Regulation of sperm storage and movement in the ruminant oviduct

PH Hung and SS Suarez

Department of Biomedical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA

Three regions of the ruminant oviduct play different roles in the progress of sperm: the uterotubal junction, isthmus, and ampulla. The uterotubal junction acts as a point of selection of sperm, requiring that sperm are progressively motile and express specific proteins in order to enter the oviduct. The isthmus stores sperm, preserving motility and viability until ovulation. Sperm are stored in the isthmus by binding to its mucosal epithelium. In bovine sperm, binding to the oviductal epithelium is promoted by proteins that are secreted by the seminal vesicles and coat the heads of sperm by associating with plasma membrane phospholipids. Putative oviductal receptors for the seminal vesicle proteins are members of the annexin protein family. Release of sperm from the storage site in the isthmus is gradual, which serves to ensure that sperm in the proper physiological state reach the oocytes at the appropriate moment and also to reduce incidence of polyspermic fertilization. The ampulla supports fertilization and may participate in guiding sperm toward the eggs. Further studies are needed to improve our understanding of the interactions between sperm and the female reproductive tract, in order to develop means to improve fertility in ruminants.

Introduction

Mammalian sperm travel a long distance relative to their size in order to reach the site of fertilization in the oviduct. When sperm attempt to enter and pass through the oviduct, they are subjected to selective processes to eliminate those of poor quality. Selective processes also regulate the numbers of sperm that reach the fertilization site, thereby reducing the incidence of polyspermic fertilization (Hunter *et al.* 1982, Hunter & Wilmut 1984). The oviduct acts as well to support fertilization, for instance, by storing sperm and maintaining their viability during storage. Furthermore, the timing of capacitation of sperm may be influenced by the oviduct in order to ensure that capacitated sperm are available at ovulation.

Three regions of the oviduct, uterotubal junction, isthmus, and ampulla (Figure 1) play different roles in selecting and supporting sperm. In this review, the current understanding of the roles of each region will be discussed, particularly with regard to ruminant reproduction.

Corresponding author E-mail: sss7@cornell.edu

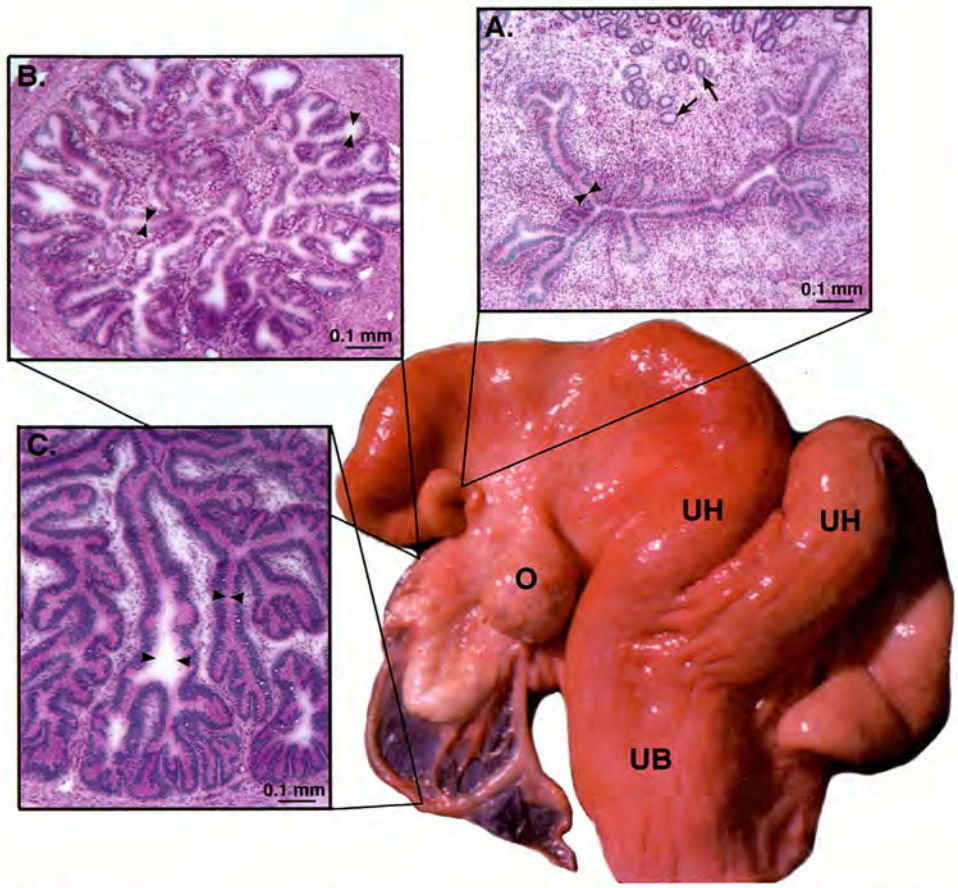

Fig. 1. Regions of the bovine oviduct. On the right, the regions of the bovine female tract are shown (**O**, ovary; **UH** uterine horn; **UB**, lower uterine body). **A.** A frozen section of the bovine uterotubal junction, stained with Periodic Acid Schiff to indicate mucopolysaccharides, and counterstained with hematoxylin. Arrowheads indicate the oviductal lumen, which is narrow and contains mucus. Arrows indicate uterine glands. **B.** A frozen section of the oviductal isthmus, stained as for A. Arrowheads indicate the oviductal lumen. **C.** A frozen section of approximately half of the diameter of the oviductal ampulla, stained as for A. Arrowheads indicate the oviductal lumen. Preparation of these tissues is described in Suarez *et al.* (1997).

The uterotubal junction regulates sperm passage into the oviduct

Regardless of the billions of sperm in the ejaculates of most ruminants, only a minority of several thousand enters the uterotubal junction and passes through it. There may be only a small window of opportunity for sperm to pass through the junction, due to the apparent ability of the junction to close down. In the mouse, patency of the junction can be seen using transillumination of the oviduct and the junction has been observed to close about an hour after mating (Suarez 1987). Three morphological characteristics of the ruminant oviduct indicate that it may do likewise. First, the connective tissue in the bovine junction wall is heavily vascularized and engorgement of the vascular beds may act to compress the lumen (Wrobel *et al.* 1993). Second,

the thick smooth muscle layer of the junction could contract to compress the lumen. Third, the tube of the junction is bent into a sigmoidal shape and the muscular ligaments attached to the junction could increase the flexure of the curve to further impede passage of sperm (Hafez & Black 1969, Hook & Hafez 1968).

In the cow, the uterotubal junction is lined with mucosal folds that form channels ending in cul-de-sacs rather than continuing into the isthmus (Figure 2) (Yaniz *et al.* 2000). When muscular and/or vascular action compresses the lumen, these dead ends might form a plug. On the other hand, when the uterotubal junction is not being compressed, the dead ends of the channels could act more like funnels to direct sperm into the isthmus.

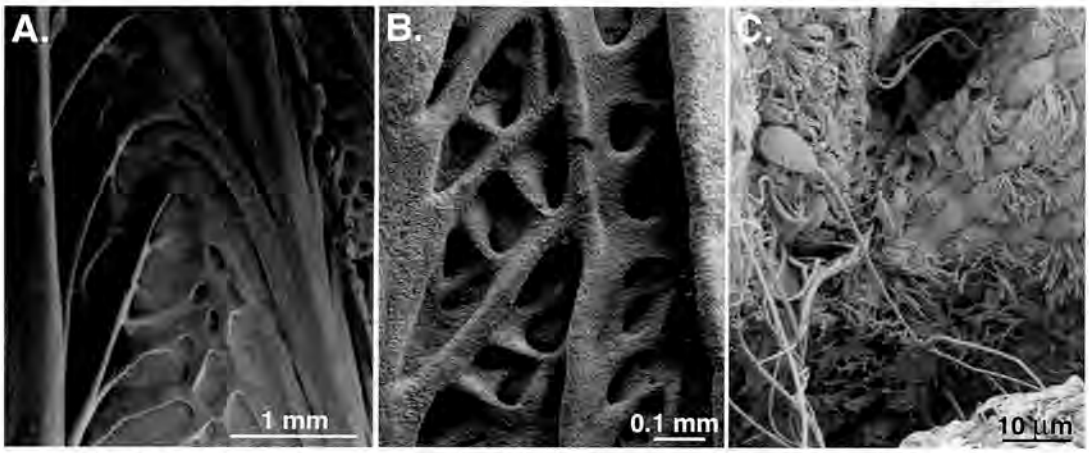

Fig. 2. Scanning electron micrographs of the bovine uterotubal junction and isthmus. **A.** The uterotubal junction is lined with mucosal folds that form channels ending in cul-de-sacs. The bottom of the figure represents the prouterine end of the uterotubal junction. **B.** Mucosal folds in the lower isthmus form pockets in the wall. **C.** Bull sperm binding primarily to cilia on the oviductal epithelium in the lower isthmus to form a reservoir. Figure 2A and 2B are courtesy of Dr. JL Yaniz (Yaniz *et al.* 2000).

When the uterotubal junction is not being closed down by vascular and/or muscular action, the junction is equipped with mechanisms to cause it to act as a point of sperm selection. There are mucous secretions in the lumen of bovine uterotubal junction (Suarez *et al.* 1997) that might filter out sperm with weak motility, as does mucus in the cervical canal (Katz *et al.* 1990). Furthermore, based on null mutant mouse models, there is evidence that mouse sperm must possess certain proteins in order to swim into the uterotubal junction (Nakanishi *et al.* 2004, Yamaguchi *et al.* 2009). For example, calmegin is a spermatogenesis-specific chaperone protein, and the sperm produced by calmegin knockout mice are unable to pass through the uterotubal junction, despite having normal morphology and motility (Ikawa *et al.* 2001). A subsequent study confirmed that each sperm must possess the proteins chaperoned by calmegin, because when females were mated with chimeric males that produced a mixture of developing germ cells in the testis that contained or were missing calmegin, the only sperm that entered into the uterotubal junctions where those in which calmegin had been expressed during development (Nakanishi *et al.* 2004). Calmegin is known to be required for the expression of ADAM3 (a disintegrin and metallopeptidase domain 3) on mature sperm, and sperm from male mice deficient in ADAM3 are also unable to pass through the uterotubal junction (Yamaguchi *et al.* 2009). It is not yet known whether similar proteins are required for ruminant sperm to pass through the uterotubal junction.

In cattle (VanDemark & Moeller 1951), sheep (Mattner & Braden 1963), humans (Kunz *et al.* 1996), and rabbits (Overstreet & Cooper 1978), a small fraction of sperm are transported through the uterus and oviduct to the site of fertilization within a few minutes of mating. This phenomenon, which has been called "rapid sperm transport", may be a consequence of muscular contractions in the female tract that occur during mating, because it occurs too rapidly for it to be the result of sperm swimming and it may not serve to bring competent sperm to the site of fertilization (Hays & VanDemark 1953). For instance, rabbit sperm that underwent rapid transport were found to have sustained damage and thus would be unlikely to fertilize (Overstreet & Cooper 1978, Overstreet & Tom 1982). It is not known whether rapid transport serves any purpose.

Sperm are stored in the isthmus

After sperm pass through the intramural portion of the uterotubal junction, most bind to the epithelium in the extramural portion and/or the lower part of the isthmus of the oviduct to form a storage reservoir. Oviductal reservoirs of sperm have been identified in a broad array of mammals, including sheep (Hunter *et al.* 1982), cattle (Hunter & Wilmut 1984), mice (Suarez 1987), and hamsters (Smith & Yanagimachi 1991). There is evidence that binding to the oviductal epithelium prolongs the motile life span of ruminant sperm. When sperm of cattle (Chian *et al.* 1995, Pollard *et al.* 1991), sheep (Lloyd *et al.* 2008, Lloyd *et al.* 2009), or red deer (Berg *et al.* 2002) were incubated with oviductal epithelium in vitro, higher percentages of the sperm remained motile over time. The mechanism for sustaining motility has been proposed to involve suppression of capacitation, because equine sperm showed delayed capacitation when incubated with oviductal epithelium (Dobrinski *et al.* 1996). Elucidating the mechanism of preservation of motility during storage might suggest new methods for storing sperm in vitro.

There is evidence that sperm at least begin to capacitate while in the oviductal isthmus (Smith & Yanagimachi 1991). It is not fully understood how sperm become capacitated in the oviduct. In vitro, it has been shown that capacitation involves loss of decapacitating factors including cholesterol, a rise in intracellular Ca^{2+}, and an increase in activity of a sperm-specific adenylyl cyclase known as SACY (Sinclair *et al.* 2000). SACY activation results in increased cAMP, which, in turn, activates protein kinase A (PKA) (reviewed by Visconti *et al.* 2002). Through a poorly understood process, activation of PKA, a serine and threonine kinase, leads to increased tyrosine phosphorylation of several sperm proteins (reviewed by Visconti *et al.* 2002), such as testis-specific serine/proline-rich protein, calcium binding protein, and others (Platt *et al.* 2009). Studies in mice (Visconti *et al.* 1995) and cattle (Galantino-Homer *et al.* 1997) have shown that protein tyrosine phosphorylation is correlated with capacitation, confirmed by the ability of sperm to undergo induced acrosome reactions and to fertilize eggs in vitro. Most of the roles of tyrosine phosphorylated proteins in sperm capacitation remain to be determined.

Bull sperm capacitation is facilitated in vitro when heparin is present (Parrish *et al.* 1988). There is evidence that the effect of heparin on sperm is mediated by bovine seminal plasma proteins (binder of sperm, BSP) (Manjunath *et al.* 2009) on the sperm surface, particularly PDC109 (also known as BSPA1/A2) (Manjunath & Therien 2002, Therien *et al.* 1997). BSP proteins are produced by the bovine seminal vesicles and adhere to the sperm surface by associating with phospholipids in the sperm plasma membrane (Desnoyers & Manjunath 1992). BSP proteins are small in mass (14-30 kDa) and each contains a unique N-terminal domain followed by two fibronectin type II domains (Calvete *et al.* 1999). The type II domains contain heparin binding sites (Chandonnet *et al.* 1990) that could be responsible for the stimulation of capacitation by heparin, and phospholipid binding sites that are responsible for binding the BSPs to the

sperm surface (Wah *et al.* 2002). BSP homologs have been reported in other ruminant species, including goats (Villemure *et al.* 2003), sheep (Bergeron *et al.* 2005), and bisons (Boisvert *et al.* 2004), and some of the homologs have been demonstrated to bind heparin (Bergeron *et al.* 2005, Boisvert *et al.* 2004, Villemure *et al.* 2003).

The process of capacitation is considered to include hyperactivation of sperm motility. Hyperactivated sperm display asymmetrical, high-amplitude flagellar beating patterns, causing vigorous, and sometimes circular, movement of free-swimming sperm (Suarez *et al.* 1983, Yanagimachi 1970). Hyperactivated motility is required for the penetration of viscoelastic substances in the oviduct (Suarez *et al.* 1991), such as mucus and the matrix of the cumulus oophorus, and for sperm penetration of the zona pellucida (Stauss *et al.* 1995). Hyperactivation is triggered by a rise of intracellular Ca^{2+}, primarily from extracellular sources (reviewed by Suarez 2008). In cattle, sperm can be induced to hyperactivate in vitro by procaine in the presence of extracellular Ca^{2+} and therefore is presumed to operate by stimulating Ca^{2+} influx (Ho & Suarez 2001, Ho & Suarez 2003). In mouse sperm, sperm-specific CatSper proteins are known to form Ca^{2+} channels in the plasma membrane of the principal piece of the flagellum (Carlson *et al.* 2005, Jin *et al.* 2007). Sperm from CatSper-null mice do not hyperactivate (Carlson *et al.* 2005, Jin *et al.* 2007, Qi *et al.* 2007) and cannot fertilize zona pellucida-intact eggs (Quill *et al.* 2003, Ren *et al.* 2001).

BSP proteins play a role in sperm storage

In hamsters (DeMott *et al.* 1995), cattle (Lefebvre *et al.* 1997), and pigs (Wagner *et al.* 2002), studies have shown that sperm-oviductal epithelium interaction is mediated by carbohydrate-recognition mechanisms. Fucose, specifically in the Lewis-A trisaccharide, was identified to be a key component of the receptor for bull sperm binding in the oviduct (Suarez *et al.* 1998). Lewis-A was used in an affinity column to pull proteins from sperm extracts that would play a role in binding sperm to the oviductal epithelium. The main protein identified by this method was PDC109 (Gwathmey *et al.* 2003, Ignotz *et al.* 2001), which is one of the BSP proteins. Gwathmey *et al.* (2006) later reported that two other BSP proteins (BSPA3 and BSP30K), each acting alone, can also induce sperm binding to oviductal epithelium in vitro. This leads to the question: Why do bull sperm need three BSP proteins when they can bind to the oviductal epithelium with any one of the BSP proteins alone? Key functions are often protected by redundancy; however, this apparent redundancy may also provide more intricate control over sperm movement out of the reservoir. Differences in the amino acid sequences of the three BSP proteins result in different patterns of surface charges on the BSP molecules (Gwathmey *et al.* 2006); therefore, it is proposed that each BSP molecule adheres to the surface of sperm and binds sperm to the oviductal epithelium with different affinities and kinetics. Altogether, the BSP proteins may act to provide a gradual release of sperm from the reservoir, in order to ensure that sperm capacitate and reach eggs at the ideal time for fertilization and yet not too many reach the egg at once, which might result in polyspermic fertilization. It is possible that BSP homologs in other ruminant species are also involved in the formation of sperm reservoir; however, this remains to be determined.

Oviductal annexins are possible binding partners of BSP proteins

When BSP proteins were used as bait to pull down oviductal ligands from a mixture of proteins extracted from apical plasma membranes of bovine isthmic epithelium, four annexin proteins (ANXA1, 2, 4, and 5) were captured (Ignotz *et al.* 2007). ANXAs comprise a large, diverse

family of Ca^{2+}- and lipid-binding proteins. In other cell types, there is evidence that ANXAs serve as membrane scaffold proteins and are involved in vesicle transport and protein secretion, but much of their functions and secretory pathways are still poorly understood (reviewed by Rescher & Gerke 2004).

In support of the proposal that ANXAs are the oviductal receptors for sperm, all four ANXAs were immunolocalized on the apical surfaces of oviductal epithelium (Ignotz *et al.* 2007). Western blot analysis confirmed that oviductal ANXAs contain fucose, and sperm binding to oviductal epithelium can be inhibited in vitro in the presence of ANXA antibodies (Ignotz *et al.* 2007). There is also evidence that one of the ANXAs (ANXA2) is a sperm binding protein on the porcine oviductal epithelium (Teijeiro *et al.* 2009). Like BSP proteins, ANXAs were also found to bind to heparin (Ishitsuka *et al.* 1998, Shao *et al.* 2006). This interesting finding implicates heparin, or a similar glycosaminoglycan, in the process of binding or release of sperm, particularly since heparin-like molecules have been identified in oviductal fluid (Parrish *et al.* 1989) and have been shown to trigger release of bovine sperm from monolayers of oviductal epithelium in vitro (Gualtieri & Talevi 2000).

Sperm movement beyond the reservoir to the ampulla

In cattle, sperm are stored in the isthmic region of the oviduct for 18-20 hours or more before being released to ascend to the ampulla (Hunter & Wilmut 1984). The release of sperm from the reservoir begins prior to ovulation, which occurs 28-31 hours after the onset of estrus in cattle (Hunter & Wilmut 1984). It is unlikely that the release of sperm is due to the loss of binding sites on the oviductal epithelium, as sperm can bind to oviducts from different stages of estrus cycle (Lefebvre *et al.* 1995). Rather, it is likely that hormonal changes that trigger ovulation also stimulate the release of factors in the oviduct that cause changes in sperm which enable them to release themselves from the oviductal epithelium.

There is evidence that sperm release themselves from the reservoir by two mechanisms: hyperactivation and shedding of sperm surface proteins during capacitation. First, hyperactivation was proposed to play a role in release of mouse and human sperm, (DeMott & Suarez 1992, Pacey *et al.* 1995), because sperm were seen to hyperactivate before pulling away from the epithelial surface. In addition, sperm from CatSper null mice, which are unable to hyperactivate, fail to move beyond the sperm storage reservoir (Ho *et al.* 2009). It is not yet known whether hyperactivation plays such a role in ruminants. Second, sperm may lose binding affinity for the oviductal epithelium by shedding BSP proteins during capacitation. It has been reported that bull sperm shed the BSP protein PDC109 during capacitation in vitro and capacitated sperm are less able to bind to epithelium unless they are treated with purified PDC109 (Gwathmey *et al.* 2003). Less is known of the roles of BSPA3 and BSP30K; however, because capacitated sperm lose binding affinity for the epithelium, one would predict that these two proteins are also shed during capacitation. Because BSPA3 and BSP30K differ from PDC109 in molecular surface charges (Gwathmey *et al.* 2006), we predict that the kinetics of loss during capacitation differs from that of PDC109. Differential loss of BSP proteins could serve to spread out the release of sperm from the reservoir and thus assure that sperm reach eggs shortly after they enter the oviduct, but that not so many reach eggs that polyspermy occurs.

After sperm are released from the reservoir, they are still required to travel a long distance before they reach the fertilization site. Furthermore, as sperm move up the isthmus into the ampulla, the diameter of the tube increases and the shape of the lumen becomes even more complicated by elaboration of the mucosal folds that create narrow, labyrinthine passages (Figure 1). How sperm find their way to the egg is still largely unknown. It has been proposed that

chemotaxis serves to guide sperm toward eggs (reviewed by Kaupp *et al.* 2008). The existence of chemotaxis has been well documented in several species of marine invertebrates (reviewed by Hildebrand & Kaupp 2005); therefore, it has been hypothesized that chemotactic factors direct mammalian sperm to eggs. In humans (Cohen-Dayag *et al.* 1995, Spehr *et al.* 2003, Villanueva-Diaz *et al.* 1990) and rabbits (Fabro *et al.* 2002), sperm reportedly turn to swim up a gradient of follicular fluid or putative chemotactic agents, indicating that chemotaxis plays a role in mammalian fertilization; however, unlike the massive response shown by various species of marine invertebrate sperm (Cook *et al.* 1994, Yoshida *et al.* 2003), only small percentages of mammalian sperm (2-12% in humans) have shown this response in vitro (Gakamsky *et al.* 2008). In ruminant species, 8-10% of frozen-thawed bull sperm were reported to orient into a gradient of follicular fluid (Gil *et al.* 2008). Some follicular fluid escapes from the oviduct with the egg mass during fertilization and thus could be present in the ampulla to attract sperm toward the site of fertilization.

Conclusions

Studies from different animal models have helped researchers to better understand the regulation of sperm storage and movement in the oviduct; however, not much research has been done in ruminant species other than *Bos taurus*. There is still much to be learned about how sperm entry through the uterotubal junction is regulated, how sperm fertility is maintained during storage in the oviductal reservoir, how sperm are released from the reservoir, and whether sperm are guided toward eggs in the ampulla by chemotaxis. Such information could prove valuable for developing new methods to improve sperm storage and the success rate of artificial insemination of domestic ruminants and endangered wildlife species.

References

Berg DK, Thompson JG & Asher GW 2002 Development of in vitro embryo production systems for red deer (Cervus elaphus). Part 1. Effect of epithelial oviductal monolayers and heparin on in vitro sperm motility and penetration of in vitro matured oocytes. *Anim Reprod Sci* **70** 65-76.

Bergeron A, Villemure M, Lazure C & Manjunath P 2005 Isolation and characterization of the major proteins of ram seminal plasma. *Molecular Reproduction and Development* **71** 461-470.

Boisvert M, Bergeron A, Lazure C & Manjunath P 2004 Isolation and characterization of gelatin-binding bison seminal vesicle secretory proteins. *Biology of Reproduction* **70** 656-661.

Calvete JJ, Campanero-Rhodes MA, Raida M & Sanz L 1999 Characterisation of the conformational and quaternary structure-dependent heparin-binding region of bovine seminal plasma protein PDC-109. *FEBS Letters* **444** 260-264.

Carlson AE, Quill TA, Westenbroek RE, Schuh SM, Hille B & Babcock DF 2005 Identical phenotypes of CatSper1 and CatSper2 null sperm. *Journal of Biological Chemistry* **280** 32238-32244.

Chandonnet L, Roberts KD, Chapdelaine A & Manjunath P 1990 Identification of heparin-binding proteins in bovine seminal plasma. *Molecular Reproduction and Development* **26** 313-318.

Chian RC, Lapointe S & Sirard MA 1995 Capacitation in vitro of bovine spermatozoa by oviduct epithelial cell monolayer conditioned medium. *Molecular Reproduction and Development* **42** 318-324.

Cohen-Dayag A, Tur-Kaspa I, Dor J, Mashiach S & Eisenbach M 1995 Sperm capacitation in humans is transient and correlates with chemotactic responsiveness to follicular factors. *Proceedings of the National Academy of Sciences of the United States of America* **92** 11039-11043.

Cook SP, Brokaw CJ, Muller CH & Babcock DF 1994 Sperm chemotaxis: Egg peptides control cytosolic calcium to regulate flagellar responses. *Developmental Biology* **165** 10-19.

DeMott RP, Lefebvre R & Suarez SS 1995 Carbohydrates mediate the adherence of hamster sperm to oviductal epithelium. *Biology of Reproduction* **52** 1395-1403.

DeMott RP & Suarez SS 1992 Hyperactivated sperm progress in the mouse oviduct. *Biology of Reproduction* **46** 779-785.

Desnoyers L & Manjunath P 1992 Major proteins of bovine seminal plasma exhibit novel interactions with phospholipid. *Journal of Biological Chemistry* **267** 10149-10155.

Dobrinski I, Suarez SS & Ball BA 1996 Intracellular

calcium concentration in equine spermatozoa attached to oviductal epithelial cells in vitro. *Biology of Reproduction* **54** 783-788.

Fabro G, Rovasio RA, Civalero S, Frenkel A, Caplan SR, Eisenbach M & Giojalas LC 2002 Chemotaxis of capacitated rabbit spermatozoa to follicular fluid revealed by a novel directionality-based assay. *Biology of Reproduction* **67** 1565-1571.

Gakamsky A, Schechtman E, Caplan SR & Eisenbach M 2008 Analysis of chemotaxis when the fraction of responsive cells is small–application to mammalian sperm guidance. *International Joural of Developmental Biology* **52** 481-487.

Galantino-Homer HL, Visconti PE & Kopf GS 1997 Regulation of protein tyrosine phosphorylation during bovine sperm capacitation by a cyclic adenosine 3′5′-monophosphate-dependent pathway. *Biology of Reproduction* **56** 707-719.

Gil PI, Guidobaldi HA, Teves ME, Unates DR, Sanchez R & Giojalas LC 2008 Chemotactic response of frozen-thawed bovine spermatozoa towards follicular fluid. *Animal Reproduction Science* **108** 236-246.

Gualtieri R & Talevi R 2000 In vitro-cultured bovine oviductal cells bind acrosome-intact sperm and retain this ability upon sperm release. *Biology of Reproduction* **62** 1754-1762.

Gwathmey TM, Ignotz GG, Mueller JL, Manjunath P & Suarez SS 2006 Bovine seminal plasma proteins PDC-109, BSP-A3, and BSP-30-kDa share functional roles in storing sperm in the oviduct. *Biology of Reproduction* **75** 501-507.

Gwathmey TM, Ignotz GG & Suarez SS 2003 PDC-109 (BSP-A1/A2) promotes bull sperm binding to oviductal epithelium in vitro and may be involved in forming the oviductal sperm reservoir. *Biology of Reproduction* **69** 809-815.

Hafez ES & Black DL 1969 The mammalian uterotubal junction. In: *The Mammalian Oviduct: Comparative Biology and Methodology*, 85-128. Eds ESE Hafez & RJ Blandau. Chicago: The University of Chicago Press.

Hays RL & VanDemark NL 1953 Spontaneous motility of the bovine uterus. *American Journal of Physiology* **172** 553-556.

Hildebrand E & Kaupp UB 2005 Sperm chemotaxis: a primer. *Annals of the New York Academy of Sciences* **1061** 221-225.

Ho HC & Suarez SS 2001 An inositol 1,4,5-trisphosphate receptor-gated intracellular Ca^{2+} store is involved in regulating sperm hyperactivated motility. *Biology of Reproduction* **65** 1606-1615.

Ho HC & Suarez SS 2003 Characterization of the intracellular calcium store at the base of the sperm flagellum that regulates hyperactivated motility. *Biology of Reproduction* **68** 1590-1596.

Ho K, Wolff CA & Suarez SS 2009 CatSper-null mutant spermatozoa are unable to ascend beyond the oviductal reservoir. *Reproduction, Fertility, and Development* **21** 345-350.

Hook SJ & Hafez ES 1968 A comparative anatomical study of the mammalian uterotubal junction. *Journal of Morphology* **125** 159-184.

Hunter RH, Barwise L & King R 1982 Sperm transport, storage and release in the sheep oviduct in relation to the time of ovulation. *British Veterinary Journal* **138** 225-232.

Hunter RH & Wilmut I 1984 Sperm transport in the cow: peri-ovulatory redistribution of viable cells within the oviduct. *Reproduction Nutrition Development* **24** 597-608.

Ignotz GG, Cho MY & Suarez SS 2007 Annexins are candidate oviductal receptors for bovine sperm surface proteins and thus may serve to hold bovine sperm in the oviductal reservoir. *Biology of Reproduction* **77** 906-913.

Ignotz GG, Lo MC, Perez CL, Gwathmey TM & Suarez SS 2001 Characterization of a fucose-binding protein from bull sperm and seminal plasma that may be responsible for formation of the oviductal sperm reservoir. *Biology of Reproduction* **64** 1806-1811.

Ikawa M, Nakanishi T, Yamada S, Wada I, Kominami K, Tanaka H, Nozaki M, Nishimune Y & Okabe M 2001 Calmegin is required for fertilin α/β heterodimerization and sperm fertility. *Developmental Biology* **240** 254-261.

Ishitsuka R, Kojima K, Utsumi H, Ogawa H & Matsumoto I 1998 Glycosaminoglycan binding properties of annexin IV, V, and VI. *Journal of Biological Chemistry* **273** 9935-9941.

Jin J, Jin N, Zheng H, Ro S, Tafolla D, Sanders KM & Yan W 2007 Catsper3 and Catsper4 are essential for sperm hyperactivated motility and male fertility in the mouse. *Biology of Reproduction* **77** 37-44.

Katz DF, Morales P, Samuels SJ & Overstreet JW 1990 Mechanisms of filtration of morphologically abnormal human sperm by cervical mucus. *Fertility and Sterility* **54** 513-516.

Kaupp UB, Kashikar ND & Weyand I 2008 Mechanisms of sperm chemotaxis. *Annual Review of Physiology* **70** 93-117.

Kunz G, Beil D, Deininger H, Wildt L & Leyendecker G 1996 The dynamics of rapid sperm transport through the female genital tract: evidence from vaginal sonography of uterine peristalsis and hysterosalpingoscintigraphy. *Human Reproduction* **11** 627-632.

Lefebvre R, Chenoweth PJ, Drost M, LeClear CT, MacCubbin M, Dutton JT & Suarez SS 1995 Characterization of the oviductal sperm reservoir in cattle. *Biol Reprod* **53** 1066-1074.

Lefebvre R, Lo MC & Suarez SS 1997 Bovine sperm binding to oviductal epithelium involves fucose recognition. *Biology of Reproduction* **56** 1198-1204.

Lloyd RE, Badia E, Fazeli A, Watson PF & Holt WV 2008 Temporal dynamics of ram sperm binding and survival during 48-h coculture with oviducal epithelial cells. *Reproduction, Fertility, and Development* **20** 835-845.

Lloyd RE, Elliott RM, Fazeli A, Watson PF & Holt WV 2009 Effects of oviductal proteins, including heat shock 70 kDa protein 8, on survival of ram spermatozoa over 48 h in vitro. *Reproduction, Fertility, and Development* **21** 408-418.

Manjunath P & Therien I 2002 Role of seminal plasma phospholipid-binding proteins in sperm membrane lipid modification that occurs during capacitation. *Journal of Reproductive Immunology* **53** 109-119.

Manjunath P, Lefebvre J, Jois PS, Fan J & Wright MW 2009 New nomenclature for mammalian BSP genes. *Biology of Reproduction* **80** 394-397.

Mattner PE & Braden AWH 1963 Spermatozoa in the genital tract of the ewe. I. Rapidity of transport. *Australian Journal of Biological Sciences* **16** 473.

Nakanishi T, Isotani A, Yamaguchi R, Ikawa M, Baba T, Suarez SS & Okabe M 2004 Selective passage through the uterotubal junction of sperm from a mixed population produced by chimeras of calmegin-knockout and wild-type male mice. *Biology of Reproduction* **71** 959-965.

Overstreet JW & Cooper GW 1978 Sperm transport in the reproductive tract of the female rabbit: I. The rapid transit phase of transport. *Biology of Reproduction* **19** 101-114.

Overstreet JW & Tom RA 1982 Experimental studies of rapid sperm transport in rabbits. *Reproduction* **66** 601-606.

Pacey AA, Davies N, Warren MA, Barratt CLR & Cooke LD 1995 Hyperactivation may assist human spermatozoa to detach from intimate association with the endosalpinx. *Human Reproduction* **10** 2603-2609.

Parrish JJ, Susko-Parrish J, Winer MA & First NL 1988 Capacitation of bovine sperm by heparin. *Biology of Reproduction* **38** 1171-1180.

Parrish JJ, Susko-Parrish JL, Handrow RR, Sims MM & First NL 1989 Capacitation of bovine spermatozoa by oviduct fluid. *Biology of Reproduction* **40** 1020-1025.

Platt MD, Salicioni AM, Hunt DF & Visconti PE 2009 Use of differential isotopic labeling and mass spectrometry to analyze capacitation-associated changes in the phosphorylation status of mouse sperm proteins. *J Proteome Res* **8** 1431-1440.

Pollard JW, Plante C, King WA, Hansen PJ, Betteridge KJ & Suarez SS 1991 Fertilizing capacity of bovine sperm may be maintained by binding of oviductal epithelial cells. *Biology of Reproduction* **44** 102-107.

Qi H, Moran MM, Navarro B, Chong JA, Krapivinsky G, Krapivinsky L, Kirichok Y, Ramsey IS, Quill TA & Clapham DE 2007 All four CatSper ion channel proteins are required for male fertility and sperm cell hyperactivated motility. *Proceedings of the National Academy of Sciences of the United States of America* **104** 1219-1223.

Quill TA, Sugden SA, Rossi KL, Doolittle LK, Hammer RE & Garbers DL 2003 Hyperactivated sperm motility driven by CatSper2 is required for fertilization. *Proceedings of the National Academy of Sciences of the United States of America* **100** 14869-14874.

Ren D, Navarro B, Perez G, Jackson AC, Hsu S, Shi Q, Tilly JL & Clapham DE 2001 A sperm ion channel required for sperm motility and male fertility. *Nature* **413** 603-609.

Rescher U & Gerke V 2004 Annexins - unique membrane binding proteins with diverse functions. *Journal of Cell Science* **117** 2631-2639.

Shao C, Zhang F, Kemp MM, Linhardt RJ, Waisman DM, Head JF & Seaton BA 2006 Crystallographic analysis of calcium-dependent heparin binding to annexin A2. *Journal of Biological Chemistry* **281** 31689-31695.

Sinclair ML, Wang XY, Mattia M, Conti M, Buck J, Wolgemuth DJ & Levin LR 2000 Specific expression of soluble adenylyl cyclase in male germ cells. *Mol Reprod Dev* **56** 6-11.

Smith TT & Yanagimachi R 1991 Attachment and release of spermatozoa from the caudal isthmus of the hamster oviduct. *Journal of Reproduction and Fertility* **91** 567-573.

Spehr M, Gisselmann G, Poplawski A, Riffell JA, Wetzel CH, Zimmer RK & Hatt H 2003 Identification of a testicular odorant receptor mediating human sperm chemotaxis. *Science* **299** 2054-2058.

Stauss CR, Votta TJ & Suarez SS 1995 Sperm motility hyperactivation facilitates penetration of the hamster zona pellucida. *Biology of Reproduction* **53** 1280-1285.

Suarez SS 1987 Sperm transport and motility in the mouse oviduct: observations in situ. *Biology of Reproduction* **36** 203-210.

Suarez SS 2008 Control of hyperactivation in sperm. *Human Reproduction Update* **14** 647-657.

Suarez SS, Brockman K & Lefebvre R 1997 Distribution of mucus and sperm in bovine oviducts after artificial insemination: the physical environment of the oviductal sperm reservoir. *Biology of Reproduction* **56** 447-453.

Suarez SS, Katz DF & Overstreet JW 1983 Movement characteristics and acrosomal status of rabbit spermatozoa recovered at the site and time of fertilization. *Biology of Reproduction* **29** 1277-1287.

Suarez SS, Katz DF, Owen DH, Andrew JB & Powell RL 1991 Evidence for the function of hyperactivated motility in sperm. *Biology of Reproduction* **44** 375-381.

Suarez SS, Revah I, Lo M & Kolle S 1998 Bull sperm binding to oviductal epithelium is mediated by a Ca^{2+}-dependent lectin on sperm that recognizes Lewis-a trisaccharide. *Biology of Reproduction* **59** 39-44.

Teijeiro JM, Ignotz GG & Marini PE 2009 Annexin A2 is involved in pig (Sus scrofa) sperm-oviduct interaction. *Molecular Reproduction and Development* **76** 334-341.

Therien I, Soubeyrand S & Manjunath P 1997 Major proteins of bovine seminal plasma modulate sperm capacitation by high-density lipoprotein. *Biology of Reproduction* **57** 1080-1088.

VanDemark NL & Moeller AN 1951 Speed of spermatozoa transport in reproductive tract of estrous cow. *American Journal of Physiology* **165** 674-679.

Villanueva-Diaz C, Vadillo-Ortega F, Kably-Ambe A, Diaz-Perez MA & Krivitzky SK 1990 Evidence that human follicular fluid contains a chemoattractant for spermatozoa. *Fertility and Sterility* **54** 1180-1182.

Villemure M, Lazure C & Manjunath P 2003 Isolation and characterization of gelatin-binding proteins from goat seminal plasma. *Reproductive Biology and Endocrinology* **1** 39.

Visconti PE, Bailey JL, Moore GD, Pan D, Olds-Clarke P & Kopf GS 1995 Capacitation of mouse spermatozoa. I. Correlation between the capacitation state and protein tyrosine phosphorylation. *Development* **121** 1129-1137.

Visconti PE, Westbrook VA, Chertihin O, Demarco I, Sleight S & Diekman AB 2002 Novel signaling pathways involved in sperm acquisition of fertilizing capacity. *Journal of Reproductive Immunology* **53** 133-150.

Wagner A, Ekhlasi-Hundrieser M, Hettel C, Petrunkina A, Waberski D, Nimtz M & Topfer-Petersen E 2002 Carbohydrate-based interactions of oviductal sperm reservoir formation-studies in the pig. *Molecular Reproduction and Development* **61** 249-257.

Wah DA, Fernandez-Tornero C, Sanz L, Romero A & Calvete JJ 2002 Sperm coating mechanism from the 1.8 A crystal structure of PDC-109-phosphorylcholine complex. *Structure* **10** 505-514.

Wrobel KH, Kujat R & Fehle G 1993 The bovine tubouterine junction: general organization and surface morphology. *Cell and Tissue Research* **271** 227-239.

Yamaguchi R, Muro Y, Isotani A, Tokuhiro K, Takumi K, Adham I, Ikawa M & Okabe M 2009 Disruption of ADAM3 impairs the migration of sperm into oviduct in mouse. *Biology of Reproduction* **81** 142-146.

Yanagimachi R 1970 The movement of golden hamster spermatozoa before and after capacitation. *Journal of Reproduction and Fertility* **23** 193-196.

Yaniz JL, Lopez-Gatius F, Santolaria P & Mullins KJ 2000 Study of the functional anatomy of bovine oviductal mucosa. *Anatomical Record* **260** 268-278.

Yoshida M, Ishikawa M, Izumi H, De Santis R & Morisawa M 2003 Store-operated calcium channel regulates the chemotactic behavior of ascidian sperm. *Proceedings of the National Academy of Sciences of the United States of America* **100** 149-154.