Original Article

E-cadherin localization in oviduct and uterine horn of swamp buffalo during estrous cycle

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Abstract

The main purpose of the present study was to investigate the localization of epithelial cadherin (E-cadherin) in the oviduct and uterine horn of swamp buffalo at the follicular and mid-luteal phases. E-cadherin is a transmembrane glycoprotein which plays a vital role in the mechanism of the maintenance epithelial architecture, cell growth and proliferation, cell to cell adhesion including cell to epithelium adhesion. Female reproductive tracts of swamp buffaloes (n=30) were collected from local abattoirs and categorized into follicular (n=15) and mid-luteal (n=15) phases. Tissue samples from the oviducts and uterine horns were regularly handled according to histological and immunohistochemical techniques. The results established that E-cadherin localization was detected in the surface epithelium of uterotubal-junction (UTJ) and isthmus of the oviducts and strong intensity was significantly observed ($P<0.05$) in the follicular phase compared to the mid-luteal phase. In the uterine horns, E-cadherin was found in the endometrial and glandular epithelium of superficial and deep endometrial glands during both phases of the estrous cycle. However, difference in the immunostaining intensity in the uterine tissues was not found between estrous phases. The current findings indicate that the existence of E-cadherin in the UTJ and isthmus, called sperm reservoir, might function with other molecular substances that appear in this region to regulate the adhesion and release of spermatozoa. In addition, E-cadherin in the endometrial and glandular epithelium is associated with the major preparation of the maintenance of epithelial cell architecture including cell growth and proliferation in uterine horn before blastocyst hatching during implantation period.

Keywords: E-cadherin, estrous cycle, sperm reservoir, oviduct, uterus, swamp buffalo

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Introduction

At present, the population of swamp buffalo recorded in Thailand is rapidly diminished by numerous reasons, especially the internal factors of this animal which are composed of delayed puberty, deprived estrus detection, lengthy calving interval, and also reproductive failures, e.g. early embryonic death and reduced conception rates (Singh et al., 2000). These causes have been the main boundaries for the improvement in swamp buffalo productivity. Consequently, necessary understanding with declaration of the molecular biology occurring in the female reproductive organs, especially in both oviduct and uterine horn, must be scrutinized to raise this animal production. In cattle, the function of female reproductive organs involves the complicated phenomenon which is controlled by the hormonal system throughout the estrous cycle, pregnancy and parturition (Thatcher, 2017). After insemination, in cattle, the oviduct plays an important role before fertilization, i.e. storing the spermatozoa in the site called sperm reservoir, conveying the oocytes from the ovaries via the infundibulum to the ampulla-isthmic junction (AIJ) where spermatozoa and oocytes encounter at fertilization (Holt and Fazeli, 2016). The uterotubal junction (UTJ) and caudal isthmus of the oviducts in pig and cattle have been confirmed to be the sperm reservoir, where the spermatozoa are stored, sperm viability is maintained, sperm capacitation is initiated and the exact amount of spermatozoa is released to the fertilization place (Hunter, 2012; Tienthai, 2015). After the achievement of fertilization, the uterine horns serve as the implantation site of blastocysts and continue to the formation of placenta in domestic animals (Imakawa et al., 2017; Sadam et al., 2017). Although the glycosaminoglycans, i.e. hyaluronan and syndecan-1, have recently appeared in the isthmus and UTJ of swamp buffalo oviducts (Tienthai, 2011), profound researches correlating with the glycoproteins serving as cell adhesion molecules in the oviducts and uterine horns of swamp buffalo are scarcely performed. Consequently, precise modulation during sperm-epithelial adhesion in the sperm reservoir or regulations of the endometrial epithelium during embryo implantation in the uterine horn of swamp buffalo is still obscured and requires supplementary information.

Among the members from superfamily of cell adhesion molecules, cadherins are known as the family of transmembrane glycoproteins associated with calcium-dependent cell to cell adhesion and they are documented according to their tissue of derivation which is composed of three types, i.e. the neural (N-), placental (P-), and epithelial (E-) cadherins (Yoshida-Noro, 1984; Nose and Takeichi, 1986; Takeichi, 1991). These glycoproteins demonstrate critical roles during embryonic development, cell growth, cell differentiation, and also adult tissue architecture maintenance (Gumbiner, 1996). Interestingly, E-cadherin was discovered as the first cadherin which functions in the facilitation of selective adhesion between epithelial cells and is involved in the preliminary attachment of early embryo to the endometrial epithelium (Dawood et al., 1998; Imakawa et al., 2017). Moreover, E-cadherin was distinguished in the oocytes, spermatozoa, oviduct epithelium and uterine epithelium of human (Tsuchiya et al., 2006) and cow (Caballero et al., 2014). In cow, the creation of sperm reservoir is associated with the sperm-epithelial binding and protein recognition depending on the stages of estrous cycle (Suarez, 2002; Suarez and Pacey, 2006; Hunter, 2012). These earlier investigations implied the interaction of gametes and the adhesion between gametes and female reproductive epithelium involved in the cell adhesion molecules, i.e. cadherins. Despite the large amount of researches performed in human and other domestic animals, no study report has scrutinized E-cadherin in the female reproductive tissues of swamp buffalo.

To better understand the principal role functioned by this molecule during the activities of gametes during estrous cycle, this study was designed to investigate the localization of E-cadherin in the oviducts and uterine horns of swamp buffalo at the follicular and mid-luteal phases by immunohistochemical techniques.

Materials and Methods

Tissue samples: Female reproductive tracts of swamp buffaloes (n=30) at various ages (2-5 years) were obtained from local abattoirs after slaughter, kept in an ice container at temperature about 4-5°C and transported to laboratory room for macroscopic examination within a few hours. The chosen stages of the estrous cycle were composed of the follicular (n=15) and mid-luteal phases (n=15), sorted out by the appearance of corpus lutea and predominant follicles on the ovaries (Chandra Roy et al., 2006). The entire oviducts were dissected out of the mesosalpinx and divided into two main segments consisting of UTJ and isthmus, whereas the uterine horns were separated from the mesometrium and the middle part of uterine horn, about 2 centimeters, was excised. All tissue samples from the oviducts and uterine horns were submerged in 10% buffered formalin as routinely prepared by histological procedures and accomplished by the use of immunohistochemistry.

Immunohistochemical detection of E-cadherin: After preservation in 10% buffered formalin for 48 hours, the oviduct and uterine tissues were handled by an automatic tissue processor (Lieca TP1020, Lieca Biosystems, Nussloch, Germany), embedded in a paraffin block and then cut into 4-µm-thick sections by the use of a rotary microtome (JUNG BIOCUT 2035, Lieca Instruments GmbH, Nussloch, Germany). The tissue sections were put on Poly-L-Lysine coated glass slides (SIGMA-ALDRICH Inc., Steinheim, Germany), heated in an incubator at 40°C for 4 hours, deparaffinized in xylene and rehydrated via graded ethanol dilutions. Afterwards, the tissue sections were immersed in 0.01 M citrate buffer (pH 6.0) in a microwave oven at 750 W about 10 minutes to retrieve antigenicity. Endogenous peroxidase activity was inhibited by submerging the tissue section in 3.0% H_2O_2 at room temperature for 20 min and non-specific background staining was diminished by incubation with normal horse serum (Vector Laboratories Inc.,

Burlingame, CA, USA). Mouse monoclonal antibody to E-cadherin (clone NCH-38, DAKO, Grostrup, Denmark) at a dilution of 1:50 was operated as primary antibody and the incubation time for the primary antibody was exactly 18 hours at 4°C. Subsequently, the sections were incubated with secondary biotinylated horse anti-mouse antibody (Vector Laboratories Inc., Burlingame) at a dilution of 1:200, followed by Avidin-Biotin Complex (ABC)-mouse reagent (Vector Laboratories Inc., Burlingame). Positive immunoreactions were envisioned using the 3, 3'-diaminobenzidine (DAB kit, Vector laboratories Inc. Burlingame) in H_2O_2 and all slides were counterstained with Mayer's hematoxylin, mounted with gelatin-glycerine mixture, and put on the sections with cover-slips. Slide tissues of mammary gland adenocarcinoma in bitch were accomplished as the positive controls (Sarli et al., 2004; Nowak et al., 2007), whereas the negative controls were completed by substituting the primary antibody with normal mouse IgG (sc-2025; Santa Cruz Biotechnology Inc., CA, USA) in the oviduct and uterine tissues as well as the mammary gland adenocarcinoma.

Categorization of positive immunohistochemistry: The oviduct and uterine slide sections were analyzed under light microscopy (BX50, Olympus, Tokyo, Japan) with a digital camera Micropublisher 5.0 (Qimage, Surrey, Canada). All section micrographs were performed using the program of Image Pro® Plus version 6 (Media Cybernatics Inc., MD, USA). In oviduct, the surface epithelium and subepithelial connective tissue (CNT) layer were investigated, whereas the uterine epithelium, superficial and deep endometrial glands as well as subepithelial CNT layer in the uterine horns were examined. Manual recording of E-cadherin positive immunostaining reaction was performed by the same person who was ignorant of the originality of animals compared to brown staining in the positive controls. The positive immunohistochemical staining intensity was elucidated into three different scores: weak = 1, moderate = 2 or strong = 3, as formerly performed by Tienthai et al. (2009).

Statistical analyzes: All observed data were statistically scrutinized using Statistical Analysis System software (SAS Institute Inc., Cary, NC, USA). Mean and standard deviation were calculated and used to demonstrate data. The intensity scores obtained from the oviduct and uterine horn at the follicular and mid-luteal phases were compared using Wilcoxon Scores test and Kruskal-Wallis test (NPAR1WAY procedure of SAS). A value of $P<0.05$ was considered statistically significant.

Results

Immunohistochemical reaction of E-cadherin in control tissues: As anticipated, the strong intensity of E-cadherin immunohistochemical staining was noticed in the canine mammary gland carcinoma, which functioned as the positive controls (Figs. 1a, b), whereas the immunostaining was not found on the negative slide controls using uterine tissues with normal mouse IgG instead of primary antibody (Figs. 2c, d). The configuration of E-cadherin immunostaining was apparently seen as dark brown staining on cell-cell boundaries of the epithelial luminal cells of ducts, called membranous staining, within the mammary gland carcinoma (Fig. 1b).

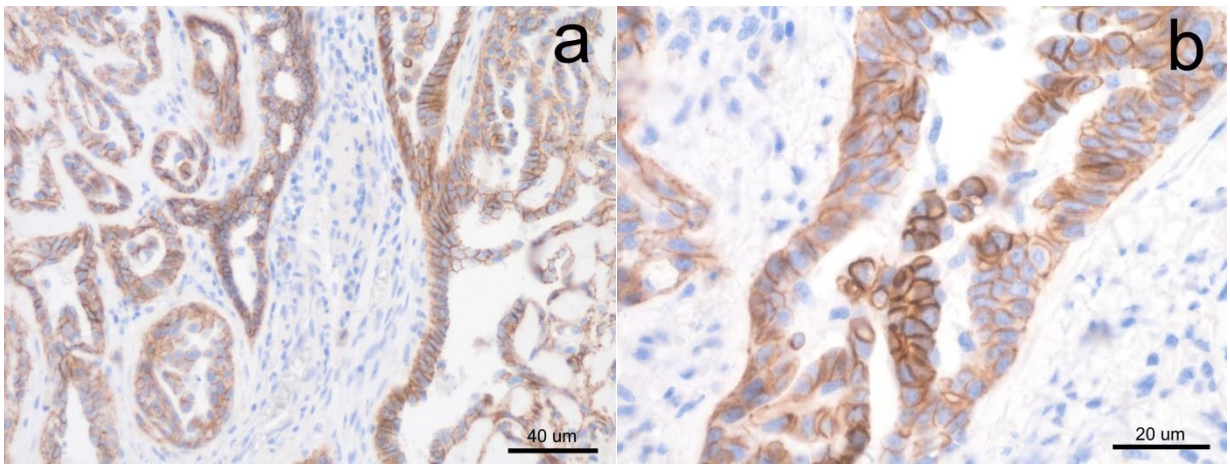

Figure 1 Intense immunohistochemical detection of E-cadherin (dark brown staining) at the epithelial cells of ducts within the mammary gland carcinoma in a bitch (positive control) at different magnifications (a, b) that predominantly demonstrate the membranous E-cadherin staining pattern (b).

Immunohistochemical reaction of E-cadherin in oviducts: Remarkably, the positive localization of E-cadherin was detected only in the epithelial surface of UTJ and isthmus in the swamp buffaloes during both selected estrous phases and the immunoreaction varied by location and intensity among individual sample (Figs. 3a-d). Strong staining was conspicuously found in site of the epithelial surface of the UTJ (Fig. 3a) and isthmus (Fig. 3c) at the follicular phase compared to the same portions at the mid-luteal phase ($P<0.05$) as depicted in Figure 4. At higher magnification, the brown immunostaining of E-cadherin appeared in the cell membrane and in the supranuclear region of dispersed epithelial cells, which is usually assumed as the secretory-like cells (Figs. 5a, b). Moreover, weak staining was rarely observed in the cytoplasm of the epithelial cells of UTJ and isthmus (Figs. 5a, b).

Immunohistochemical reaction of E-cadherin in uterine horns: The E-cadherin expression demonstrated positive staining in the uterine epithelium including the epithelial linings of superficial and deep endometrial glands (Figs. 6a-d) of swamp buffalo uterine horns. Moderate to strong immunoreaction was intermittently observed throughout the length of uterine epithelium and also in the epithelial lining of superficial and deep endometrial glands. However, the intense immunohistochemical staining found in the uterine surface and epithelial lining of the endometrial gland was not significantly different between the follicular and mid-luteal phases (Fig. 7). Considering the intensity region of the uterine epithelium, the pattern of strong membranous E-cadherin immunoreaction was sporadically noticed, whereas the weak staining was frequently observed in the cytoplasm of uterine epithelial cells (Figs. 8a, b).

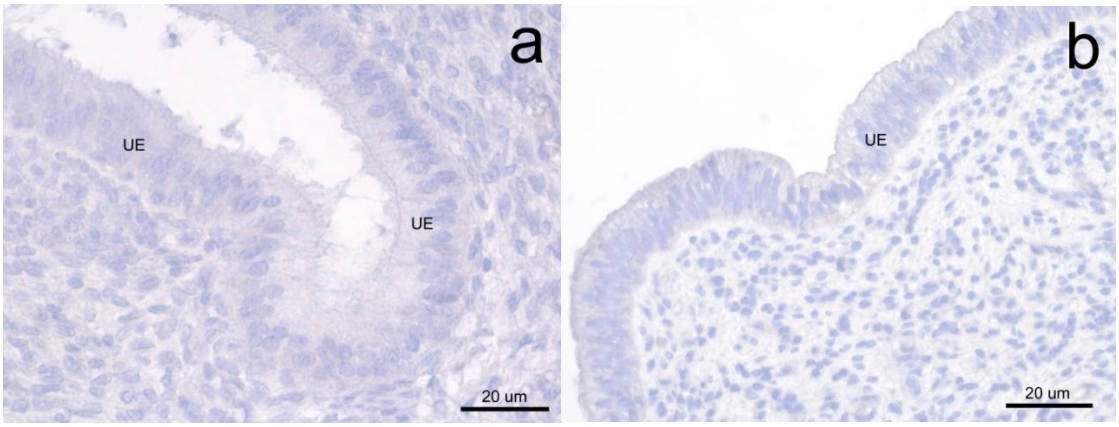

Figure 2 No immunohistochemical detection of E-cadherin in the epithelial cells of uterine epithelial surface (UE) and other cells in the subepithelial connective tissues of the buffalo uterine tissue sections by the use of normal mouse IgG by which primary antibody was replaced (negative control).

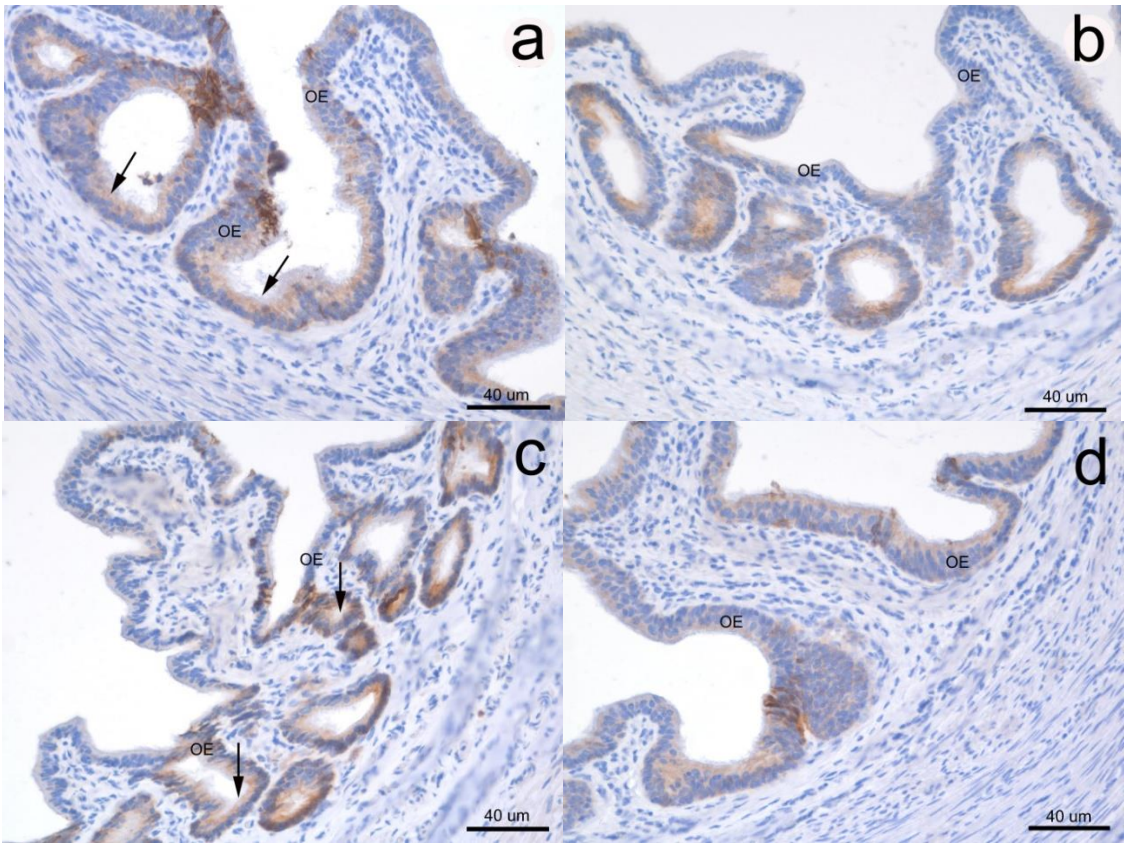

Figure 3 Immunohistochemical localization of E-cadherin in the uterotubal junction (a, b), and isthmus (c, d) of swamp buffalo oviducts at follicular (a, c) and mid-luteal (b, d) phases. Moderate to intense E-cadherin detection was found at the lateral and apical cell membranes of the oviduct epithelium (OE) and positive immunoreactivity was sporadically depicted throughout the epithelial lining. Notice the epithelial crypts of UTJ and isthmus presented in both UTJ and isthmus (black arrows).

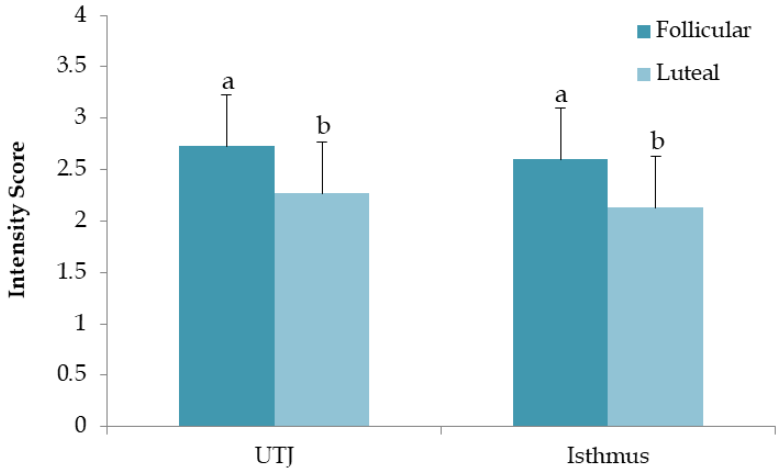

Figure 4 Intensity scores of E-cadherin immunostaining in the surface epithelium of uterotubal junction and isthmus of the swamp buffalo oviducts at follicular and mid-luteal phases. Values are presented as mean ± SD. Different letters between bars show significant difference (P<0.05).

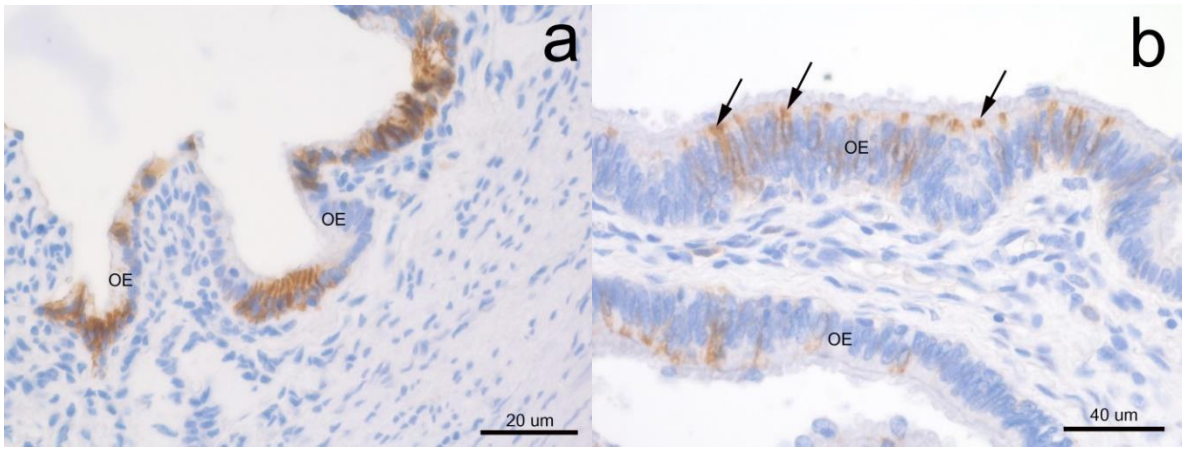

Figure 5 Immunohistochemical localization of E-cadherin in the oviduct epithelium (OE) of UTJ (a) and isthmus (b) of swamp buffalo oviduct during follicular phase at higher magnification demonstrating the lateral and apical cell membrane immunoreactivity. Notice the supranuclear domains in the oviduct epithelial cells establishing immunostaining (black arrows in Figure 4a).

Discussion

This report describes, for the first time, a detailed investigation into the immunohistochemical localization of E-cadherin in the female reproductive organs, at least in the oviduct and uterine horn, of swamp buffalo. Furthermore, the findings provide the confirmation that E-cadherin might cooperate with other substances to modulate the interaction of gametes or blastocysts with oviduct or uterine epithelium during the estrous cycle. In the positive control (mammary gland carcinoma), the appearance of E-cadherin was characterized by brown immunostaining on cell-cell borders of the epithelial luminal cells of ducts, corresponding to previous studies (Sarli et al., 2004; Nowak et al., 2007), indicating the satisfactory efficiency of the monoclonal antibody and the detection procedures in the present study.

In farm animals, it is becoming progressively clear that the oviducts play an essential role in the complex evidences, i.e. the transportation of oocyte and spermatozoa and the development of embryo, which signify the accomplishment of fertilization (Suarez, 2002; Brussow et al., 2008; Hunter, 2012). The UTJ and caudal isthmus are segments of the oviduct which serve as sperm reservoir where a number of functions are performed, e.g. the conservation of sperm fertilizing ability to increase the survival time during sperm storage until ovulation, the control process of sperm capacitation and release to the fertilization site (Pollard et al., 1991; Hunter, 2012; Tienthai, 2015). The regulations for creation of the sperm reservoir are composed of various factors, but the most important factor is the sperm-epithelium binding with cell adhesion molecules in the selected period of estrous cycle (Tienthai, 2011; Tienthai, 2015). Cadherins are a family of surface glycoprotein cell adhesion molecules that establish the capability of binding with high specificity to other cadherins on neighboring cells (Marrs and Nelson, 1996). Among the superfamily of cadherins (E-, N- and P-cadherins), it has been suggested that E-cadherin is a huge group that mediates calcium-dependent intercellular adhesion and is sheltered from proteolysis through stabilization by calcium ion without undertaking conformational changes (Takeichi, 1995). E-cadherin immunostaining in the UTJ and isthmus of swamp buffalo was observed at the basolateral and apical domains of epithelial cells during both follicular and mid-luteal phases as previously reported in bovine (Caballero et al., 2014).

Although the location and function of sperm reservoir have not been clearly investigated in swamp buffalo, the UTJ and caudal isthmus have been confirmed as the site of sperm reservoir in cow (Suarez, 2002) and the modulation of sperm binding oviduct explants has been performed in water buffalo (Saraf et al., 2017). Furthermore, E-cadherin localization was firmly detected in mammalian spermatozoa (Rufas et al., 2000; Tsuchiya et al., 2006) and the reallocation in E-cadherin expression was analyzed in capacitated-spermatozoa released from oviduct epithelial cell co-cultures, representing the association of E-cadherin in assembly or disassembly of the oviduct-sperm reservoir (Caballero et al., 2014). Therefore, the localization of E-cadherin detected in the UTJ and isthmus in the present study could confirm the involvement of cell adhesion molecules in the formation of sperm reservoir in swamp buffalo.

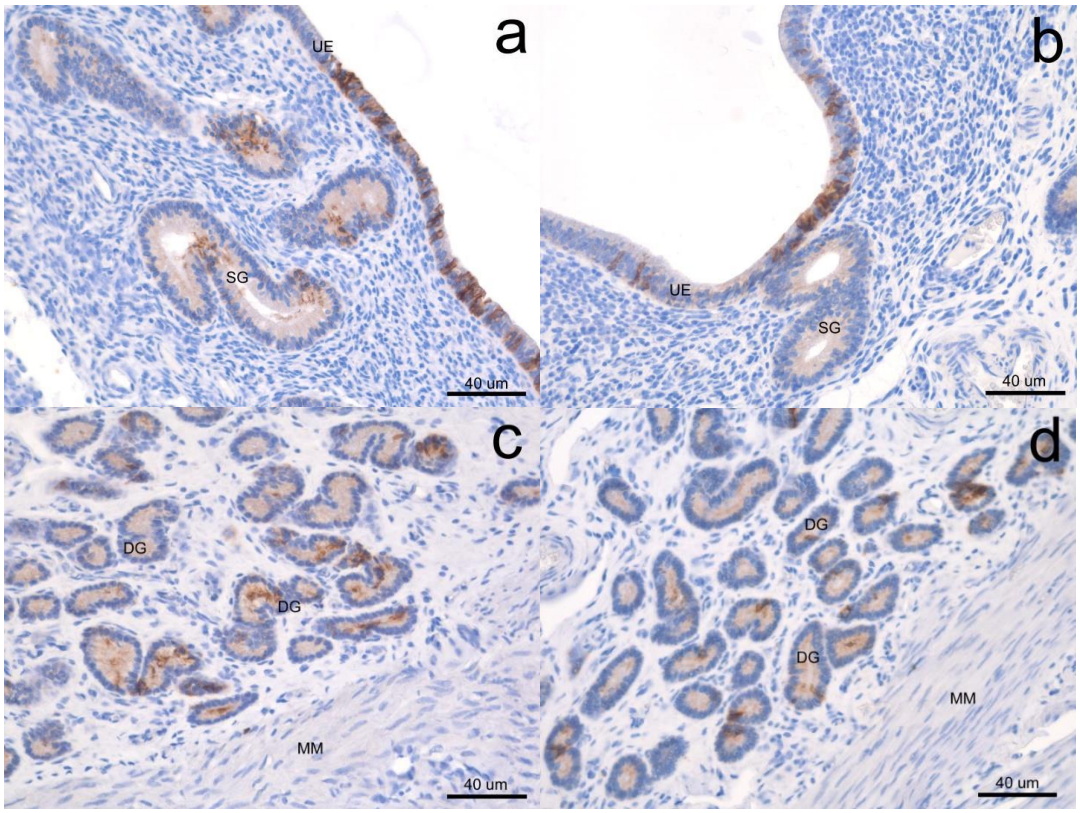

Figure 6 Immunohistochemical localization of E-cadherin in the uterine horns (a-d) of swamp buffalo at follicular (a, c) and mid-luteal (b, d) phases. Moderate to intense E-cadherin immunostaining was found in the uterine epithelial surface (UE), superficial endometrial glands (SG) and deep endometrial glands (DG), whereas other cells within the connective tissue and muscle cells in the smooth muscle layer (MM) were not seen the immunoreactivity during both phases.

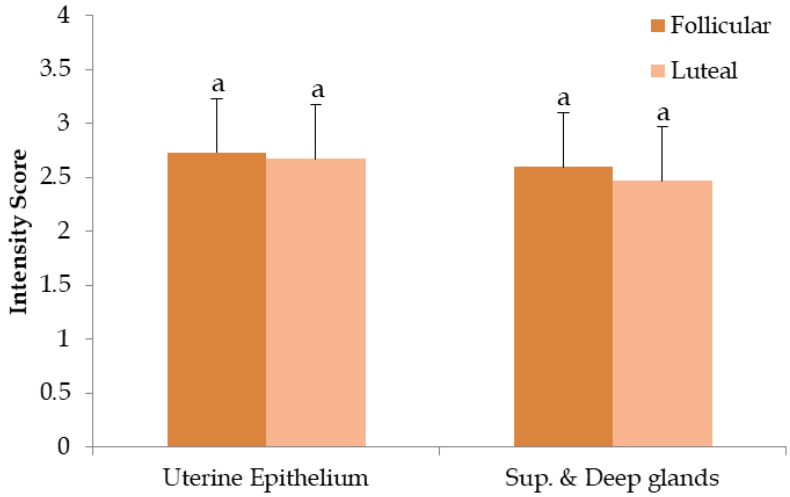

Figure 7 Immunohistochemical localization of E-cadherin in the uterine horns (a-d) of swamp buffalo at follicular (a, c) and mid-luteal (b, d) phases. Moderate to intense E-cadherin immunostaining was found in the uterine epithelial surface (UE), superficial endometrial glands (SG) and deep endometrial glands (DG), whereas other cells within the connective tissue and muscle cells in the smooth muscle layer (MM) were not seen the immunoreactivity during both phases.

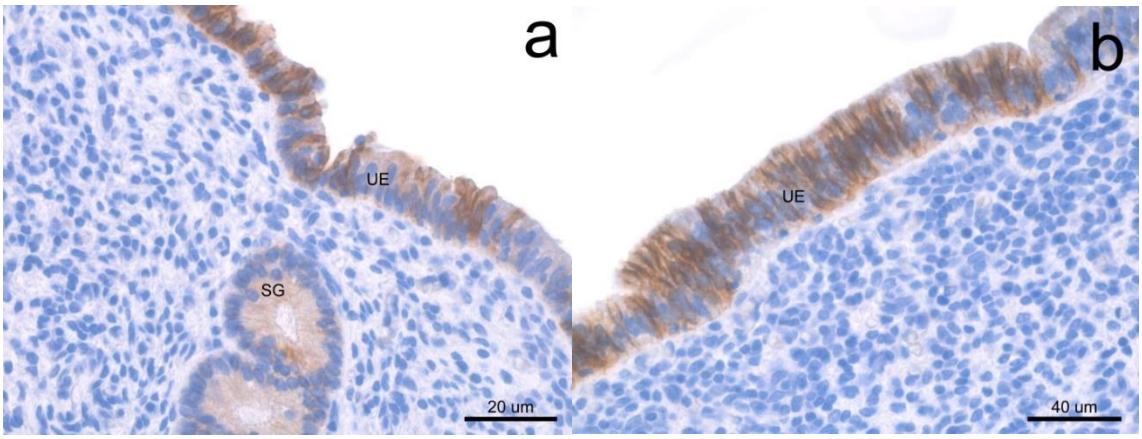

Figure 8 Immunohistochemical localization of E-cadherin in the uterine epithelial surface (UE) of swamp buffalo uterine horns at follicular (a) and mid-luteal (b) phases during higher magnification representing cellular membranous immunoreactivity. Notice the weak immunostaining within the cytoplasm of uterine epithelial cells and in some epithelial cells of superficial uterine glands (SG).

Considering the expression of E-cadherin, the stronger intensity of E-cadherin immunoreaction in the epithelial cells of UTJ and isthmus revealed significant ($P<0.05$) difference at the follicular phase compared to the mid-luteal phase. Interestingly, information on the pig oviduct indicated that the largest subpopulation of spermatozoa in the UTJ and isthmus at pre- and peri-ovulation period (follicular phase) was viable and uncapacitated, whereas most spermatozoa at post-ovulation (luteal phase) in this location were already capacitated (Rodriguez-Martinez et al., 2001; Tienthai et al., 2004; 2015). Similarly, earlier reports suggested that numerous categories of glycoproteins seemed to be absent from the plasma membrane covering the head at specific time of sperm capacitation, and that this event was related to a decrease in oviduct epithelium-sperm binding (Suarez et al., 1998; Tollner et al., 2008). Moreover, E-cadherin localization found in bovine spermatozoa demonstrated different patterns between bound and unbound spermatozoa using oviduct epithelial cell layer (Caballero et al., 2014). These investigations implied the alteration of the cell adhesion molecules in spermatozoa due to assembly or disassembly of the oviduct epithelium of sperm reservoir related to capacitation mechanism during selected phase of estrous cycle. Moreover, the release of bovine bound spermatozoa from oviduct epithelium culture was stimulated by heparin (Gualtieri et al., 2005) and released spermatozoa were noticed to be principally intact acrosome but adapted their functional state (Medeiros and Parrish, 1996). Definitely, these released spermatozoa revealed the modification in E-cadherin localization caused by increase in intracellular calcium, permitting these spermatozoa release from the site of sperm reservoir (Caballero et al., 2014). Therefore, it is possible that the strong intensity of E-cadherin immunoreaction in the sperm reservoir was related to the stages of estrous cycle, especially at the time of spermatozoa binding to the epithelial lining, and E-cadherin intensity might certainly be scrutinized in the swamp buffalo spermatozoa during attachment to and/or release from the epithelium of sperm reservoir. To confirm the assumption above in swamp buffalo, E-cadherin expression in the non-capacitated and capacitated viable spermatozoa and the epithelium of sperm reservoir with bound or unbound spermatozoa at selected phases of the estrous cycle by the use of immunofluorescence, western blotting and RT-PCR are required in forthcoming research.

As reported in former studies, E-cadherin immunopositivity was existent in the blastomeres of mouse (Ogou et al., 1982) and human (Bloor et al., 2002) embryos. Since implantation is a complicated event associated with the early embryo and the endometrial epithelial cells, consequently, the present study initially tried to investigate E-cadherin localization in the uterine horns during the follicular and mid-luteal phases. The immunolocalization of E-cadherin was detected in the endometrial and glandular epithelial linings of the swamp buffalo uterine horns and the immunoreaction was absent in the uterine connective tissue and smooth muscle, corresponding to the report of previous studies (Tabibzadeh et al., 1995; Beliard et al., 1997; Tsuchiya et al., 2006; Payan-Careira et al., 2016) of various species. Moreover, these former studies suggested that strong intensity of E-cadherin immunoreactivity was distinguished as a typical membranous staining in all epithelial cell types and weak intensity staining was seen within cytoplasm of these epithelial cells, similar to the present results. Considering the reproductive cycle, E-cadherin in normal human uterus is expressed throughout the menstrual cycle (Beliard et al., 1997; Tsuchiya et al., 2006), similar to the localization of E-cadherin in bovine (Caballero et al., 2014) and swamp buffalo uterine horns observed in the present study which showed no significant difference between the follicular and mid-luteal phases. However, the intensity variation of E-cadherin immunoreaction was reported in ewes; the increase in E-cadherin expression in the endometrial luminal and glandular epithelium was on day 16 to day 10 of the estrous cycle (day 0 = estrus) and by day 16 with the beginning of implantation, whereas the decrease in E-cadherin expression was noticed between day 10 and day 14 of the estrous cycle and after day 10 of pregnancy (Satterfield et al., 2007). These findings support the clear appearance of E-cadherin during the follicular to mid-luteal phases in the swamp buffalo endometrium in the present study because the tissue samples were collected roughly on days 18-20 and on days 8-10 of the cycle (day 0 =

estrus). It is known that ruminants are categorized as showing no menstruation and have a non-invasive type of implantation when the conceptuses are attached to the endometrial epithelial cells (Betteridge and Flechon, 1988; Bai et al., 2012). With these different reasons, the endometrial and glandular epithelial cells are necessary to adjust and provide the suitable mechanisms and microenvironment depending on the physiological status of these animals. The intense appearance of E-cadherin in the endometrial and glandular epithelial cells might represent the major functions of E-cadherin as the maintenance of epithelial growth and cell proliferation (van der Linden et al., 1995) during the follicular and mid-luteal phases in swamp buffalo. The decrease in E-cadherins at the time of implantation could be associated with the modification of adheren junction properties which facilitate the interaction between embryo and the endometrial epithelial cells (Bartly et al., 2014; Payan-Careira et al., 2016). Although the present study and other former studies indicated that the expression of E-cadherin in the endometrium was independent of the selected phase of estrous cycle or menstrual cycle, researches in human (Shih et al., 2004) and animals (MacCalman et al., 1994; Payan-Careira et al., 2016) reported that both estrogen and progesterone were able to induce E-cadherin transcription. Therefore, the variability of E-cadherin protein and transcription in the endometrial epithelial cells throughout the estrous cycle and also the time at implantation in swamp buffalo must be investigated by western blotting and RT-PCR in future project.

In conclusion, this study is the first to describe E-cadherin localization in the sperm reservoir of the oviduct and the endometrium of uterine horns in swamp buffalo during the follicular and mid-luteal phases. These findings support the suggestion that E-cadherin in the UTJ and isthmus of the oviduct might cooperate with other substances, i.e. hyaluronan and syndecan-1 (Tienthai, 2011), in the formation and regulation of sperm reservoir by assembly and disassembly of spermatozoa during the estrous cycle. In addition, E-cadherin expression in the endometrial and glandular epithelial cells of uterine horns is associated with the conservation of epithelial architecture and proliferation at the period of estrous cycle to prepare appropriate morphology and microenvironment before implantation and pregnancy.

Acknowledgements

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บทคัดย่อ

การแสดงออกของ E-cadherin ในท่อนำไข่และปีกมดลูกกระบือปลักในช่วงวงรอบการเป็นสัด

ไพศาล เทียนไทย

วัตถุประสงค์ของการศึกษานี้ เพื่อตรวจสอบลักษณะการแสดงออกของ epithelial cadherin (E-cadherin) ในท่อนำไข่และปีกมดลูกของกระบือปลักซึ่งอยู่ในระยะฟอลลิคูลาร์และระยะลูเทียลช่วงกลาง E-cadherin เป็น transmembrane glycoprotein ชนิดหนึ่งซึ่งมีบทบาทสำคัญในกลไกการปรับสภาพของเยื่อบุ การเพิ่มจำนวนเซลล์ การยึดเกาะระหว่างเซลล์กับเซลล์ และการยึดเกาะระหว่างเซลล์กับชั้นเยื่อบุ เก็บอวัยวะสืบพันธุ์ของกระบือปลักเพศเมียจากโรงฆ่าสัตว์ทั้งหมด 30 ตัว คัดแยกตามวงรอบของการเป็นสัด โดยแบ่งเป็นระยะฟอลลิคูลาร์ จำนวน 15 ตัว และระยะลูเทียลช่วงกลาง จำนวน 15 ตัว จากนั้น นำตัวอย่างเนื้อเยื่อที่ได้จากท่อนำไข่และปีกมดลูกมาผ่านกระบวนการเตรียมเนื้อเยื่อทางจุลกายวิภาคศาสตร์ และศึกษาด้วยเทคนิคทางอิมมูนโนฮีสโตเคมี การศึกษาพบว่า E-cadherin มีการปรากฏในชั้นเยื่อบุส่วน uterotubal junction (UTJ) และอิสธ์มัสของท่อนำไข่ทั้งในระยะฟอลลิคูลาร์และลูเทียลช่วงกลาง ซึ่งความเข้มในการติดสีปรากฏชัดเจนอย่างมีนัยสำคัญทางสถิติ ($P<0.05$) ในระยะฟอลลิคูลาร์เมื่อเปรียบเทียบกับระยะลูเทียลช่วงกลาง การปรากฏของ E-cadherin พบได้เช่นกันในชั้นเยื่อบุของปีกมดลูกและเยื่อบุของต่อมมดลูกชั้นผิวและชั้นลึกทั้งในระยะฟอลลิคูลาร์และลูเทียลช่วงกลาง ซึ่งความเข้มของการติดสีไม่พบความแตกต่างอย่างมีนัยสำคัญทางสถิติ ($P<0.05$) การศึกษาวิจัยนี้บ่งชี้ว่า การแสดงออกของ E-cadherin บริเวณ UTJ และอิสธ์มัสของท่อนำไข่อาจทำงานร่วมกับสารอื่น ๆ ที่พบภายในที่กักเก็บตัวอสุจิในการควบคุมการยึดเกาะตัวอสุจิกับเยื่อบุท่อและการปลดปล่อยตัวอสุจิออกจากเยื่อบุท่อ ขณะที่การแสดงออกของ E-cadherin ในปีกมดลูกอาจเกี่ยวข้องกับกลไกการปรับสภาพของเยื่อบุและการเจริญเพิ่มจำนวนขึ้นของเซลล์เยื่อบุ เพื่อเตรียมพร้อมเยื่อบุก่อนที่จะมีการฝังตัวของตัวอ่อนระยะบลาสโตซีสต์

คำสำคัญ: E-cadherin วงรอบการเป็นสัด บริเวณกักเก็บตัวอสุจิ ท่อนำไข่ มดลูก กระบือปลัก

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