Morphological studies of the lingual gland of 
*Onychodactylus japonicus* (the 60th stage)  
—Two kinds of granules—

Iwao Sato, Makiichi Kobayashi, Tooru Sato* and Takeshi Muraki**  
*Department of Anatomy (Chief: Prof. Yuji Nagumo), Nippon Dental University,  
9-20 Fujimi 1-chome, Chiyoda-ku, Tokyo 102, Japan  
**Department of Anatomy (Chief: Prof. Takuro Suzuki), St. Marianna University,  
School of Medicine

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**Introduction**

Numerous reports are available on the major salivary glands of mammals. Only a very small number of studies, however, are available of the minor salivary glands, particularly the lingual gland, especially in amphibians. Only the study by Fahrmann3) was reported recently. In the present study, morphological studies were performed on the cells in the terminal portion of the lingual gland of *Onychodactylus japonicus*, in the 60th stage of metamorphosis, and on their secretory granules.

**Materials and Methods**

*Onychodactylus japonicus* maintained in a thermostatic water bath at 11-13°C after hatching were used. As food, thread earthworm (*Limnodrilus*) were given from the 50th stage when the mouth opens up (with a mean body length of 2.8 cm and a body weight of 0.3 g). After the 60th stage (with a body length of 3.5 cm and a body weight of 0.5 g) pebbles were used to make a land in the corner of the water bath to form a natural habitat.

Part of the mandible was obtained from 10 *Onychodactylus japonicus* (60 stage), prefixed in glutaracodylate (pH 7.2), postfixed in 1% osmic acid (pH 7.4), embedded with in Epon 812 and prepared into thin sections. After staining in toluidine blue, the sections were first studied under a light microscope. Some of the sections were subjected to double staining with uranyl sulfate-lead citrate and studied under a transmission electron microscope (Hitachi H700).

**Results**

1. Light microscopic findings

Glandular-acinar cells were distributed all over the tongue. Granules stained deeply by toluidine blue (type A) and those stained rather lightly were mixed together (Fig. 1). No metachromasia was induced by toluidine blue in any of these granules. The nucleus of these glandular-acinar cells was located in the basal portion and was oval in shape. Myoepithelial cells appeared to surround the glandular-acinar cells. The glandular-acinar

![Fig. 1 Granules deeply stained with toluidine blue (A type, white arrow) and those lightly stained (B type, black arrow). ×350](image-url)
cells were stained lightly by toluidine blue.

2. Transmission electron microscopic findings

The glandular-acinar cells of the tongue assumed a conical shape, with a long diameter of about 3 μm and width of about 2 μm.

![Fig. 2 Glandular acinar cells](image)

The nucleus was located on the basal side. The nucleolus showed good development in the Pars granulosa. Rough-surfaced endoplasmic reticulum was well developed and numerous secretory granules are noted. ×2700

N; nucleus
Mt; mitochondria
SER; rough-surfaced endoplasmic reticulum
Seg; secretory granule

![Fig. 3 Two kinds of secretory granules](image)

Agranule with homogeneous electron density (A type) and a granule with central core (B type) are found. ×5000

A; A granule
B; B granule

The nuclei were adjacent to the basal portion and the external nuclear membrane consisted of an internal and external nuclear capsule with nuclear membrane pores measuring 0.1 μm (Fig. 2). Two kinds of secretory granules were found (Fig. 3). One was a large homogeneous granule which had a slightly high electron density. In an enlarged picture, the electron density is either high or low. This was called ‘A’ granule (Fig. 4).

The other kind of granule was somewhat smaller in size, with a central core with high electron density, surrounded by a light halo with low electron density. This biphasic granule was called ‘B’ granule (Fig. 5).

![Fig. 4 A granule](image)

High electron density Seg I
Low electron density Seg II
×30000

![Fig. 5 Magnification of a ‘B’ granule](image)

Black arrow indicates site of fusion with SER
×30000
Discussion

*Onychodactylus japonicus* changes its mode of living before and after metamorphosis, with consequent changes in its eating habits\(^4\). Along with this, it is readily conceivable that the morphology and functions of the lingual glands also change. In the Amphibia without major salivary glands, the lingual and palatine glands have probably developed markedly with remarkable functional changes\(^4\). *Onychodactylus japonicus* has markedly developed lingual glands as compared to mammals and the entire tongue may be said to consist of lingual glands alone. The biphasic secretory granules like the B granules of *Onychodactylus japonicus* are seen in the cells of the parotid and Ebner glands\(^5,6\). Some rodents such as rats, mice and hamsters have two kinds of secretory granules, type A and B, with biphasic granules. Unlike *Onychodactylus japonicus*, however, these biphasic granules consist of a halo with a high electron density surrounding a core with a low electron density\(^7,8\). The B type biphasic granules of *Onychodactylus japonicus* have a central core with a high electron density surrounded by a halo with a low electron density. Both, however, contain mucopolysaccharides and proteins\(^7,8\). The glandular acinar cells of the parotid and submandibular glands of Rhesus monkeys (*Macaca irus*) also have secretory granules with a central core. The portion of this granule with a low electron density stains positive for silver by the methenamine silver method\(^3,6,9\), and therefore definitely contains carbohydrate. In mammals such as cows, cats, some bats, domestic rabbits and hamsters, taking the parotid gland for example, the secretory granules are homogeneous, whereas, in the monkey, biphasic secretory granules with a small central core with a high electron density and triphasic granules were noted\(^6,10\). A comparison between man, Rhesus monkeys and rodents revealed a histological resemblance of the secretory granules of Rhesus monkeys to those of rodents rather than to those of humans\(^8,10-12\). Among the mammals, dogs and pigs have biphasic granules similar to those of monkeys. Thus some differences are noted in the secretory granules of different animals, but a general resemblance is recognized. The kind of secretory granules depends not so much on the species, genus or order of the animals, as on the mode of living and especially the eating habits.

Acknowledgement

Many characteristics were found in the lingual glands in *Onychodactylus japonicus* with poor development of the major salivary glands. Two kinds of granules were found in the cells located in the terminal portion of the lingual gland, one being a mucous homogeneous granule (A type) and the other with a central core surrounded by a light halo with a low electron density (B type). Attempts were made to investigate the structure and functions of these granules.

References

