ENUMERATION OF EPIDIDYMAL CELLS IN THE SRI LANKAN GOAT: A LIGHT MICROSCOPIC STUDY

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Introduction

The epididymis is considered to be the main organ of sperm maturation. Although the epididymis has been divided grossly into head, body and tail the histological classification divides the epididymal duct into various segments such as initial segment, intermediate zone, caput etc. (Nicander and Glover, 1973). The epididymis is mainly composed of a highly coiled epididymal duct, which is lined by a pseudostratified columnar epithelium. The epithelium is composed of six main cell types; principal, basal, apical, halo, narrow and clear cells (Nicander and Glover, 1973).

Principal cells are the main cell type found in the epithelium of the epididymis in all mammals. These cells are tall columnar cells that synthesize high amount of proteins. Basal cells are triangular in appearance and lie on the basement membrane. Apical cells have apically located spherical nuclei and do not contact the basement membrane. These cells have the ability to endocytose substances from the lumen (Robaire, *et al.*, 2006). Halo cells are small round cells with a narrow rim of clear cytoplasm. Further, these cells are considered as intraepithelial lymphocytes, which may be important in immune functions. Narrow cells have elongated nuclei near the apical region and cell bodies tapering towards basement membrane. They are important in endocytosis and secretion of H^+ ions. The clear cells contain lipid droplets near the base, pale staining vesicles near the apex, and lysosomes in the middle region. These cells have functions similar to that of narrow cells. (Robaire, et al., 2006)

Detail information on the distribution of different cell types in different regions of the epididymis, particularly in the local goat is scarce. Hence, a study was undertaken to identify the proportional distribution of the different cell types in the different regions of the epididymis of the Sri Lankan goat.

Materials and Method

Testes from six adult male goats (n=6) together with the epididymides were collected from the Kandy abattoir and brought to the laboratory in ice. The epididymis was carefully dissected from each testis and separated into six regions namely proximal head (H1), distal head (H2), proximal body (B1), distal body (B2), proximal tail (T2), and distal tail (T2) based on the gross morphology.

Preparation of histological sections

Tissue samples of 0.5 cm thickness were taken from the center of each region of the epididymis and fixed in buffered formal saline. The fixed tissue samples were processed, 5 µm thick paraffin sections were prepared and stained with Haematoxylin and Eosin (H&E). Two separate slides were prepared from each region of the epididymis of each animal and stained with Periodic Acid Schiff (PAS) stain to demonstrate the presence of carbohydrates.

Enumeration of cells

A thousand cells were counted from each region of the epididymides using H&E stained tissue sections under high power (X 100) of a light microscope. Of the PAS stained tissue sections, two hundred cells were counted to determine the proportion of PAS positive cells.

Data analysis

Data were grouped according to the regions and cell types, and expressed as Mean ± SEM.

The variation of cell types within each region and among different regions were analyzed by ANOVA, and mean separation was done by Fisher's test using Minitab 14. The values were considered significant when $p<0.05$.

Results

Principal cells, basal cells, apical cells, halo cells, narrow cells, and clear cells were detected in all six regions and the percentage distribution of these cells are given in Table-1. The percentage of principal cells varied from 76 to 84 in different regions and the lowest percentage was found in the B1 region. On the other hand, percentage of the basal cells was high in the B1 region compared with that of the H1 region. The percentages of the apical cells were low in B1 and T1 regions compared with that of H2 region. The lowest percentage of halo cells was found in the H2 region and was

Table 1. The percentages of different cell types present in different regions of the epididymal duct

Region	Principal Cells	Basal Cells	Apical Cells	Halo Cells	Narrow Cells	Clear Cells
H1	84.5 ± 2.1 ^{a,u}	11.8 ± 1.9 ^{ac,v}	1.8 ± 0.4 ^{ac,w}	1.5 ± 0.5 ^{ab,w}	0.4 0± 0.17 ^{a,w}	0.12 ± 0.06 ^{a,w}
H2	83.9 ± 1.9 ^{ac,u}	12.8 ± 1.6 ^{ab,v}	2.15 ± 0.4 ^{ac,w}	0.8 ± 0.2 ^{ac,w}	0.35 ± 0.12 ^{a,w}	0.05 ± 0.05 ^{a,w}
B1	76.2 ± 1.1 ^{bc,u}	19.6 ± 1.0 ^{b,v}	0.82 ± 0.3 ^{bc,w}	2.9 ± 0.7 ^{b,x}	0.35 ± 0.19 ^{a,w}	0.23 ± 0.13 ^{a,w}
B2	77.8 ± 1.9 ^{bc,u}	17.2 ± 2.1 ^{bd,v}	1.2 ± 0.5 ^{bcde,w}	1.7 ± 0.4 ^{bc,w}	0.17 ±0.1 0 ^{a,w}	0.15 ± 0.06 ^{a,w}
T1	78.6 ± 1.5 ^{ac,u}	16.7 ± 1.0 ^{bc,v}	0.88 ± 0.3 ^{bc,w}	3.3 ± 0.9 ^{b,x}	0.23 ± 0.10 ^{a,w}	0.22 ± 0.10 ^{a,w}
T2	80.3 ± 3.0 ^{ac,u}	14.7 ± 2.7 ^{bc,v}	1.7 ± 0.6 ^{cde,w}	3.1 ± 1.0 ^{b,x}	0.20 ± 0.13 ^{a,w}	0.08 ± 0.04 ^{a,w}

$^{a-g}$ Means (n=6) ± SEM within same column with no common superscript differs significantly ($P < 0.05$); $^{u-z}$ Means (n=6) ± SEM within same raw with no common superscript differs significantly ($P < 0.05$).

significantly lower than those of B1, T1 and T2 regions. The narrow cells and clear cells were equally distributed in all regions of the epididymis.

The principal cells were the predominant cell type present in all the six regions. In any region, the second highest cell type was the basal cells. Of the different regions, the percentage of the apical cells found in B1, T1 and T2 regions was significantly lower than the halo cells present in the same regions.

Discussion

Epididymal morphology has been studied extensively in many species. In the goat it has been reported that only three cell types; namely, principal cells, basal cells and intraepithelial lymphocytes/ macrophages are found in all regions while the apical cells are limited to the head and the body regions of the epididymis (Goyal and Williams, 1991). However, the present study showed all these cell types throughout the epididymis. In contrast to previous studies in goats (Goyal and Williams, 1991) and in several other species (Nicander and Glover, 1973), it was found that the clear cells were present in all regions in the Sri Lankan local goats. This may be attributed to the fact that clear cells are difficult to identify with H&E staining only. In the present study, the tall columnar cells with a foamy appearance in H&E stained sections were considered to be clear cells. Their identity was confirmed by PAS staining. Even though earlier studies showed apical cells in few regions, in the present study it was observed in all the regions.

Conclusion

Although six cell types have been described in the epididymis of mammals not all are present in all the species studied so far. However, from the present study it is possible to conclude that all these cell types are present in the epididymis of the goat; although the results need to be further confirmed by electron microscopy.

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