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EFFECT OF BOAR SEMINAL PLASMA ON UTERINE AND OVIDUCTAL MOTILITY IN OESTROUS GILTS*

By

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VIRING, SVEN and STIG EINARSSON: *Effect of boar seminal plasma on uterine and oviductal motility in oestrous gilts.* Acta vet. scand. 1980, *21*, 607—616. — The effect of boar seminal plasma was tested on the muscular activity of the uterine horns and of the isthmic part of the oviducts in anaesthetized oestrous gilts. The motility was recorded by using balloon-tipped catheters placed in the middle part of the uterine horns and in the isthmus close to the uterotubal junction. The following test solutions were used: sperm-free seminal plasma and isotonic glucose solution. In the isthmic part a test solution called OLEP was also used. OLEP is a buffer solution with electrolyte concentrations, pH and osmotic pressure similar to seminal plasma.

Neither seminal plasma nor the buffers used affected the motility of the uterine horns. Seminal plasma decreased the spontaneous motility of the isthmus, mainly characterized by a reduced amplitude. No effect was, however, obtained with the different buffer solutions used. It is therefore supposed that an unidentified substance with relaxative effect on the isthmic muscle is present in the boar seminal plasma.

boar; seminal plasma; uterus; oviduct; oestrus; gilt.

The semen is introduced directly into the uterus during natural or artificial insemination in the pig. The spermatozoa reach the upper parts of the oviducts shortly after mating/insemination (e.g. *Burger* 1952, *First et al.* 1968, *Baker & Degen* 1972). Radiolabelled substances of different molecular sizes, mixed with seminal plasma, were present in the ampullar part of the oviducts of gilts in measurable amounts within 5 min after intra-uterine deposition (*Viring et al.* 1980). It is therefore quite clear that contractile activity of the genital tract itself must be mainly responsible for the rapid transport.

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Several factors are considered to effect the sperm transport within the genital tract. *Baker et al.* (1968) suggested that some minimum volume of semen must be introduced to achieve optimum fertility, and the sperm concentration must also be maintained above some minimum value.

Also the seminal plasma itself has been ascribed a promoting effect on sperm transport within the genital tract of female pigs (*Leidl* 1968, *Viring & Einarsson* 1980). Based on experimental inseminations by surgical means, *Viring & Einarsson* supposed the seminal plasma effect to be localized mainly to the isthmic part of the oviduct close to the uterotubal junction.

In in vitro experiments using isolated uterine preparations from oestrous gilts (*Einarsson & Viring* 1973) or from rats (*Bower* 1974) as test objects, boar seminal plasma caused an inhibition of the spontaneous motility, with lowered frequency and sometimes with a decrease in amplitude of the contractions. No relaxation of the myometrial activity in the presence of seminal plasma was, however, recorded in unanaesthetized pigs, using chronically implanted extraluminal strain gauge transducers (*Bower*).

The object of the present study was to test, with intraluminal balloons, whether boar seminal plasma affects the motility of the uterus as well as of the isthmic part of the oviduct close to the uterotubal junction.

MATERIALS AND METHODS

Twelve crossbred (Swedish Landrace × Swedish Yorkshire breed) gilts were used in the two experiments presented. The gilts were checked for oestrus daily in the presence of a vasectomized boar. The gilts were used for the experiments approx. 24 h after the onset of their second or third heat.

The gilts were operated upon under general anaesthesia with a 5 % solution of thiopentone sodium (Pentothal, Abbott) injected into a cannulated ear vein. By repeated injections the gilts were kept anaesthetized during the whole experimental period (3—7 h).

Experiment I

Six gilts were used. A midline incision was made in the abdominal wall and the uterine horns were localized and exposed. Through a small incision in the middle of each uterine horn a

polyethylene tube (PE 90—0.86 mm i.d. × 1.27 mm o.d. Clay-Adams, Division of Becton and Dickinson and Company, N. J., USA), with a balloon (18/55/36 or 18/18/16, London Rubber Industry LTD, England) in the end was inserted. The polyethylene tube was fixed to the uterine wall, using 3—0 silk thread. The tube was connected to a pressure transducer. Earlier the recording system had been carefully filled with saline and the balloon sealed in the end. The uterine motility was recorded with a polygraph (Model 79C) using 7PL DC preamplifiers and Statham p-23 AC pressure transducers (Grass medical instruments, Mass., USA). After checking that the recording system was functioning, the midventral incision was closed.

After recording of the spontaneous motility for at least 1 h, a rubber catheter (*Melrose & O'Hagan* 1961) was inserted into the cervix by way of the vagina for insemination of test solutions.

The following test solutions were used:

1. Sperm-free boar seminal plasma
2. 5.5 % isotonic glucose solution

On each occasion 35 ml test solution was inseminated.

Experiment II

Six gilts were used in this experiment. The genital tract was exposed through a midline incision in the abdominal wall. A pressure recording system was established (Fig. 1). One recording catheter was inserted into each oviduct through a small longitudinal incision in the oviductal wall about 4 cm from the uterotubal junction. The polyethylene tube was of the same size as described in Experiment I. In the end of the tube a microballoon was fixed. The balloons, made of silicone tube (0.787 mm i.d. × 1.118 mm o.d., MDX-4-4515 HHI 072, Extracorporeal Medical Specialities, Pennsylvania, USA), were attached directly to the tube with a double 4—0 silk suture. The recording system was carefully filled with saline and the microballoons sealed in the end. The catheters were fixed to the oviduct wall, using 3—0 silk suture after placing the free end of the balloon about 5 mm from the uterotubal junction. The recording equipment was the same as in Experiment I (Fig. 1).

The uterine horns were ligated with catgut (No. 2) 20 cm from the uterotubal junction. The spontaneous uterine motility was identical before and after ligation (unpublished observation).

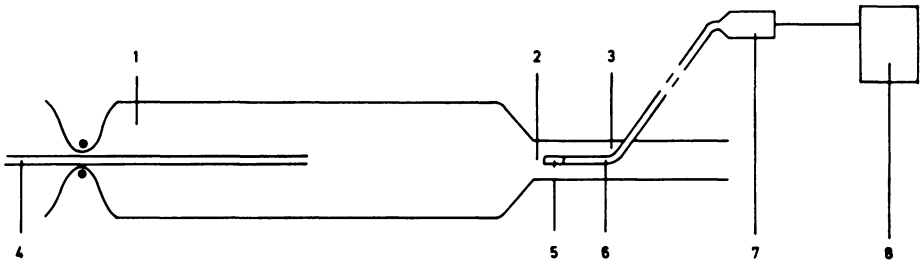

Figure 1. A drawing demonstrating recording equipment and positions of the tubes and balloon within the genital tract in Experiment II.

1. Uterine horn
2. Uterotubal junction
3. Isthmic part of the oviduct
4. Infusion tube
5. Balloon (sealed silicone tube)
6. Recording catheter
7. Pressure transducer
8. Polygraph

Polyethylene tubes, of the same size as described above, were inserted into the ligated part of the uterine horns through small incisions in the uterine wall. The catheters were fixed to the uterine wall. All the catheters were passed out of the midventral incision. After checking that the recording system was functioning, the midventral incision was closed.

The spontaneous motility of the isthmus was always recorded for at least ½ h before administration of test solutions. In four gilts 10—20 ml boar seminal plasma was slowly infused into one of the ligated uterine horns through the inserted polyethylene tube and 10—20 ml 5.5 % isotonic glucose solution into the other ligated horn. In two gilts 10—20 ml OLEP (*Larsson & Einarsson* 1976) was infused into the ligated part of each uterine horn. The motility of the isthmus was recorded for at least 1 h after administration of test solutions.

OLEP is a buffer solution with electrolyte concentrations, pH and osmotic pressure similar to seminal plasma.

RESULTS

Experiment I

The spontaneous uterine motility is presented in Figs. 2, 3. The frequency of concentrations was 6—10 per 10 min and the duration of the concentrations 0.5—1 min. The intra-luminal pressure of the contractions ranged between 30 and 45 mm Hg.

All six gilts were inseminated with seminal plasma. In none

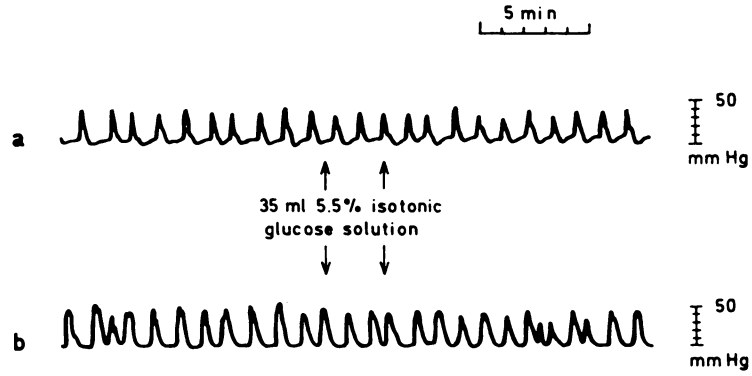

Figure 2. The uterine motility before and after insemination of isotonic glucose solution into the uterine horns: (a) right horn, (b) left horn.

of them did the contraction pattern change (Fig. 3). In three gilts 5.5 % isotonic glucose solution was inseminated. No change of the spontaneous contraction pattern was recorded (Fig. 2).

Experiment II

Preliminary results from this experiment were presented earlier by *Viring et al.* (1976). The spontaneous motility of the recorded part of the oviducts was as a rule regular and similar on both sides in all four gilts. Besides heavy contractions with a frequency of about one per min, small frequent peaks were recorded (Figs. 4—6).

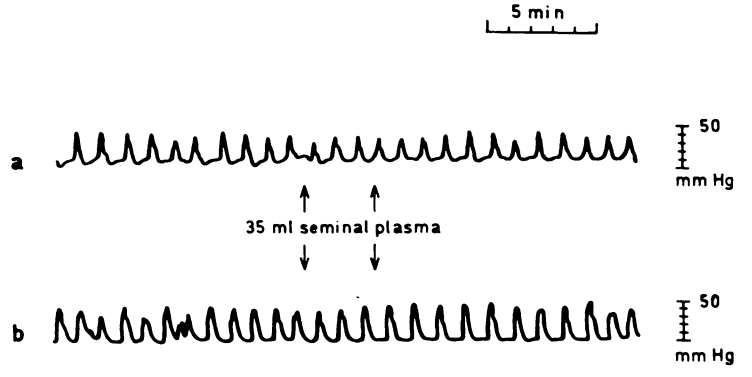

Figure 3. The uterine motility before and after insemination of seminal plasma into the uterine horns: (a) right horn, (b) left horn.

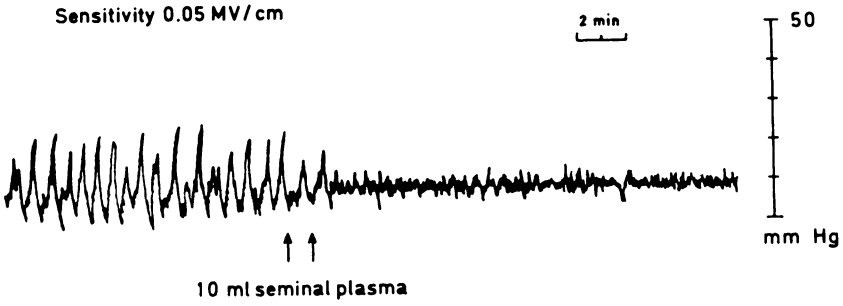

Figure 4. The motility of the isthmic part of the oviduct close to the uterotubal junction before and after insemination of seminal plasma into the ligated top part of the uterine horn.

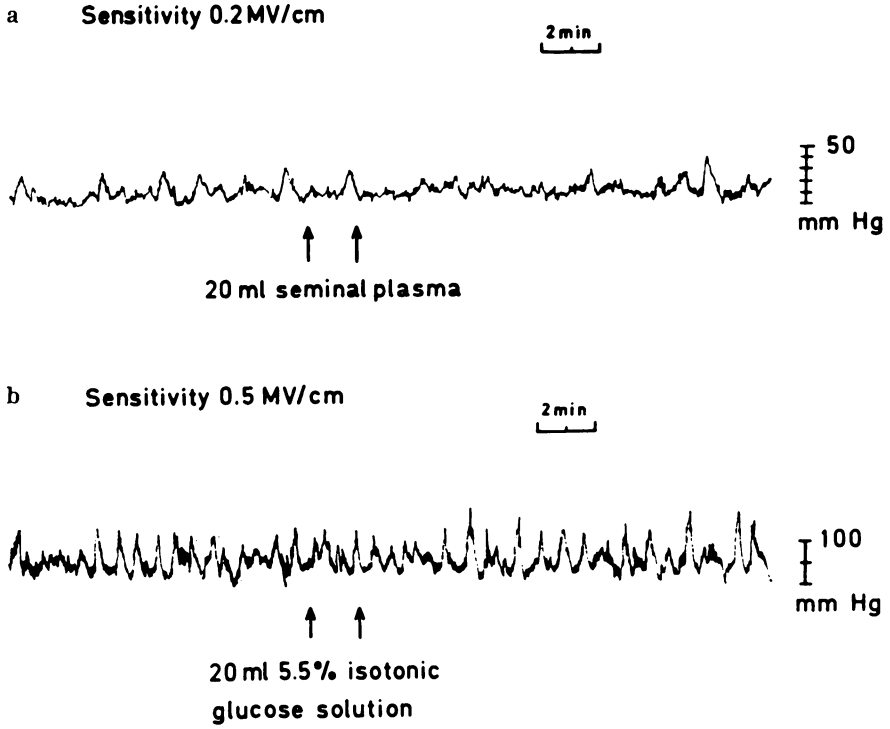

Figure 5. The motility of the isthmic parts of the oviduct in one gilt before and after insemination into the ligated part of one uterine horn close to the uterotubal junction, a) 20 ml seminal plasma, b) 20 ml 5.5 % isotonic glucose solution.

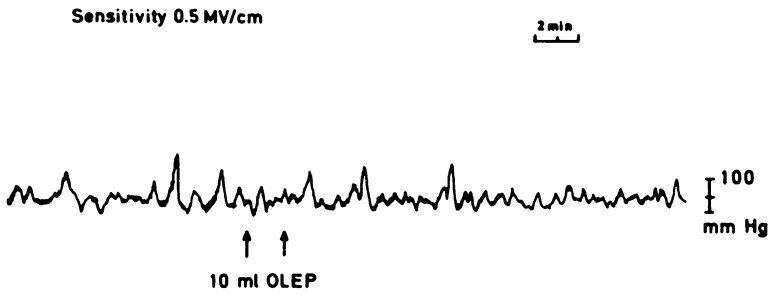

Figure 6. The motility of the isthmic part of the oviduct close to the uterotubal junction before and after insemination of 20 ml OLEP into the ligated part of the uterine apex.

A decreased contractility was demonstrated after introduction of seminal plasma in all four gilts tested (Figs. 4, 5). The changed motility was recorded almost immediately or within 5—10 min and with a duration of at least 30 min. The change of motility was mainly characterized by a reduced amplitude. The intraluminal pressure of the contractions was reduced to 35—80 % of the original in the presence of seminal plasma. No effect was in any case recorded on the control side injected with isotonic glucose solution (Fig. 5). In the two gilts inseminated with OLEP solution no change of the contraction pattern of the isthmus part was recorded (Fig. 6).

DISCUSSION

The spontaneous motility of the uterus and of the isthmic part of the oviduct was as a rule stabilized within 30 min. The uterine contraction pattern was almost identical with that reported by *Zerobin* (1968) and the oviductal motility with that reported by *Zerobin & Spörri* (1972) using anaesthetized female pigs.

Seminal plasma did not significantly affect the spontaneous uterine motility. The inhibition of the uterine contractions obtained in vitro by addition of small quantities of boar seminal plasma into the organ bath, Tyrode solution (*Einarsson & Viring* 1973) was not recorded in any case in the present in vivo study.

The difference in reactivity of the myometrium to seminal plasma between in vitro and in vivo studies is difficult to explain. One explanation might be an insufficient sensitivity of the in

vivo recording system to small changes in the motility in the motility pattern.

Bower (1974), using unanaesthetized, and *Zerobin*, using anaesthetized oestrous female pigs, recorded an increased myometrial activity after insemination of seminal plasma. A similar increased activity was accomplished by insemination of a glucose-$NaHCO_3$ extender (*Bower*). This response may be dependent on the volume of fluid inseminated and explain the discrepancy in relation to the present results.

A decreased contractility of the isthmus part of the oviduct was demonstrated in all cases after introduction of seminal plasma into the ligated part of the uterine horns. In in vitro studies using longitudinally cut strips of the porcine isthmus, boar seminal plasma increased the tonus as well as the amplitude of the contractions in four of six tested animals. In the other two cases no effect or a decrease of the activity was recorded (*Einarsson & Viring*). The discrepancy in results between in vitro and in vivo studies might be due to different sensitivity of isthmic circular muscle and isthmic longitudinal muscle to seminal plasma.

The method used in the in vitro studies (*Einarsson & Viring*) was particularly sensitive to changes in the longitudinal muscle elements of the oviduct, while it was less suitable for detecting changes in oviductal circular muscle. The in vivo method used in our experiments is more likely to detect changes in tubal circular muscle and would only secondarily detect changes in the longitudinal elements of the oviductal muscle system.

Solutions with similar ionic strength, electrolyte concentrations, pH and osmotic pressure as seminal plasma did not change the contraction pattern of the isthmus. The changed pattern of the isthmic motility was therefore not caused by, e.g., the electrolytes in the seminal plasma.

Several studies in different species have been carried out concerning effects of different prostaglandins on the oviductal motility in vitro as well as in vivo. *Brundin* (1965) reported that PGE_1 caused a relaxation of the isthmic circular muscle in the rabbit. Furthermore *Spilman & Harper* (1973) found that several prostaglandins of the E series suppressed spontaneous activity and often completely abolished oviductal muscle activity in rabbits. The effects obtained with PGE on rabbit isthmus in vivo are almost identical with those obtained with seminal plasma on

porcine isthmus in the present study. No detectable prostaglandin PGE_2 was, however, found in boar seminal plasma (*Hunter* 1975, *Poyser & Hunter* 1975 cited, *Poyser* 1980 personal communication). Therefore boar seminal plasma is supposed to contain some other substance of unknown identity with relaxative effect on the isthmic part of the oviduct in pigs.

The constricted lumen of the isthmic part of the porcine oviduct during oestrus together with the oedematous condition of the longitudinal folds are thought to be of functional importance for sperm transport (*Hunter*). Therefore it seems logical to assume that the relaxative effect of seminal plasma on the porcine isthmus (at least close to the uterotubal junction) facilitates sperm transport into the oviducts. Sperm distribution studies have also confirmed this theory (*Viring & Einarsson* 1980). More spermatozoa on an average were namely always recovered from oviducts connected to uterine horns inseminated with spermatozoa suspended into seminal plasma.

REFERENCES

Baker, R. R. & A. A. Degen: Transport of live and dead boar spermatozoa within the reproductive tract of gilts. J. Reprod. Fertil. 1972, *28*, 369—377.

Baker, R. D., P. J. Dziuk & H. W. Norton: Effect of volume of semen, number of sperm and drugs on transport of sperm in artificially inseminated gilts. J. Anim. Sci. 1968, *27*, 88—93.

Bower, R. E.: Factors affecting myometrial activity in the pig. Thesis, Univ. Minnesota 1974.

Brundin, J.: Distribution and function of adrenergic nerves in the rabbit fallopian tube. Acta physiol. scand. 1965, *66*, Suppl. 259.

Burger, J. F.: Sex physiology of pigs — studies on the boar (part 5). Onderstepoort J. vet. Res. 1952, Suppl. 2, 116—131.

Einarsson, S. & S. Viring: Effect of boar seminal plasma on the porcine uterus and the isthmus part of oviducts in vitro. Acta vet. scand. 1973, *14*, 639—641.

First, N. L., R. E. Short, J. B. Peters & F. W. Stratman: Transport and loss of boar spermatozoa in the reproductive tract of the sow. J. Anim. Sci. 1968, *27*, 1037—1040.

Hunter, R. H. F.: Physiological aspects of sperm transport in the domestic pig, sus scrofa. II. Regulation, survival and fate of cells. Brit. vet. J. 1975, *131*, 681—690.

Larsson, K. & S. Einarsson: Fertility of deep frozen boar spermatozoa. Influence of thawing diluents and of boars. Acta vet. scand. 1976, *17*, 43—62.

Leidl, W.: Die akzessorischen Geschlechtsdrüsen der Tiere und ihre Bedeutung für die Fertilität. (The accessory sex glands of animals and their importance for fertility). Proc. 6th World Congr. Fertil. Steril., Tel Aviv 1968, 277—283.

Melrose, D. R. & C. O'Hagan: Investigations into the techniques of insemination in the pig. Proc. 4th Int. Congr. Anim. Reprod., The Hague 1961, IV, 855—859.

Spilman, C. H. & M. J. K. Harper: Effect of prostaglandins on oviduct motility in estrous rabbits. Biol. Reprod. 1973, *9*, 36—45.

Viring, S. & S. Einarsson: Influence of boar seminal plasma on the distribution of spermatozoa in the genital tract of gilts. Acta vet. scand. 1980, *21*, 598—606.

Viring, S., S. Einarsson & J.-O. Lindell: Effect of boar seminal plasma on the contractility of the porcine oviduct in vivo. Proc. 8th Int. Congr. Anim. Reprod., Cracow 1976, 326—328.

Viring, S., S. Einarsson, B. Jones & K. Larsson: Transuterine transport of small- and medium-sized molecules deposited in the uterus in gilts. J. Reprod. Fertil. 1980, *59*, 459—462.

Zerobin, K.: Untersuchungen über die Uterusmotorik des Schweines. (Investigation of the uterine motility in pigs). Zbl. Vet.-Med. 1968, Reihe A, *15*, 740—798.

Zerobin, K. & H. Spörri: Motility of the bovine and porcine uterus and fallopian tube. Advanc. vet. Sci. 1972, *16*, 303—354.

SAMMANFATTNING

Galtspermaplasmas effekt på muskelaktiviteten i livmoder och äggledare hos gyltor i brunst.

Galtspermaplasmas effekt på muskelaktiviteten i livmoderhornen och i äggledarnas isthmusdel undersöktes på brunstiga gyltor under allmännarkos. Kontraktionsmönstret registrerades med hjälp av ballonger som placerats i livmoderhornens mitt och i isthmusdelen nära „uterotubal junction". Ballongerna var via katetrar anslutna till tryckregistreringsapparatur. Spermiefri spermaplasma och isoton glykoslösning användes som testvätskor. Vid tryckregistrering av isthmusdelen användes dessutom en buffertlösning benämnd OLEP, som har ett elektrolytinnehåll, pH och osmotiskt tryck som överensstämmer med spermaplasma. Varken spermaplasman eller elektrolytlösningen påverkade det spontana kontraktionsmönstret i livmoderhornet. Däremot dämpade spermaplasma den spontana aktiviteten i isthmusdelen av äggledaren, huvudsakligen genom en minskning av amplituden. Ingen effekt kunde ses efter tillsats av buffertlösningarna. En oidentifierad komponent med avslappande effekt på isthmusregionens muskulatur antas finnas i galtspermaplasma.

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