

## LABORATORY OF MORPHODIVERSITY

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The aim of this laboratory is to observe the variety of morphogenetic processes that occur in the course of the ontogenesis of multicellular organisms and to analyze the mechanisms of such processes, mainly by morphological methods. The accumulation of such analyses of the embryogenetic processes of related species is expected to provide an insight into the evolution of morphogenetic processes. This laboratory uses the wings of lepidopteran insects as the main target of morphological studies.

### I. Wing outline shape formed by cell death

The wings of the lepidopteran insects (butterflies and moths) develop from the wing imaginal disc, which is a hollow sac made of simple epithelium. Due to its simple construction, this tissue is a good material for studying cellular interactions in the course of morphogenesis.

The outline shape of the adult wing is often different from that of the pupal wing. This difference is brought about by the programmed cell death of the marginal area of the pupal wing. The marginal dying area is called “the degeneration region” and the internal area which develops into the adult wing is called “the differentiation region”.

Cell death in the degeneration region proceeds very rapidly and finishes in a half-day to one-day period in *Pieris rapae* and in several other examined species. It was shown that the dying cells in the degeneration region have characteristics in common with the apoptotic cell death in mammalian cells, such as fragmented and condensed nuclei containing short DNA fragments detected by TUNEL staining. The cells in the degeneration region are actively engulfed by the macrophages in the cavity beneath the wing epithelium. The macrophages seem to be concentrated beneath the degeneration region by the strong adhesion between the basal surfaces of the dorsal and ventral epithelium in the differentiation region, which occurs at the time of prominent cell death and excludes macrophages from the differentiation region. Thus realized concentration of macrophages seems to accelerate the shrinkage of the degeneration region.

A possible physiological role of the cell degeneration at the wing margin is to make space for the growth of the marginal scales. Marginal scales are extremely elongated scales that grow densely on the edge of the wing. These scales are considered to be important in stabilizing the turbulence occurring posterior to the wing. The movements of the marginal scales are closely monitored by sensory scales and bristles growing among them (Yoshida and Emoto, Zool. Sci. 28, 430-437, 2011).

Trachea and tracheoles are both important in delivering air into the wing and their patterns coincide with that of the boundary of degeneration and differentiation zones at the distal end of the wing. According to previous observations,

the pattern formation of wing epithelium is often dependent on tracheal and tracheole patterns. Basic research on the development of tracheal pattern formation is being done by scanning and transmission electron microscopy to describe the exact pathway and time course of the formation of the elaborate pattern of trachea and tracheoles and to establish the cytological and developmental relationship between the formation of the tracheal pattern and the epithelial cell pattern.

### II. Wing morphogenesis and the growth of marginal scales in small moths

In the small moths which have very long scales along their wing margins, the cuticle of the pupal wings does not appear to be large enough to house these scales. We examined the developmental process of the pupal wings of three species of the small gelechiid moths and found that, concomitant with the programmed cell deaths at the wing margin, there occurs a shrinkage of the differentiation region which has not been observed in large winged butterflies and moths. The shrinkage concomitant with the cell deaths causes the space between the pupal wing and the cuticle to extend, which appears to contribute to the growth of long marginal scales.

Microscopic observation of the long marginal scales revealed that they have a novel morphology and we are studying the effect this morphology may have on the mechanical interaction between marginal scales and further on the air flow over the wing during flight.



Figure 1. Adult specimen of *Metzneria lappella*, one of the gelechiid moths examined, with long scales along the wing margin.

### III. Other research activities

This laboratory also conducts morphological observation of various animal tissues by scanning and transmission electron microscopy and immuno-electron microscopic analyses in collaboration with other laboratories of NIBB. Training in specimen preparation and instrument operation for such observations is also given.