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Research Article
Morphology of the Tracheal Epithelium in the Quail (*Coturnix coturnix japonica*)

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Abstract

Background and Objectives: Despite the importance of the tracheal epithelium in the avian respiratory pathology, the histology of the trachea has not been fully recorded. The main purpose of this study was to extend the microscopic investigations of the tracheal epithelium in the quail, in order to add some information on the fine structure of the luminal surface. **Materials and Methods:** The structure of the luminal surface of the tracheal epithelium of the quail was studied using scanning electron microscopy (SEM) and light microscopy (LM) histochemistry. **Results:** The pseudo stratified columnar epithelium composed mainly of ciliated cells and dispersed islands of single or groups of non-ciliated cells. The non-ciliated cells comprised a variety of basal, goblet cells and PAS positive cells exhibiting merocrine secretion. **Conclusion:** It was assumed that the tracheal epithelium of the quail closely resembles that of the chicken but the identity of the epithelial non ciliated cells still remains enigmatic as is the case with these cells of other bird species.

Key words: Ciliated epithelium, goblet cell, merocrine secretion, avian, quail

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The histology of the trachea in the quail has been described by means of optical microscopy by Cevic-Demircan *et al.*[1]. The epithelium of the trachea contained a ciliated pseudo stratified cuboidal epithelium. Intraepithelial glands were present in the epithelium and goblet cells were observed[1].

The detailed histology of the chicken's tracheal epithelium has been investigated by Purcell[2], Walsh and McLelland[3], Lai and Ibrahim[4]. The authors identified by transmission electron (TEM) and scanning electron microscopy (SEM) four main cell types. Three of them were commonly named ciliated, goblet cells and basal cells, whereas, the fourth type differed according to the author. Purcell[2] named these cells mucous gland cells, Walsh and McLelland[3] called them non-ciliated columnar cells, whereas, Lai and Ibrahim[4] claimed that they represent an immature type of goblet cells.

The histology of the trachea in birds has been the subject of limited research by various scientists both in terms of normal and pathological conditions of the respiratory system.

The luminal surface of the trachea was used as a suitable model for evaluating possible changes after the vaccination of avian species[5,6].

In view of the Japanese quail as a bird of biomedical and agricultural interest, it was important to record the normal morphology of the tracheal epithelium in order to establish a baseline of information for comparative purposes.

MATERIALS AND METHODS

This study was undertaken from January, 2014-March, 2015 at the laboratory of Anatomy, Histology and Embryology in Karditsa, Greece. All procedures were carried out in accordance with guidelines established by the University of Thessaly, Greece, for the use of animals. Six 3-months old Japanese quails from a minimal disease flock were used. The birds were euthanized by the administration of sodium pentobarbitone into the wing vein. Pieces of trachea which were cut longitudinally, were washed four times, for 15 min each time, with sodium cacodylate buffer (pH 7.2), transferred for 1 h to 1% Osmium tetroxide (OsO_4) and dehydrated in graded acetone. Tissues were critical point-dried in carbon dioxide (72-75 barr), mounted onto stubs and sputter coated with platinum and gold in a Bal-Tec sputter coater. Specimens were observed in a JEOL, JSM 840 scanning electron microscope. Other pieces of trachea were processed for light microscopy. The samples were embedded in paraffin wax. The sections, 5 μm thick, were stained with haematoxylin and eosin and periodic acid-schiff (PAS).

RESULTS

Examination of the luminal wall of the trachea with the scanning electron microscope revealed two principal surfaces, ciliated and non-ciliated. In this epithelium, the ciliated cells which formed the ciliated surface were abundant without visible borders among them (Fig. 1). The median height of the cilia was 4 μm whereas, the high density of the cilia did not permit the detailed observation of the underlying apical surface of the ciliated cells. However, at the zones of transition between ciliated and non-ciliated cells, microvilli among the cilia could be observed. Among the cilia, patches of non ciliated cells were recorded. The luminal surface of the non ciliated cells exhibited various appearances (Fig. 1). Sites containing one (Fig. 1b) or more cells (Fig. 1d) exhibiting the typical appearance of goblet cells were observed . The goblet cells were bulged into the lumen of the trachea and were surrounded by a narrow band of cytoplasm (Fig. 2). At their apical surface, there were no cilia but numerous microvilli arose. On occasion cells were seen which, whilst showing characteristics of actively secreting non-ciliated cells, had a few cilia on their luminal surface (Fig. 1a,c). These cells may represent transitional forms between the non-ciliated and ciliated cells.

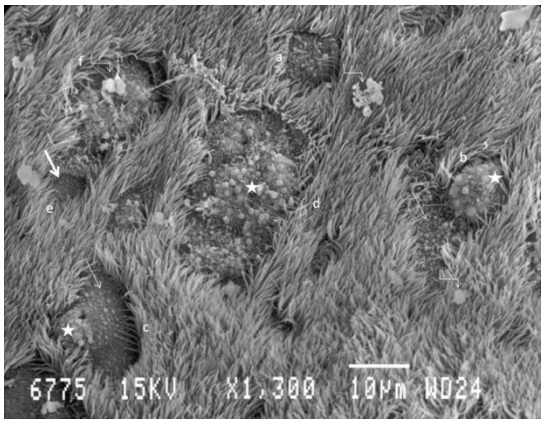

Fig. 1: A typical ciliated region of the trachea. Dispersed among the ciliated cells are a number of typical goblet cells (asterisk). The curved arrows point to discharged mucous. The arrows point to non ciliated cells

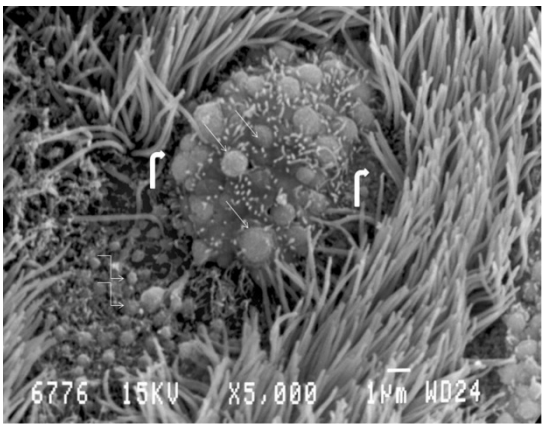

Fig. 2: Luminal surface of a goblet cell. The apical surface is bulged exhibiting numerous protrusions (thin arrows) and microvilli. The curved thick arrows denote the zone between the cell and the surrounding cilia. The curved thin arrows show small protrusions which probably belong to an adjacent non ciliated cell

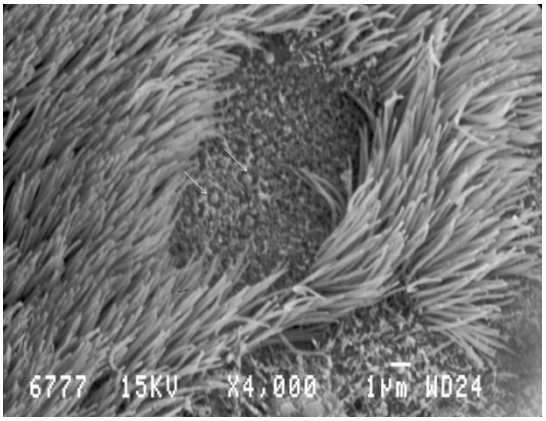

Fig. 3: Luminal surface of a non ciliated cell encircled by cilia. Note the small protrusions (thin arrows) exhibiting merocrine secretion

Mucous occurred as discrete droplets, which frequently were being entangled in the cilia as well as released from goblet cells (Fig. 1f).

Some non-ciliated cells (Fig. 1e, 3) showed evidence of an active merocrine secretory phase. Merocrine secretion was indicated by individual cells bearing small protrusions into the tracheal lumen. On occasion, secretory vesicles appeared to be in the process of budding from the ends of these protrusions. The rest of the apical surface was flat (Fig. 3).

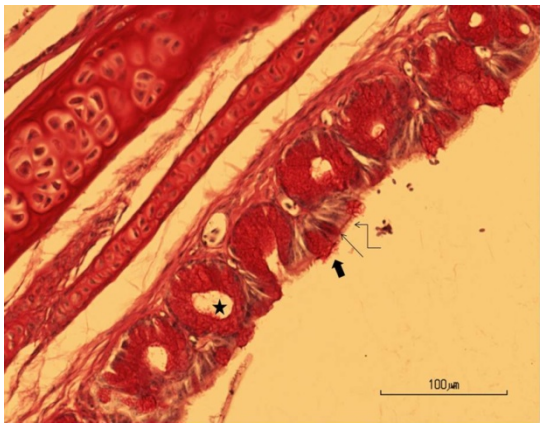

Fig. 4: Light micrograph of the trachea. The pseudo stratified ciliated columnar epithelium is interrupted by numerous simple acinar, mucous glands (asterisk) which invaginate into the shallow lamina propria. Basal cells (arrow heads) are noted beneath mucous cells. PAS positive cell (thin arrow), goblet cell (fat arrow), columnar ciliated cell (crooked arrow)

Light microscopy (Fig. 4) indicated that the lining is pseudo stratified ciliated columnar epithelium with numerous simple acinar mucous glands invaginating into a shallow lamina propria. Tracheal glands were comprised of goblet cells and basal cells. Basal cells of the tracheal glands were identical to those of the epithelium proper. They formed a discontinuous demilune around the basal and lateral aspects of the gland and were continuous with the fenestrated layer of basal cells of the epithelium proper. The tracheal epithelium proper was comprised of four types of cells: Basal cells, PAS positive goblet cells, ciliated cells and PAS positive columnar cells (Fig. 4).

DISCUSSION

SEM and LM observations showed that the tracheal epithelium of the quail, was composed of ciliated and non- ciliated cells. The non-ciliated cells comprised the goblet cells and PAS positive columnar cells. The tracheal lining of the quail was a ciliated columnar epithelium rather than cuboidal epithelium as described by Cevic-Demircan *et al.*[1]. The cellular population of the quail's tracheal epithelium closely resembled that of chicken.

The luminal morphology of the avian tracheal epithelium has been the subject of research in order to clarify some issues related with avian respiratory problems[7,8]. The SEM

appearance of the majority of epithelial cells resembles the descriptions of Lai and Ibrahim[4] in the chicken. According to the authors, both SEM and TEM have shown that ciliated cells are the most numerous cell types in the tracheal epithelium. However, the authors claimed that the goblet cells present various apical surfaces according to their status of function. The non ciliated columnar cells reported by Walsh and McLelland[3], may coincide with the non-ciliated cells observed in this study. The authors, claimed that the cytoplasm of these cells did not exhibit secretory material. However, their appearance in TEM micrographs resembled the non-ciliated cells of the current research.

Despite the fact that the ultra structure of the epithelial cells of the avian trachea has been investigated in detail by transmission electron microscopy, there is still an obscurity about the identity of the non-ciliated cells[2,4,9].

In the present study, with the SEM and LM observations of the luminal surface, no definite deductions could be made. On the other hand, the existence of the goblet cells in various stages of maturation as the unique population of non-ciliated cells cannot be accepted.

An immuno-histochemical study of the tracheal avian epithelium could be useful to clarify furthermore the cellular identification of the avian trachea.

CONCLUSION

SEM observations and LM staining with PAS showed that the tracheal epithelium of the quail, in line with other avian species is mainly composed of columnar ciliated and non ciliated cells. The non ciliated cells comprised the goblet cells and the PAS positive columnar cells exhibiting merocrine secretion. The identity of the non ciliated cells requires further investigation.

SIGNIFICANCE STATEMENT

This study discovers the morphology and identity of the cells of the quail's epithelium that can be beneficial for better understanding the avian respiratory physiology and pathology. This study provides new data, which are useful baseline information on the quail's epithelium histology.

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