Molecular Biology of the Cell
Vol. 10, 5–8, January 1999

Video Essay

Oocyte Pickup by the Mammalian Oviduct[V]

Prue Talbot, Christine Geiske, and Michael Knoll

Department of Biology, University of California, Riverside, California 92521

Submitted September 14, 1998; Accepted October 26, 1998
Monitoring Editor: Jennifer Lippincott-Schwartz

INTRODUCTION

Mammals ovulate an oocyte cumulus complex (OCC), which contains, in addition to the oocyte, numerous cumulus cells and an extensive extracellular matrix (Mahi-Brown and Yanagimachi, 1983; Talbot and DiCarlantonio, 1984). OCCs are ovulated into the peritoneal or bursal cavity and then picked up by the infundibulum of the oviduct and carried into the ampulla where fertilization occurs (Harper, 1994; Hunter, 1994). Pickup of the OCC by the oviduct is the first in an important series of gamete transport events that assure entry of blastocyts into the uterus at the proper time for implantation. The infundibulum has a highly convoluted surface characterized by numerous deep crevices and ridges leading to the opening or ostium through which the OCC enters the oviduct (Figure 1A). The epithelium on the exterior surface of the infundibulum is enriched with cilia that beat in the direction of the ostium (Figure 1B). Although these cilia create currents capable of moving small particles such as *Lycopodium* spores (Norwood, *et al.*, 1978), ciliary currents alone cannot move larger objects, such as OCCs (0.7–1.0 mm diameter), into the ostium. Pickup of OCCs occurs by means of an adhesive interaction between the OCC and cilia coupled with ciliary beating that pulls the OCC toward the ostium (Norwood, *et al.*, 1978). The following video sequences show the two mechanisms that allow particles to be moved by the cilia on the outer surface of the infundibulum and the importance of adhesion in OCC pickup.

VIDEO SEQUENCES

Video Sequence 1: Movement of Lycopodium Spores in Oviductal Ciliary Currents

A dissected hamster infundibulum has been mounted in a tissue culture dish to examine movement of *Lycopodium* spores on its surface (Knoll and Talbot, 1998). Cilia cover the entire surface of the infundibulum but are in focus only at the periphery. In this video sequence, most spores tumble over the surface of the infundibulum in ciliary currents, while several spores are either stuck in crevices or are attached to cilia and remain stationary (Figure 2A). Oviductal cilia beat at 6–8 hertz when cultured at this temperature (DiCarlantonio *et al.*, 1995). Because spores are relatively small particles, they are transferred rapidly to the ostium. Spores move over the surface of the hamster infundibulum at a rate of ~70–100 μm/s when incubation is done at ambient temperature.

Video Sequence 2: Movement of an OCC on the Surface of the Oviduct

Cilia on the outer surface of the infundibulum normally function in picking up the OCC after ovulation and transferring it rapidly into the lumen of the oviduct (Figure 2B). A methylene blue–stained OCC has been placed on a hamster infundibulum and viewed with a stereoscopic microscope. Cells of the OCC are stained blue and are separated by extracellular matrix, which is unstained. Pickup of the OCC by the oviduct occurs in two phases. First the OCC attaches to the tips of the cilia and glides over the surface of the infundibulum to the ostium at a rate of 50–60 μm/s at ambient temperature (Huang *et al.*, 1997). The video shows this event in approximately real time. Then the OCC enters the ostium and undergoes churning activity as it becomes compacted to a size that can be accommodated by the lumen of the infundibulum. The video shows entrance of the OCC into the ostium approximately five times faster than real time. When positioned at the same starting place on an infundibulum, OCCs will repeatedly travel over the same path on the surface each time (Huang *et al.*, 1997).

Only a small fraction of the OCC surface touches the cilia at any one time. Because the matrix is elastic, it stretches out in front and behind the main body of the OCC, thereby increasing the area of contact and improving adhesion. The extension of cells and matrix at the leading edge of the OCC is characteristic of normal pickup, and the extension is easily able to penetrate into the narrow opening of the ostium. The cells and matrix trailing the OCC remain attached to cilia as the main body of the OCC moves away from them toward the ostium. Eventually the trailing matrix snaps either

[V] Online version of this essay contains video material for Figure 2. Online version available at www.molbiolcell.org.

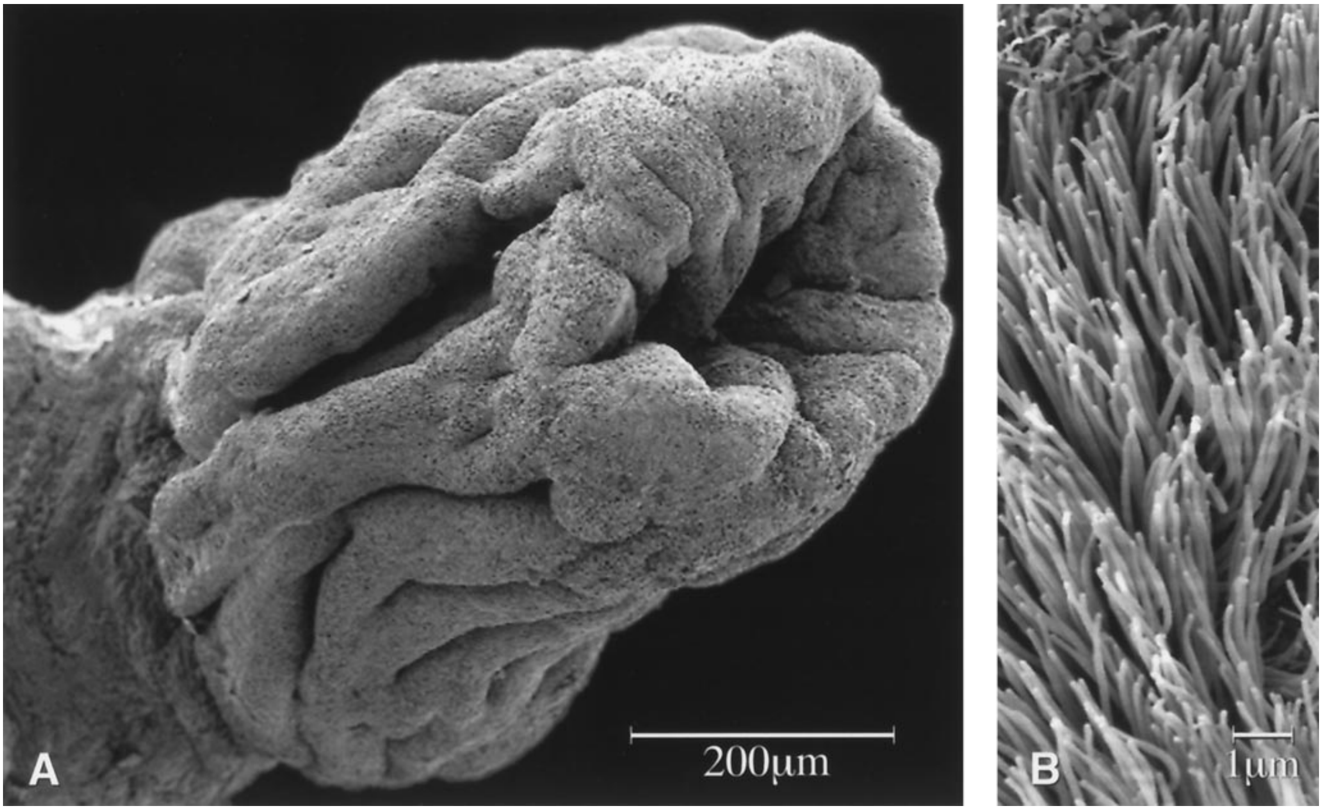

Figure 1. Scanning electron micrograph (A) of a hamster infundibulum and a higher-magnification view of its surface showing oviductal cilia (B). The ostium of the infundibulum is partially opened.

because it releases from the cilia or because it tears. Tearing is known to occur as scanning electron micrographs have revealed matrix trails left by OCCs on infundibular surfaces. Movement of the matrix extending from the leading edge into the ostium is important in anchoring the OCC to the oviduct as the main body of the OCC makes a 180° turn and enters the ostium. The main body of the OCC is usually too large to move directly into the lumen of the infundibulum, and the OCC churns while the cilia inside the infundibulum pull on the its matrix and compact the OCC into a smaller size that can pass through the ostium.

Video Sequence 3: Movement of an OCC into the Ostium

This camera view looks down the oviduct and presents a bird's eye view of an OCC entering the ostium (Figure 2C). This process takes longer than movement of the OCC over the surface of the oviduct and is therefore shown approximately six times faster than real time. The OCC is large relative to the size of the ostium, which often is completely closed as entry begins. At the beginning of this sequence, several cells and the matrix at the leading edge enter the right side of the ostium. As cilia pull the matrix, the OCC churns until it becomes spherical and sufficiently compacted to enter the ostium completely. The oocyte, which is located in the center of the OCC at the beginning of this sequence, is squeezed to the periphery during churning and enters the ostium last. This explains why OCCs recovered from the ampulla of the oviduct often have eccentrically located oocytes (Corselli and Talbot, 1987). Repositioning the oocyte to the periphery of the OCC potentially shortens the distance a sperm must pass through the matrix before reaching the zona pellucida of the oocyte. As a result of churning, the OCC has a smaller diameter, and the matrix is compacted and less elastic.

Video Sequence 4: Entrance into the Ostium

This video was made at higher magnification to better show the interface between cells of the OCC and the surface of the infundibulum as the OCC is entering the ostium (Figure 2D). Because this infundibulum has already picked up an OCC, the ostium is partially open and easier to visualize. Cumulus cells, but not the matrix between them, are stained with methylene

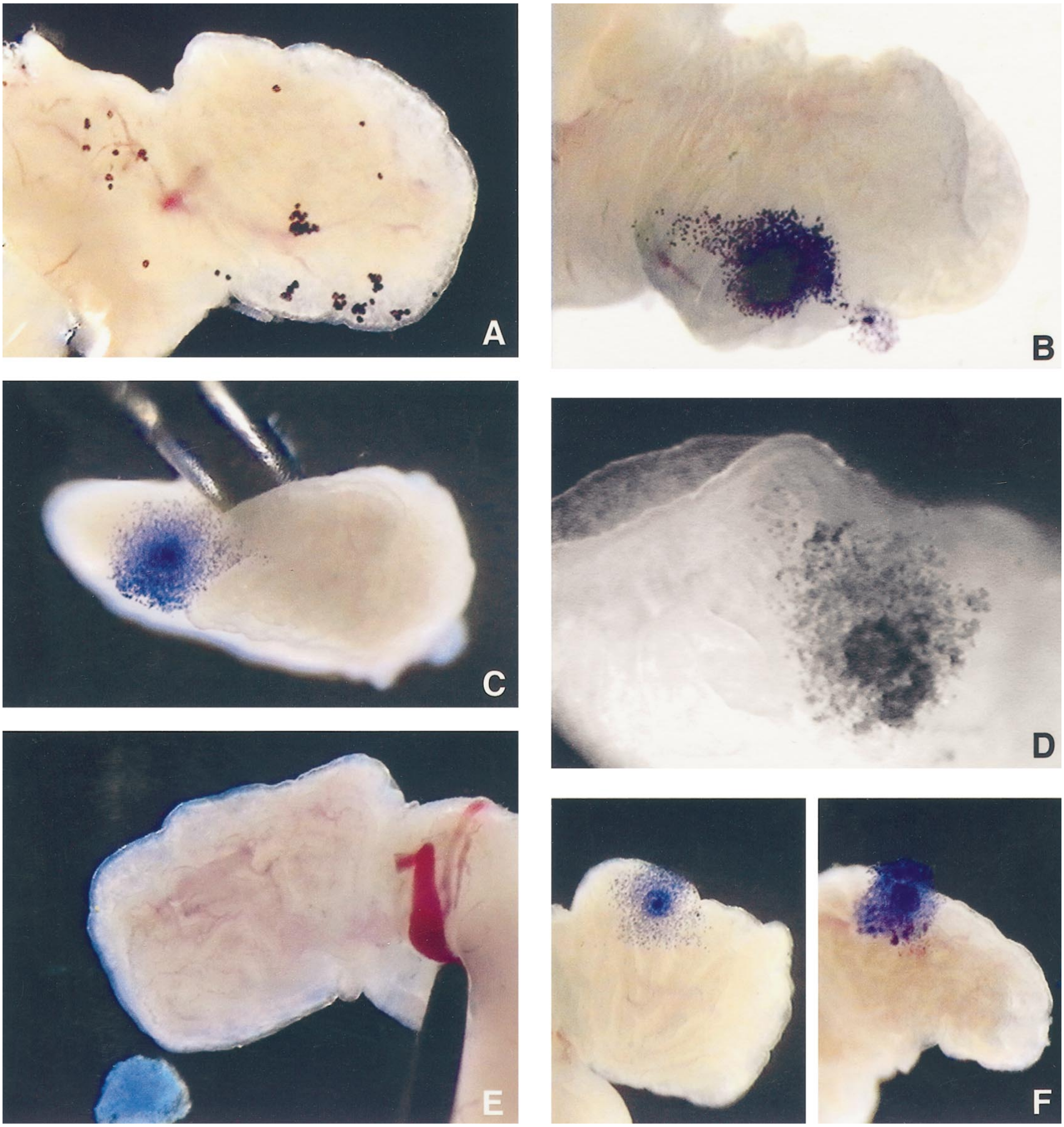

Figure 2. Movies showing movement of *Lycopodium* spores and OCCs on the surface of hamster infundibula. (A) *Lycopodium* spores being picked up by currents above hamster oviductal cilia. (B) Stained OCC traveling over the surface of a hamster infundibulum. (C) Bird's eye view of the ostium showing entry of a stained OCC. (D) Higher-magnification video showing movement of an OCC into the ostium. (E) Video showing pickup of an OCC that has been recovered from inside an infundibulum. The matrix of the OCC has been compacted and is no longer elastic. (F) Pair of movies showing the effect of cigarette smoke on OCC pickup. The infundibulum on the left has been exposed to sidestream smoke solution, whereas the one on the right was incubated in control medium only.

blue. Cumulus cells glide over the surface of the cilia. Cells and matrix at the leading edge move out ahead of the main body of the OCC and enter the ostium first. As the leading cells and matrix penetrate deeper into the lumen of the infundibulum, adhesion between the main body of the OCC and cilia on the surface is disrupted, and the main body snaps forward and is drawn down into the infundibulum. Adhesion between the cilia and matrix of the OCC ensures that the OCC successfully makes a 180° turn as it enters the ostium without falling off the oviduct.

Video Sequence 5: Effect of Insufficient Adhesion on OCC Pickup

This sequence shows pickup of an OCC that has been recovered from the inside of an infundibulum (Figure 2E). Because the OCC has been compacted, it is smaller in diameter, its cells are difficult to resolve, and its matrix is less elastic. The matrix is still somewhat adhesive and can attach to the cilia well enough to allow the OCC to be pulled along an edge of the infundibulum. However, the matrix is not elastic enough to enable an extension of matrix to form at the leading edge of the OCC. As a result, the OCC is not adhering well to the cilia when it approaches the ostium, and rather than make a 180° angle turn to enter the ostium, the OCC falls off the oviduct. This sequence illustrates that matrix elasticity is necessary for formation of an adhesive extension at the leading edge of the OCC and that this extension is important for turning into the entrance of the oviduct.

Video Sequence 6: Effect of Cigarette Smoke on OCC Pickup

Cigarette smoke causes the oviductal ciliary to beat frequency and oocyte pickup rate to decrease (Knoll *et al.*, 1995; Knoll and Talbot, 1998). When smoke-treated infundibula are placed in fresh culture media, the ciliary beat frequency recovers, but unexpectedly, the OCC pickup rate does not recover and may continue to decrease (Knoll and Talbot, 1998). This decrease in pickup rate appears to be related to a change in the adhesive interaction between the cilia and OCC matrix. The video sequence on the right shows a control infundibulum and OCC moving at normal speed during a 10-s interval. The sequence on the left shows an infundibulum pretreated with sidestream smoke solution. Although the ciliary beat frequency has recovered from the smoke treatment, the OCC pickup rate continues to be depressed, and in this 10-s sequence, the OCC on the treated infundibulum moves very little. The OCC on the treated infundibulum eventually moved to the ostium, but it took >3.5 min. After longer exposures to smoke solutions, OCCs are not picked up at all, despite the fact that cilia beat at normal frequency (Knoll and Talbot, 1998).

SUMMARY

The transportation of mammalian oocytes into the oviduct is governed by ciliary beating and proper adhesion of the OCC matrix to the tips of the infundibular cilia. Ciliary beating alone can transport small particles such as spores but is not sufficient for OCC pickup. Proper adhesion is particularly important for entry of the OCC into the ostium, during which time the OCC matrix is compressed and the oocyte becomes eccentrically positioned. Chemicals in cigarette smoke affect both ciliary beat frequency and adhesion, and the effects on adhesion are not readily reversible.

ACKNOWLEDGMENTS

We thank Mark Radleigh, Sohail Wasif, and Ray Talbot for their help in preparing the manuscript. This work was supported by California Tobacco-Related Disease Research Program grant 6RT-0039.

REFERENCES

Corselli, J., and Talbot, P. (1987). In vitro penetration of hamster oocyte-cumulus complexes using physiological numbers of sperm. Dev. Biol. *122*, 227–242.

DiCarlantonio, G., Shaoulian, R., Knoll, M., Magers, T., and Talbot, P. (1995). Analysis of ciliary beat frequencies in hamster oviductal explants. J. Exp. Zool. *272*, 142–152.

Harper, J.K. (1994). Gamete and zygote transport. In: The Physiology of Reproduction, ed. E. Knobil and J.D. Neill, New York: Raven Press, 123–185.

Huang, S., Driessen, N., Knoll, M., and Talbot, P. (1997). In vitro analysis of oocyte cumulus complex pick-up rate in the hamster *Mesocricetus auratus*. Mol. Reprod. Dev. *47*, 312–322.

Hunter, R.H.F. (1994). Modulation of gamete and embryonic microenvironments by oviduct glycoproteins. Mol. Reprod. Dev. *39*, 176–181.

Knoll, M., Shaoulian, R., Magers, T., and Talbot, P. (1995). Ciliary beat frequency of hamster oviducts is decreased in vitro by exposure to solutions of mainstream and sidestream cigarette smoke. Biol. Reprod. *53*, 29–37.

Knoll, M., and Talbot, P. (1998). Cigarette smoke inhibits oocyte cumulus complex pick-up by the oviduct independent of ciliary beat frequency. Reprod. Toxicol. *12*, 57–68.

Mahi-Brown, C.A., and Yanagimachi, R. (1983). Parameters influencing ovum pickup by oviductal fimbria in the golden hamster. Gamete Res. *8*, 1–10.

Norwood, J.T., Hein, C.E., Halbert, S.A., and Anderson, R.G.W. (1978). Polycationic macromolecules inhibit cilia-mediated ovum transport. Proc. Natl. Acad. Sci. USA *75*, 4413–4416.

Talbot, P., and DiCarlantonio, G. (1984). The oocyte-cumulus complex: ultrastructure of the extracellular components in hamsters and mice. Gamete Res. *10*, 127–142.