

# Preliminary comparative immunocytochemical study of respiratory tract endocrine cells in certain rodents

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*Studies were performed on 3 species belonging to two families: Microtidae (7 common voles and 7 pine voles), and Muridae — 10 Wistar rats. In rodents the airways endocrine cells (ECs) are localised in the epithelium lining the larynx, trachea, bronchi, bronchioles and lung. CGRP-, synaptophysin (SY)-, calcitonin (CT)-, neuron-specific enolase (NSE)- and chromogranin A (CA)- immunoreactivity were nearly totally co-localised in ECs. In the region of the tracheo-larynx junction, CGRP- and NSE-positive cells were observed in the epithelium of the glands. It is surmised that some of the CGRP-positive ECs do not generate CT and CA, for the most part in ECs. In Microtidae ECs were more abundant than in the rat and were found even in the epithelium lining of the inside larynx in the transition region before the trachea.*

**key words: rodents endocrine cells, larynx, trachea, lung, immunocytochemistry**

## INTRODUCTION

Many of the endocrine cells (ECs) dispersed in various organs are derived from neural crest ectoderm. These cells secrete similar hormones [1, 3, 4]. ECs appears in the mammal airways as scattered single ECs and, less frequently, as innervated groups — “the neuroepithelial bodies (NEBs)” [2]. No description of ECs occurrence in the airway epithelium of small wild rodents living in Central Europe can be found in the available literature. The aim of the present investigations was to compare airways ECs of rats (the Muridae family) and rodents from the Microtidae family, namely pine voles and common voles. These rodents occupy a different ecological space from rats and live in a different environment, spending a very long time in the burrow. In winter these rodents not migrate into farm buildings. It is

thus probable that the regulatory processes controlling their metabolism and respiration may be slightly different from those in rats.

## MATERIAL AND METHODS

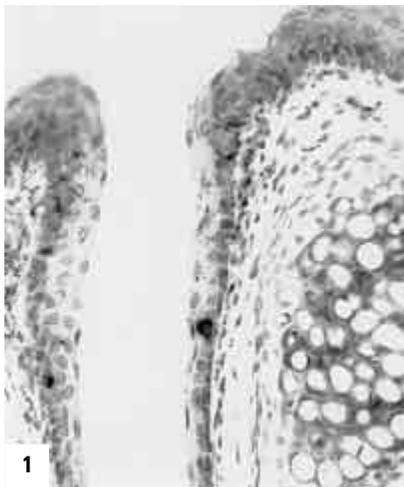
The material used for analysis was collected in 1996–1997. Tissues were taken from the upper and lower airways of 3 rodent species belonging to two families: Microtidae (7 common voles — *Microtus arvalis* and pine voles — *Pitymys subterraneus*), and Muridae — 10 Wistar rats. Two specimens were extracted, the larynx together with the trachea and the right lung with the lobar bronchi. The entire lungs were fixed in Bouin’s fluid with intratracheal infusion, and both specimens were then fixed separately for the duration of 18 h. Paraffin 5- $\mu$ m sections were taken. Immunocytochem-

ical reactions were performed using ABC technique (detail see [1]) Specific antibodies were used against calcitonin (CT), CGRP, neuron-specific enolase (NSE), chromogranin A (CA), somatostatin (SS) and synaptophysin (SY).

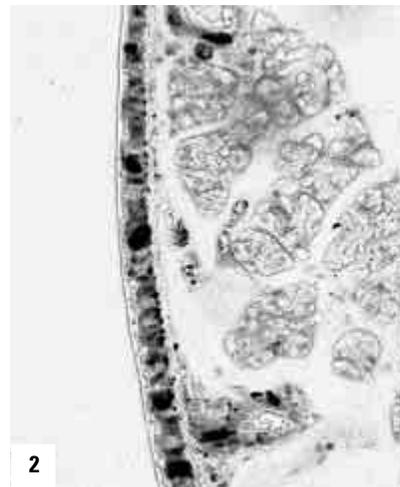
### RESULTS AND DISCUSSION

CGRP was a very good marker for all ECs in the epithelium lining of the rodents' respiratory tracts from the larynx to the lung alveolar epithelium (Fig. 1–3). Numerous CGRP- and NSE- immunoreactive nerve fibres were distributed around the airways epithelium (Fig. 1, 2, 4). In the higher internal part of the larynx, still showing the surface covered with

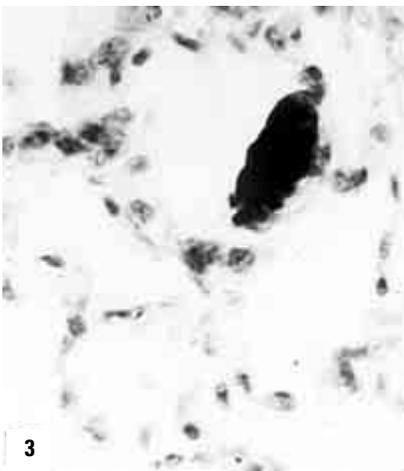
stratified squamous non-keratinized epithelium, only a few single ECs were found (Fig. 1). Particularly in the rat, ECs were found sporadically in this location. In the common vole and pine vole, however, they were observed more often, especially in the region of the tracheo-larynx junction. In the epithelium of this area, NEBs composed of at least a few, more seldom a dozen cells, were also observed (Fig. 2). Moreover, in the mucous membranes of the junction area numerous glands were observed. In the epithelium of these glands, a small amount of single CGRP- and NSE-positive ECs was found (Fig. 2). In the epithelium, located beneath the trachea, single ECs and NEBs were distributed



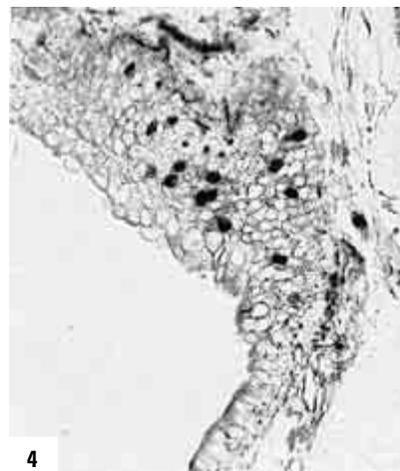
**Figure 1.** Longitudinal section through a pine vole larynx. CGRP-immunopositive single EC are visible against the stratified squamous non-keratinized epithelium ( $\times 200$ ).



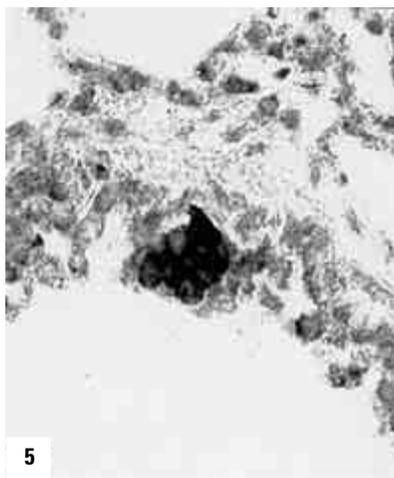
**Figure 2.** Mucous membrane of the common vole tracheo-larynx junction. Immunopositive CGRP reaction in numerous ECs of the epithelium and in ECs occurring in glandulae tracheales, as well as in nerve fibres ( $\times 200$ ).



**Figure 3.** Rat lung. CGRP-immunopositive reaction is visible against all cells in NEB of the lung alveolus ( $\times 400$ ).



**Figure 4.** Slantwise section through the pine vole trachea — region of the bifurcation. Numerous NSE-immunoreactive single ECs and nerve fibers are visible against the tracheal epithelium ( $\times 400$ ).



**Figure 5.** Rat lung. Strong CT-immunopositive reaction in NEB cells located in the epithelium lining the bronchiales ( $\times 400$ ).



**Figure 6.** Mucous membrane of the common vole tracheo-larynx junction. Only one CT-positive cell is visible ( $\times 200$ ).

very unevenly. These were mainly concentrated around the opening of the trachea glands (Fig. 2). NEBs observed here were generally of small size. The observed density of ECs population present throughout the trachea epithelium was higher in both wild rodent species in comparison to the rats. In all the animals particularly numerous ECs were observed in the bifurcation zone of trachea, bronchi, and bronchioles (Fig. 4, 5). In the lower airways ECs appeared in small numbers in all the animals and no significant difference between the species could be observed. ECs were particularly rarely found in alveolar ducts, and alveolar sacs (Fig. 3). CT-positive cells (Fig. 6) were observed in smaller numbers than NSE-, and CGRP- positive cells (Fig. 2). Control reactions yielded negative results. SS was observed occasionally in a very small number of ECs.

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