Successful non-surgical deep intrauterine insemination with small numbers of spermatozoa in sows

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A 100-fold reduction of the standard dose for artificial insemination in pigs (3 x 10^9 spermatozoa in 80–100 ml fluid) can be used when spermatozoa are deposited surgically next to the uterotubal junction. The present study was performed to develop a technique for non-surgical deep intrauterine insemination in pigs without sedation of the animal. In Expt 1, sows were weaned, treated to induce oestrus and used to evaluate the difficulties involved in the insertion of a flexible fibre optic endoscope through the cervix and along the uterine horn. Deep uterine catheterizations were performed on each sow at 30–40 h after hCG treatment in the crate in which the animal was housed. The endoscope was inserted through an artificial insemination spirette, moved through the cervical canal and propelled forward along one uterine horn until the entire endoscope was inserted. In 30 sows (90.9%) no or minor difficulties were observed during insertion and in these animals the procedure was completed in 4.1 ± 0.26 min. Insertion of the endoscope through the cervical canal was not possible in only one sow (3.03%). In Expt 2, endoscopic deep intrauterine insemination at 36 h after hCG treatment was performed in 15, 18 and 13 sows with 100, 20 or 5 x 10^7 spermatozoa, respectively, resulting in farrowing rates of 86.6%, 88.9% and 92.3%, respectively; there were no significant differences among groups. Farrowing rates after deep intrauterine inseminations were also not different from those achieved after standard intracervical insemination with 3 x 10^9 spermatozoa (control group: n = 48; 87.5%). Mean litter size (9.41 ± 0.38 to 10.02 ± 0.25) was also similar among the different experimental and control groups. In conclusion, endoscopic non-surgical deep intrauterine inseminations can be performed quickly in sows, and normal farrowing rates and litter sizes can be obtained after insemination with a small number of spermatozoa.

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complex anatomy of the genital tract in sows, mainly the cervical canal and the length and coiled nature of the uterine horns, has impeded the development of a procedure for non-surgical insemination into the uterine horns. The present study was conducted to develop a fibre optic endoscope technique for non-surgical deep intrauterine insemination in pigs, without sedation of the animal.

Materials and Methods

Animals

Experiments were conducted under field conditions in a commercial pig farm in Murcia (Spain). For a period of 10 weeks, crossbred sows (n = 8 – 10) were selected each week on the day of weaning. Sows were allocated individually to crates in a mechanically ventilated confinement facility and were fed with a commercial ration twice a day. Water was provided ad libitum during 2 h after feeding.

Induction and detection of oestrus

Oestrus was induced by an i.m. injection of 1250 iu equine chorionic gonadotrophin (eCG) (Folligon; Intervet International BV, Boxmeer) at 24 h after weaning, followed 72 h later with 750 iu human chorionic gonadotrophin (hCG) (Chorulon; Intervet International BV). Oestrus detection was performed twice a day (07:00 h and 17:00 h), beginning 2 days after injection of eCG, by allowing females nose to nose contact with a mature boar and by applying back pressure. Sows that showed a standing oestrous reflex were considered to be in oestrus and were used for the experiments, but only when oestrus was detected within 24 h of hCG injection.

Experiment 1

Deep uterine catheterization. Thirty-eight sows (2–6 parity) were weaned at 21.72 ± 0.36 days, treated to induce oestrus and used to develop a technique for deep uterine insertion of a flexible fibre optic endoscope (Karl Storz, Tuttlingen).

The flexible fibre optic endoscope (length 1.35 m, outer diameter 3.3 mm and single inner instrument channel of 1.2 mm in diameter) was used in combination with a cold light source (Karl Storz) and a video camera (Karl Storz). A video recorder was connected to the video camera and a television monitor. Between insertions the flexible fibre optic endoscope was cleaned externally and internally with sterile physiological saline at 35°C. After the last insertion of each experiment day, the flexible fibre optic endoscope was soaked in an antiseptic solution, as recommended by the manufacturers, for 30 min, and the instrument channel was flushed with 20 ml of the same solution. The flexible fibre optic endoscope and its instrument channel were rinsed repeatedly with redistilled and purified water. Physiological saline was deposited by gravity through the instrument channel of the endoscope during the insertions to visualize the different parts of the genital tract (cervical canal, uterine body and uterine horn). Images were recorded and studied in detail.

Deep uterine catheterizations were performed on each sow at 30–40 h after hCG treatment in the crate in which the animal was housed; the procedure was performed without sedation. Before insertion, the spiral end of a commercial AI spirette (Minitub, Tiefenbach) was lubricated with a non-spermicidal sterile lubricating jelly, and a silicon spray (Willy Rusch Ag, Kernen) was used to lubricate the flexible fibre optic endoscope externally. After thorough cleansing of the perineal area of the sows, the spirette was inserted through the vagina into the cervix to produce a cervical lock, which was used to manipulate the flexible fibre optic endoscope. The flexible fibre optic endoscope was inserted through the spirette, moved through the cervical canal and propelled forward along one uterine horn until the entire endoscope had been inserted. The difficulties encountered during insertion of the endoscope in relation to the resistance felt by the inseminator were scored subjectively on a scale of 1 to 3 (1 = no or minor difficulties, almost no resistance; 2 = medium or high difficulties, medium or severe resistance; 3 = impossible, maximum resistance). The duration of the procedure was recorded in each case. In addition, all sows were observed for oestrus at days 18–24 after uterine catheterization.

Laparoscopy evaluations. Three sows (2–3 parity) were examined by laparoscopy at 32–34 h after hCG treatment to check progression of the flexible fibre optic endoscope into the uterine horn. On the day of hCG injection, sows were moved from the commercial farm to the Veterinary Hospital of the University of Murcia. The sows were sedated with an i.m. injection of azaperone (3 mg kg−1) (Stresnil; Lab Esteve Veterinaria, Barcelona). General anaesthesia was induced intravenously by a combination of xylazine (1 mg kg−1) (Rompum; Bayer, Barcelona) and ketamine (3 mg kg−1) (Imalgene 1000; Lab Rhoie Merieux, Barcelona), and was maintained with halothane (0.7–1.5%). Laparoscopies were performed as described by Palombo et al. (1999). Briefly, a 2 cm vertical incision was made infra-umbilically. A Veress needle was inserted into the peritoneal cavity and CO2 was insufflated to a pressure of 12 mm Hg using a Laparoflator (Karl Storz). A 10 mm 0° laparoscope (Karl Storz) was inserted and intra-abdominal placement was verified. When the genital tract was viewed through the laparoscope, the flexible fibre optic endoscope was inserted as mentioned earlier and the progression of the endoscope along one uterine horn was observed.

Experiment 2

Deep uterine insemination. Three sexually mature hybrid boars of proven fertility and with satisfactory semen characteristics in the 10 weeks preceding the experiment were selected as semen donors. A sperm-rich fraction was
collected once a week from the three boars using the gloved hand method and evaluated for sperm concentration with a haemocytometer and for motility and morphology using a light microscope. Immediately after evaluation, semen was diluted to \(200 \times 10^6\) cells ml\(^{-1}\) in Beltsville thawing solution (BTS) diluent (Pursel and Johnson, 1975) at 35°C. The sperm suspensions from the three boars were mixed and kept for a maximum of 24 h at 17°C. Immediately before the inseminations, semen samples were resuspended if necessary in BTS diluent at the same temperature to provide sperm doses of \(100 \times 10^7\), \(20 \times 10^7\) and \(5 \times 10^7\) mixed sperm cells (5 ml\(^{-1}\)) for deep uterine inseminations (experimental groups). Forty-six sows (2–6 parity) were weaned at 21.74 ± 0.32 days, treated to induce oestrus and inseminated once in one uterine horn at 36 h after hCG treatment. The sows were assigned randomly to the experimental groups. Non-hormonally treated sows (n = 48; duration of lactation 21.35 ± 0.30 days; 2–6 parity) that showed onset of oestrus at 4 days after weaning were used for standard artificial insemination (control group). These sows were inseminated twice at 0 and 24 h after the onset of oestrus with traditional insemination doses of spermatozoa (3 × 10⁹ spermatozoa diluted to 100 ml in BTS diluent) prepared from the same semen samples that were used for the experimental groups.

Insertions of the flexible fibre optic endoscope were performed as described earlier, but without visualization of the genital tract. Therefore, introduction of saline was not necessary. Before inseminations, the instrument channel of the flexible fibre optic endoscope was rinsed with BTS diluent and refilled with approximately 2 ml BTS extender at 30°C. Predetermined insemination doses of spermatozoa in a volume of 5 ml at 30°C were flushed into one uterine horn using a 5 ml disposable syringe attached to the instrument channel of the flexible fibre optic endoscope. An extra 5 ml BTS diluent alone was used to force all spermatozoa out of flexible fibre optic endoscope. The flexible fibre optic endoscope was cleaned as described earlier, except that between inseminations BTS diluent at 30°C was used instead physiological saline. Pregnancy was diagnosed at days 24–28 after insemination by transcutaneous ultrasonography (Pie Medical, Maastricht). All pregnant animals were allowed to carry litters to term and farrowing rates and litter sizes were recorded.

### Statistical analysis

Difficulties during insertion of the flexible fibre optic endoscope, pregnancy rates and farrowing rates from different groups were compared using the chi-squared test with Yates correction. Duration of the procedure and litter size were analysed by one-way ANOVA using the general lineal model of SYSTAT (Wilkinson and Howe, 1992). When ANOVA revealed a significant effect, values were compared by Tukey’s test. All values are given as mean ± SEM. Differences were considered to be significant at \(P < 0.05\).

### Results

#### Experiment 1

Thirty-eight sows were treated hormonally to induce oestrus. Two sows (5.26%) showed no obvious signs of oestrus at the time of hCG injection and these animals were not used. Thirty-three sows were used for the development of the deep uterine catheterization technique and the other three were subjected to examination by laparoscopy. The difficulties encountered during the insertion of the flexible fibre optic endoscope and the time required to complete the procedure are shown (Table 1). Insertion of the endoscope through the cervix into one uterine horn was possible in about 97% of the sows. The behaviour of the sows during the procedure was good (no reaction: 84.85%) or moderate (minimum reaction: 15.15%) in all cases, and was considered to be similar to the reaction of sows during standard AI.

The entry of the cervix appeared closed in all animals (Fig. 1a) but was easily dilated with the tip of the flexible fibre optic endoscope (Fig. 1b). A gentle and steady pressure was necessary to move the endoscope through the cervical folds. The endoscope progressed easily until the two last cervical folds were reached, and this stage was consistently the most difficult and time-consuming part of the procedure. Slight bleeding into the cervical canal was observed in three sows (one from the group with no or minor difficulties during insertion and two from the group with medium–high difficulties). Upon entering the uterine body, the endometrial folds were apparent (Fig. 1c,d) and occluded visualization of the bifurcation between the uterine horns. Septum uterus was not seen in any of the animals. One uterine horn could be visualized (Fig. 1e) until approximately the second curvature of the uterus, at which point the tip of the flexible fibre optic endoscope was adjacent to the uterine wall and vision was occluded. However, the endoscope progressed without difficulty along the uterine horn until the entire endoscope was inserted. During removal of the endoscope from the genital tract, a visible mark on the endometrium of the first uterine curvature was observed in seven animals. The cervical

### Table 1. Results from scoring the difficulties encountered during insertion of an endoscope through the cervical canal and into one uterine horn in sows

<table>
<thead>
<tr>
<th>Difficulty (score)</th>
<th>Number of sows (%)</th>
<th>Time (min) (mean ± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>30 (90.9)a</td>
<td>4.1 ± 0.26a</td>
</tr>
<tr>
<td>2</td>
<td>2 (6.06)b</td>
<td>11.0 ± 4.00b</td>
</tr>
<tr>
<td>3</td>
<td>1 (3.03)b</td>
<td>–</td>
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Difficulties were scored subjectively on a scale of 1 to 3: 1 = no or minor difficulties; 2 = medium–high difficulties; 3 = impossible.

abValues within columns with different superscripts are significantly different \((P < 0.001)\).
Fig. 1. Endoscopic images of different parts of the genital tract during oestrus in sows. The insertions of the endoscope were performed in sows housed in gestation crates, with no sedation. (a) Artificial insemination spirette (blue colour) inserted conventionally into the cervix. The continuation of the cervical canal can be seen. (b) Cervical canal with cervical folds. Normally the canal is curved and has a smooth white mucosal membrane that is distinctly different from the border of the uterine body. (c) Passage from the cervix into the uterine body. A proliferation of endometrial folds was observed in all cases. (d) Lumen of the uterine body showing detail of the uterine folds. (e) Lumen of uterine horn. (f) Appearance of the cervical canal during withdrawal of the endoscope. The cervical canal is circular, notably dilated and the cervical folds are not visible. The cervical canal closed suddenly when the tip of the endoscope was removed from the canal, adopting a configuration like that shown in (a).
canal was very dilated with a diameter similar to the uterine body (Fig. 1f); however, the canal closed as the flexible fibre optic endoscope was being removed. No complications or infections were observed after the procedure in any of the sows, and all sows returned to oestrus at 21.5 ± 0.24 days.

Laparoscopic observations are shown (Fig. 2). The progression of the flexible fibre optic endoscope along the uterine horn was clearly visible in the three sows. The tip of the flexible fibre optic endoscope reached approximately the middle or first portions of the anterior third of the uterine horn. In all cases, the uterine horn adapted to the flexible fibre optic endoscope and formed a spiral shape.

**Experiment 2**

Of the 94 sows used, deep insemination was performed in one uterine horn of 46 sows using the flexible fibre optic endoscope. Pregnancy and farrowing rates are shown (Fig. 3).

High farrowing rates were achieved by endoscopic deep uterine insemination of 100, 20 or 5 × 10⁷ spermatozoa (86.6%, 88.9% and 92.3%, respectively), and the mean values among the groups were not significantly different. Farrowing rates after deep uterine insemination were not different from those achieved after standard intracervical insemination with 3 × 10⁹ spermatozoa (control group: 87.5%). Mean litter size was similar among the different deep uterine insemination and control groups (9.61 ± 0.29, 9.75 ± 0.31, 9.41 ± 0.38 and 10.02 ± 0.25 for deep uterine insemination with 100, 20 and 5 × 10⁷ spermatozoa and the control group, respectively; Fig. 3).

**Discussion**

Non-surgical transcervical catheterization of the uterus has been used in cows, horses and dogs for diagnosis and treatment of reproductive disorders (Devine and Lindsay, 1984; Bracher et al., 1992; Watts and Wright, 1995). However, there have been no reports of transcervical catheterization in sows. The major obstacle to this procedure is the anatomy of the cervix and uterus in pigs. The cervical folds and the length and coiled nature of the uterine horns have discouraged attempts at non-surgical transcervical introduction of a catheter into the uterine horn. The present study is the first to report a technique for non-surgical insertion of an instrument deep into the uterine horn of a sow without sedation of the animal.

In Expt 1, a procedure was developed to gain access to the uterine horn through the cervix using a flexible fibre optic endoscope. This procedure can be performed without any particular technical difficulties in sows because the flexible fibre optic endoscope has the necessary propulsion force to pass through the cervix and the required flexibility to progress along the uterine horn. It is not necessary to have an optic system to insert the instrument through the cervix and into one uterine horn.
With this technique, it was possible to pass the cervical canal and to reach one uterine horn in 3–7 min in about 90% of the sows. Insertion of the flexible fibre optic endoscope through the cervix was not possible in only one sow (3.03%). The major difficulty in inserting the flexible fibre optic endoscope into the uterine horn was encountered at the two last cervical folds, which often appeared tightly closed; however, with gentle pressure from the endoscope the folds could be dilated in most of the sows. The cervical distensibility differed among animals, but neither parity number nor duration of lactation (data not shown) was implicated. Individual variation could be the main reason for the differences seen in the dilation of the cervical canal and the passage of the endoscope. Whether such individual variation depends on breed has not been evaluated.

At the conclusion of the procedure, as the flexible fibre optic endoscope was being withdrawn, the cervical canal presented a tubular configuration, the cervical folds were not visible and maximum dilation was observed in all sows. Different hormones may have been liberated as a result of the passage of the endoscope through the cervix and along the uterine horn, which may have influenced the dilation of the cervical canal. Oxytocin concentrations may be influenced by events that occur near the time of mating, including stimulation of the genital tract and uterine distension in mares (Nikolakopoulus et al., 2000), cows (Schams et al., 1982) and sows (see Soede, 1993). Sayre and Lewis (1996) indicated that exogenous oxytocin induces cervical dilation in oestrous ewes within approximately 10 min and for a period of up to 6 h, and permits transcervical entry into the uterus. Uterine contractions, enzymatic processes, collagenolysis and release of prostaglandins are some of the possible mechanisms of action of oxytocin for inducing cervical dilation discussed by these authors. However, in the present study, cervical dilation was too rapid, approximately 3–4 min after insertion of the flexible fibre optic endoscope, and disappeared too quickly, during removal of the endoscope, to have been caused by an increase in oxytocin in response to the cervical–uterine stimuli. Additional work is required to determine the factors implicated in this type of cervical dilation.

During insertion of the endoscope, no chemical restraint of the female was required because the sows showed no clinical reactions, indicating that this procedure is a relatively non-traumatic method that is well tolerated by sows, as is also the case in cows (Devine and Lindsay, 1984). All sows used in this experiment showed no symptoms of uterine infection in the days after hysteroscopy and they returned to oestrus after a normal period. This finding indicates that the risk of inducing uterine infection during the procedure is very low, probably because sows are resistant to infectious endometritis when under the influence of oestrogen during oestrus (De Winter et al., 1994, 1996).

Although a visible mark on the endometrium wall was observed in some animals during removal of the flexible fibre optic endoscope from the genital tract, the results of the present study indicate that the endoscopic procedure for non-surgical deep uterine insemination does not have a detrimental effect on the fertility of hormonally treated oestrous sows. Farrowing rates and litter sizes obtained after non-surgical deep uterine insemination with small numbers of spermatozoa (a minimum of $5 \times 10^7$ spermatozoa per sow) were not different from those in sows inseminated with traditional doses of spermatozoa ($3 \times 10^9$ spermatozoa per insemination, two inseminations per sow).

During the last 50 years, use of a large quantity of liquid (50–200 ml; Wiggins et al., 1951; Polge, 1956; Dziuk and Henshaw, 1958) and a large number of spermatozoa ($5–10 \times 10^9$ spermatozoa per insemination dose; Stratman and Self, 1960; Baker et al., 1968) deposited intracervically during AI have been recommended to achieve maximum fertility in pigs. Although considerable advances have been made in AI technology in pigs (including new diluents, new methods to evaluate quality and functional sperm characteristics, procedures to detect the timing of ovulation), few modifications have been made in recommendations on the number of spermatozoa and the dose volume per insemination. However, new methods of insemination with a small number of spermatozoa are required to exploit the availability of new technologies, such as sperm sorting in which the number of viable spermatozoa is reduced (Johnson, 1997). It is now known that the number of spermatozoa per insemination can be reduced markedly when the spermatozoa are deposited into the uterine horn. In cattle, insemination with small numbers of spermatozoa (1.0–2.5 $\times 10^5$ total sperm cells per inseminate) into the uterine horn can result in acceptable pregnancy rates compared with those in controls ($2.5 \times 10^6$ spermatozoa per dose) (Seidel et al.; 1997). In mares, a 500-fold reduction in the number of spermatozoa can be made using hysteroscopic-guided insemination of Percoll-treated spermatozoa onto the uterotubal junction before ovulation (Morris et al., 2000). These authors obtained a high incidence of fertilization and embryonic development after insemination with only $1 \times 10^6$ spermatozoa. Successful surgical intrauterine insemination with a small number of spermatozoa has been reported for hormonally stimulated prepubertal gilts (Krueger et al., 1999) and for sows (Krueger and Rath, 2000). These investigations have demonstrated that the number of spermatozoa used for surgical intrauterine insemination (next to the uterotubal junction) can be reduced to $1 \times 10^7$ spermatozoa without compromising fertility. The results from Expt 2 in the present study indicate that when spermatozoa are deposited non-surgically deep in one uterine horn, normal farrowing rates and litter sizes can be obtained by inseminating only $5 \times 10^7$ spermatozoa per sow. Experiments are being conducted to determine the minimum number of spermatozoa required to maintain optimal fertility using this new technology. In the present experiment, sows were not inseminated with small numbers of spermatozoa using the standard AI procedure because it is reported widely that $3 \times 10^9$ spermatozoa in a volume of...
70–100 ml are needed to achieve optimal fertility when the semen is deposited into the cervical canal. Therefore, it might be assumed that traditional AI with 5–15 × 10⁷ spermatozoa in a total volume of 10 ml would give poor fertility results.

The reasons why the number of spermatozoa per insemination dose can be decreased when the insemination is performed deep in the uterine horn are not clear. Although contractions of the myometrium are vigorous during oestrus, and these should assist transport and redistribution of the semen between the two uterine horns, an initial distribution of semen in the uterus may be achieved as a result of the force of ejaculation and the volume of fluid involved (see Hunter, 1982). Thus, the high volume of semen deposited during natural mating may favor displacement of a portion of the ejaculate to the region of the uterotubal junction, which is bathed in a sperm suspension by the completion of mating (see Hunter, 1982). Spermatozoa from this portion could enter the oviducts and establish the sperm reservoir soon after mating. Similar events may occur when the animals are inseminated artificially using a dose volume of 80–100 ml. The requirement for a large volume at insemination is supported by the finding that either spermatozoa in the oviducts or accessory spermatozoa were found in gilts inseminated intracervically with only 20 ml of semen containing 1 × 10⁹ spermatozoa (Stratman and Self, 1960; Baker et al., 1968). However, in the present study, high fertility rates were obtained after non-surgical deep intrauterine inseminations with a dose volume of only 10 ml even when the number of spermatozoa was decreased to 5 × 10⁷.

Therefore, deposition of a small number of spermatozoa in a small volume of liquid close to the uterotubal junction may be similar to an aliquot of the ejaculate or of a standard AI dose arriving at the uterotubal junction during service or AI, respectively, as has been suggested in horses (Morris et al., 2000). This is in agreement with the suggestion of Hunter (1982) that, from the point of view of the future development of AI, it would be useful to develop a technique that permitted deposition of a few millilitres of semen at the top of each uterine horn against the uterotubal junction. However, the results from the present study indicate that it is not necessary to perform insemination in the exact region of the uterotubal junction to obtain adequate farrowing rates and litter size when using a small number of spermatozoa in a small volume of medium. In the present study, the semen was deposited in the middle or the first portions of the anterior third of one uterine horn, as determined by laparoscopy. Furthermore, only one insemination in one uterine horn was performed because it was not possible to visualize the uterine bifurcation owing to the endometrial characteristics of the uterine body during oestrus. These conditions were sufficient to enable formation of a sperm reservoir in the caudal isthmus of at least one oviduct, because no significant differences were observed in farrowing rates compared with those obtained after standard AI (3 × 10⁹ spermatozoa in 80 ml liquid, two inseminations per sow). Whether the number of spermatozoa inseminated can be reduced below 5 × 10⁷ by non-surgical insemination closer to the uterotubal junction requires investigation. Litter size in each group was also not statistically different from the control group. Although not evaluated in the present study, it is possible that the hormonal treatment used in the sows that underwent deep uterine insemination increased the number of ovulations in each ovary and that a greater number of oocytes could be fertilized in the oviduct ipsilateral to the uterine horn inseminated. This theory could explain the large number of piglets born under the experimental conditions in the present study if fertilization occurred in only one oviduct.

Although the effectiveness of deep uterine insemination with small numbers of spermatozoa, in terms of farrowing rate and litter size, might be considered excellent, it is necessary to take into account the fact that the sows used for deep uterine insemination in the present experiment were treated hormonally for induction of oestrus. In a situation in which the timing of ovulation is not controlled precisely with hormones, the requirements in relation to the number of spermatozoa and number of deep inseminations per oestrus may be different.

In conclusion, endoscopic non-surgical deep uterine insemination can be performed quickly in sows. Normal farrowing rates and litter sizes can be obtained by inseminating a small number of spermatozoa (5–20 × 10⁷) in a small volume of fluid (10 ml). Although endoscopic deep uterine insemination is an attractive technique for insemination of a small number of spermatozoa in sows, the flexible fibre optic endoscope is an expensive and fragile instrument to be used under field conditions. It will be necessary to develop new devices to exploit the potential use of non-surgical deep uterine insemination. As it is not necessary to have an optic system to pass through the cervix or to gain entry to the uterine horn, the new devices could be made on the basis of the propulsion force and flexibility of the flexible fibre optic endoscope used in this work. Preliminary results using a new catheter for non-surgical deep uterine insemination in sows have been reported (Martinez et al., 2001). Experiments are underway to determine the effectiveness of this new technology under different conditions, including the use of frozen semen, flow-sorted spermatozoa and at natural post-weaning oestrus in sows. Non-surgical deep uterine insemination could have a high economic impact on the use of fresh and frozen semen in the AI industry in pigs and could assist in the adoption of other biotechnologies, such as gender preselection.

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