# Observation of isthmic epithelial cells from fallopian tubes at follicular phase by light and scanning electron microscope\*

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## **Case report**

## Introduction

Fallopian tubes in humans are musculomembranous tubes between ovaries and uterus about 10-15 cm in length. They are divided into four segments: Pars interstitialis, isthmus, ampulla and infundibulum. Their walls are composed of mucosa, muscular layer and serosa. Fallopian tubes receive the ovum expelled by the ovary and carry it toward the uterus. During the ovulation period, fallopian tubes exhibit active role that their lumens provide an adequate environment for fertilization and muscular lavers contract rhythmically for moving eggs. Endosalpinx layer and lamina propria of the mucosa project many folds into the lumen. These folds are different in the segments of the tubes. Isthmic segment which is containing adrenergic receptors, plays a role as a sphincter. The effect of ovarian hormones makes differences on the structure and form of endosalpinx layer. These differences and sphincter increase the meeting chance of spermatozoa to ovum. The movement of the spermatozoa is too much rapid to be accounted for by their intrinsic motility. Fertilization usually occurs in the ampulla near its junction with the isthmus and initial development of the conceptus to the morula stage (1,2). In chronic salpingitis, endosalpinx folds defects cause some physiological problems on fertilization or transporting zygote into uterine cavity (3).

The aim of tubal sterilization is to prevent meeting of spermatozoa with ovum in fallopian tubes. Therefore no embryo will be produced and contraception will be continuous (4).

Here, we studied with a family having enough children. She was sterilized with Modified Pomeroy Method (4) at the 10th days of menstrual cycle. We aimed to determine (investigate) ultrastructure of A patient, at the age of 36, with regular menstruation, with two children admitted for tubal sterilization.

The blood sample was taken and centrifuged at 3000 rpm, 4<sup>o</sup>C for 30 min. Serum was transferred to new tube. FSH (Follicle-stimulating hormone, J&J, Amerlex-M) and LH (Luteinizing hormone, J&J, Amerlex-M) levels were detected by magnetic separation method. Coated tubes method was used for estradiol hormone analysis (17 ß Estradiol RIA CT, Radim, Italy). The measurement was done in Iso Data 100 series USA Gamma counter (5). All analysis were done in Gülhane Military Medical Academy of Nuclear (GATA), Department Medicine. Phannestiel minilaparatomy was performed by general; anesthesia. Two tissue samples from right and left fallopian tubes of isthmic region were taken by Modified Pomeroy Tubal sterilization method (4). Samples were divided into two parts by microsurgery scissors. One of them was prepared for light microscopy (LM) and scanning electron microscopy (SEM).

For LM, the sample was fixed in % 10 buffered formalin for a day, dehydrated in ethanol, after Xylol application, the sample was embedded into paraffin. The sections were cut in 3-5 micron thickness by Shandon Lipshan microtome (Rotary microtome-Detroit, USA) and were stained with Haematoxyline-Eosin (H&E) (6). The slides were examined under LM (American Optical Cooperation Spencer). The microphotographs were taken with Zeiss II microscope.

The sample for SEM was fixed in 2.5 % glutaraldehyde in sodium phosphate buffer (pH 7.2) (7). The sample then was washed in the same solution and postfixed in 1 %  $0s0_4$  in sodium phosphate buffer (pH: 7.2) for 1 hour. After that, it was rinsed in sodium phosphate buffer for 1 hour, dehydrated in graduated acetone series (50 %, 70 %, 90 %, 100 %) for 10 minutes for each one. The samples were dried at critical point (31.5 °C) with liquid C0<sub>2</sub>, in Polaron Critical Point Drying Apparatus. The dried samples were mounted to stups with silver glue and coated

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with gold (Polaran PS 100 E 510 Sputter Coater). The samples were examined with SEM (Japan Jeol JSM 840 Model) and microphotographs were taken.

Blood levels of FSH: 3.2 mlU/ml, LH: 8.8 mIU/ml, E2:310 pg/ml were determined by RIA in GATA Nuclear medicine laboratory, Ankara.

In the isthmic region of fallopian tube endosalpinx has shown some foldings. The lumen of the tubes were lined by simple ciliated columnar epithelium, and nonciliated-secretory cells also, few peg cells and reserve cells are present in the epithelium (Fig 1). The simple ciliated columnar epithelium has looked like pseudostratified epithelium depends on sectioning. The number of ciliated cells is more than that of nonciliated cells in this sample. Both type of cells were arranged in groups (Fig 2). Microvilli in nonciliated cells were, numerous and could be seen easily in SEM. The long cilia clusters were abundant on the upper surface of ciliated cells. Some pits were seen among the cells (Fig 2).



Figure 1. The isthmic region of fallopian tube endosalpinx lumen lined with ciliated ( $\rightarrow$ ), nonciliated-secretory ( $\Rightarrow$ ) and reserve ( $\rightarrow$ ) cells, (LM, H&E,X560).



Figure 2. Cell groups of ciliated ( $\rightarrow$ ) and nonciliatedsecretory cell ( $\Rightarrow$ ) and some pits (SEM,x 2.000).

## Discussion

Ciliated cells and nonciliated-secretory cells in fallopian tube endosalpinx were effected from the hormones. They were changed morphologically and histologically during the menstruation period. These cells don't show secretion activity in the end of pregnancy. They are known to be in rest during this period (2).

Küni and Kaya have show that (8) siliogenesis was increased and glycogen was stored during proliferative period. During the ovulation period glycogen was thrown to tubal lumen together with cytoplasmic parts, glycogen secretion was decreased during the luteal phase (8).

In the second half of the pregnancy, at the beginning of the puerperium, number of ciliated cells increases at ampulla and fimbria as they decrease at isthmus. However, their number decreases and they get smaller at puerperium and lactation period. Their ciliary movement are related with ovarial hormone. During and after ovulation, cilia motility increases to maximum degree because of high level of progesterone release. In addition to that, ciliary motility of ampulla and fimbria tends to synchronize the exist of ovum. In the end of secretion phase, the number of ciliated cells are decreased. Ciliated cells and siliogenesis in adult help ovum transport to spermatozoa and fertilization developes (9).

Birkenfeld et al.(10) have shown that, in rabbit oviduct normal distribution of nonciliated-secretory cell and ciliated cells were affected after a 10 mg clomiphen citrate treatment. The cells were differentiated and nonciliated cells were released their secretion by apochrin or merochrin type.

Oviduct of new born rat was examined by Yılmazer et al. (11). They have shown that centriole morphogenesis was continued during the 14th day. In addition that, developing ciliated cells and some cells contained precursor material not like mature centriol were also observed.

The SEM studies on cow oviduct epithelium was shown that ciliated cells were abundant in fimbria and ampulla during follicular phase. However, epithelium cells were lost their cilia in lutheal phase and nonciliated cells were observed in isthmic and ampulla- isthmic region (12).

In this study, ciliated cells and nonciliatedsecretory cells were observed together in isthmic region of fallopian tubes of reproductive woman at the 10th day of menstrual cycle by light microscope and scanning electron microscope. The number of ciliated cells was increased in isthmic region of fallopian tubes by the effect of estrogen in the proliferative phase.

Li and Chen (13) studied comparatively ligated and normal oviduct epithelium in nine women. They have shown that,scar tissue in the ligated oviduct where was 0.5 cm from both the distal and proximal ends of the ligation scar, had foldings towards mucosa. In this region, the cells lost their cilia and had abundant microvilli. However, 1 cm away from this region, normal ciliated cells were observed. Tubal microsurgery reanastomosis operation can be performed to women who have infertility problems because of tubal obstruction, divorce, remarriage and reversal of sterilization. The location and structure of scar tissue of fallopian tubes must be observed by laporoscopy before microsurgery for the success of reanastomosis, 1 cm. proximal and 1 cm. distal part of scar tissue must be removed to get positive results.

If infertility problem is still not solved in this kind of operation, the importance of tubal morphological changes was considered. Therefore these group of people treated with another kind of assisted reproductive technology (ART) such as IVF (in vitro fertilization) or sperm microenjection to ovum and ET (embryo transfer).

Li and Chen (13) have shown oviductal stomata's among some secretory cells of hydrosalpinx epithelia in ligated oviduct. In this study, some pits like oviductal stoma was observed among normal isthmic epithelial cells. These pits should be investigated by transmission electron microscope (TEM) in detail.

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