success rate of 81% (success rate of 92% for the more certain case- i.e. for about two third of diagnoses)
- Sex diagnosis was impossible in 4% of the cases.
- Early sex diagnosis does not endanger the fetus.
- Most common errors: tail mistaken for a female genital tubercle, umbilical cord mistaken for the male genital tubercle, incorrect determination of the orientation of the fetus.
- Success rate is higher after day 60 of pregnancy.
- The sex of the fetus and the lactation number of the cow had no influence on the success rate of the sex diagnosis.

Box 2 : Characteristics of the early sex diagnosis (from day 55 to day 65).

Conclusion

Fetal sex determination in cattle using ultrasonography can be performed during two different periods of the pregnancy.

During an early period (from day 55 to day 65), sex diagnosis is based on the relative location of the genital tubercle. For the male fetus, the scrotal swellings and the body of the penis can also be seen. During this early period, the success rate is 81 %. (This success rate could certainly be improved with a bit more practice.) Fetal sex determination between day 55 and day 65 is a fast, early, reliable and safe technique.

Fetal sex determination can also be performed during a late period (from day 80 to day 100). Sex diagnosis is then based on the visualisation of the external genitalia of the fetus : the scrotum and the penis for the male fetus and the mammary teats and genital tubercle for the female fetus.

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