Surface morphology of oviductal epithelial cells in stages of

follicular and luteal phases of buffalo

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Abstract

In this study, we examined the luminal surfaces of epithelial cells that lined on the ampulla-

isthmus of the buffalo oviduct during the follicular and luteal phases of the estrous cycle by using

scanning electron microscopy. The luminal epithelium of ampulla-isthmus from the oviduct during

the follicular phase showed dense population of ciliated cells. The cilia of ciliated cells partially

covered the apical surface of nonciliated cells. However, only one-third of epithelial cells lining the

ampulla during luteal phase were ciliated cells. The apical surfaces of the nonciliated cells were

round shape. It is obvious that, during the estrous cycle, the buffalo oviductal epithelium changes

its structural features. It is known that the buffalo is commercial livestock in Thailand. We believed

that examination of the epithelial cell of buffalo oviduct might contribute to increasing our knowledge

of buffalo reproductive.

Key words: oviduct, follicular phases, luteal phases, scanning electron microscopy

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การศึกษาเซลล์บุผิวท่อนำไข่ของกระบือในระยะฟอลลิคูลาร์และ ระยะลูเทียล โดยใช้กล้องจุลทรรศน์อิเล็กตรอนชนิดส่องกราด

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**สถาบันพัฒนาการท่องเที่ยวเพื่ออนุรักษ์สิ่งแวดล้อม มหาวิทยาลัยศรีนครินทรวิโรฒ

บทคัดย่อ

จากการศึกษาเซลล์บุผิวของท่อนำไข่ของกระบือในส่วนแอมพูล่าในระยะฟอลลิคูลาร์ พบเซลล์ ที่มีชีเลีย(ciliated cell) อยู่อย่างหนาแน่น โดยมีเซลล์ที่ไม่มีชีเลีย (non ciliated cell) แทรกอยู่เพียงเล็กน้อย ในขณะที่ในระยะลูเทียล พบเซลล์ที่มีชีเลียจำนวนน้อย แต่พบเซลล์ที่ไม่มีชีเลียเป็นส่วนใหญ่การ เปลี่ยนแปลงจำนวนของเซลล์บุผิวของท่อนำไข่ทั้งสองระยะนี้สอดคล้องกับหน้าที่ของเซลล์ในแต่ละช่วงของ รอบสืบพันธุ์ ระยะฟอลลิคูลาร์อยู่ภายใต้อิทธิพลของฮอร์โมนเอลโตรเจน จะมีผลกระตุ้นการเจริญของ ซีเลีย ซึ่งช่วยในการพัดโบกทำให้มีการเคลื่อนที่ของเซลล์ไข่ที่สุกแล้วเดินทางในท่อนำไข่ได้สะดวกขึ้น ใน ขณะที่ระยะลูเทียลอยู่ภายใต้อิทธิพลของฮอร์โมนโปรเจสเตอโรนจะยับยั้งการเจริญของซีเลีย เนื่องจาก ไม่มีการตกไข่ในระยะนี้ จึงไม่จำเป็นที่จะต้องมีซีเลีย การศึกษาครั้งนี้ยืนยันลักษณะโครงสร้างของเซลล์ บุผิวท่อนำไข่ของกระบือสอดคล้องกับหน้าที่ในแต่ละช่วงของรอบสืบพันธุ์ และเนื่องจากกระบือจัดเป็น สัตว์เศรษฐกิจที่สำคัญของประเทศไทย ความรู้พื้นฐานนี้จึงเป็นข้อมูลที่สำคัญอันหนึ่งที่จะใช้ในการศึกษา ด้านต่างๆ ของระบบสืบพันธุ์ของกระบือต่อไป

คำสำคัญ: ท่อนำไข่ ระยะฟอลลิคูลาร์ ระยะลูเทียล จุลทรรศน์อิเล็กตรอนแบบส่องกราด

Introduction

The mammalian oviduct is important for the transportation of the ovulated oocyte, sperm and developing embryos. It maintains and modulates oviductal fluid which is a selective transudate of serum. Oviductal epithelium provides the necessary environment for the final maturation process of male gamete, fertilization and embryonic development.

The oviductal epithelium consists of two cell types: ciliated and nonciliated cells. The nonciliated or secretory cells synthesize and release glycoproteins which are dissolved in the oviductal fluid.² In some mammal, the glycoproteins secreted by oviductal nonciliated cells have been shown to associate with ovulated oocyte and developing embryo. 3,4 The oviductal secretion may regulate or may be involved in various physiological functions. Furthermore, it has been reported that the development of embryos in vitro is enhanced by co-culturing with oviductal epithelial cells or with their conditioned medium.⁵ Pavasuthipaisit and colleagues⁴ reported that porcine oviductal cells produced a soluble component which enhanced embryonic development to the blastocyst stage in vitro and its effect was not species-specific. Their findings suggest that the oviductal secretory cells and their secretion play important roles on reproductive and developmental events occurred in the oviduct.7

In the present study, the luminal surfaces of ampullar-isthmic junction of the buffalo oviduct during the follicular and luteal phases were observed by means of scanning electron microscopy in order to demonstrate their morphological changes during the estrous cycle. Furthermore, the oviductal epithelium provides the necessary environment for the maturation process of sperm, fertilization and embryonic development. The results from this study are basic knowledge for understanding the reproduction of Thai buffalo which is at risk of extinction.

Materials and methods

Ten buffalo ovaries were collected after slaughter and classified for the phase of estrous cycle. The oviducts at the luteal and follicular phases were removed and placed in phosphatebuffered saline, pH 7.4. The ampullar-isthmic area of oviducts was excised and fixed by immersing in 2.5% glutaraldehyde for 3 h. The tissue was cut in to pieces so that the luminal surface of each piece was approximately 3 mm in diameter before being postfixed with 1% osmium tetroxide at 4°C for 1 h. The tissues were dehydrated through graded series of ethanol (50-100%). The pieces of oviduct were placed in the small stainless-steel mesh basket of a critical point drying (CPD) apparatus. The chamber was sealed and flooded with liquid CO₂. After finishing the process of CPD, the



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pieces of tissue were mounted on stubs with carbon conducting tape, coated with gold at 20 nm thick in an ion sputtering and examined with a scanning electron microscopy operating at 15 kV.

Results

Follicular Phase

The ampullar-ishmic junction of the buffalo oviducts during the follicular phase appeared to have extensive ciliation (Figure 1A). The cilia were consistently protrusion over the apex of the nonciliated cells which were covered from being visible to some extent. The apical surfaces of the nonciliated cells were spherical in shape (Figure 1B). Numerous short microvilli were protrusion from the apical surfaces of the nonciliated cells.

Luteal Phase

During the luteal phase, approximately one-third of the epithelial cells in the ampullar-isthmic junction were decreased. The cilia protruded uniformly into the lumen. The apical surfaces of the nonciliated cell were round shape and the microvilli were decreased when compared to those in the follicular phase. The nonciliated cells were numerous and morphologically distinct from the ciliated cell in this region of the oviduct (Figures 2A, B).

Discussion

This research demonstrates that the buffalo's oviduct undergoes alteration in the numbers of ciliated and nonciliated cells during the estrous cycle. The results were only derived from the follicular and luteal phases, but not the entire estrous cycle. The ciliated cells in ampulla-isthmus were predominant during the follicular phase, and they are cells decreased during the luteal phase. Similar cyclic changes have been examined in the Chinese Meishan pig.6 In the ampulla, the numbers of ciliated cells on the surface of the bovine oviductal epithelium increased during the follicular phase and decreased during the luteal phase. In the ampullar region during follicular phase, the number and the length of cilia are responsible for picking up and transportation the ovulated oocytes.^{3,4}Therefore, numerous ciliated epithelium at ovulation must be attained. During the luteal phase, the nonciliated cells were more abundant than the ciliated cells and were generally spherical in shape.^{3,4} Nonciliated cells secrete the secretion to support the embryonic development. The contact between these cells and the sperm has been reported to play an important role in maintaining sperm viability in rabbit.⁸ In sheep, sperm are observed on the apical plasma membrane of the oviductal

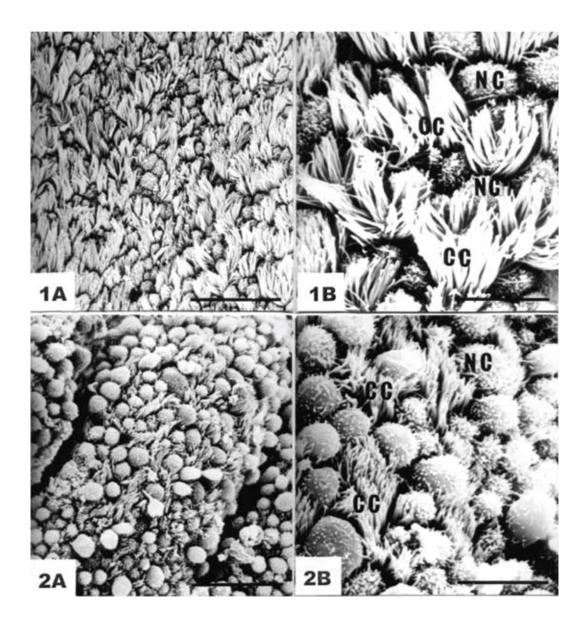


Figure 1A Scanning electron micrograph in oviduct showing the high number of ciliated cells during follicular phase Bar = $30 \ \mu m$

- Figure 1B A high magnification of scanning the figure 1A showing, the protrusion of the cilia of ciliated cells (CC) over the nonciliated cells in oviduct during the follicular phase (NC). Bar = $10~\mu m$
- Figure 2A Scanning electron micrograph showing the lower number of ciliated cells than but a lot of nonciliated cells in oviduct during the luteal phase Bar = $30 \mu m$
- Figure 2B A high magnification of the figure 2A in oviduct showing numerous nonciliated cells during the luteal phase. Bar = $10~\mu m$



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epithelial cells.⁹ Attachment to oviductal epithelial cells has a beneficial effect on the maintenance of sperm viability during storage in the oviduct, but its mechanism is unknown.¹⁰

The present study reveals that there are marked differences in epithelial cells of the buffalo's oviduct between the follicular phase and the luteal phase. Many studies have demonstrated that the cytodifferentation of epithelial cells of the oviduct affected by ovarian steroids. The regular cycle of ciliogenesis and deciliation of the epithelial cells in the mammalian oviduct is depended on the levels of circulating estrogen and progesterone. The ciliogenesis in the follicular phase occurs under the influence of estrogen, while suppression of the ciliogenesis leading to deciliation at midcycle is under the influence of progesterone.

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