

## Subtribes Malaxidinae and Liparidinae (Orchidaceae, Malaxideae) – taxonomic divergency as effect of pollination isolation

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**Abstract** – Fundamental for the process of divergence within tribe Malaxideae (especially between subtribes Malaxidinae and Liparidinae) are: different landing places for visiting insects on flower, insect visiting position in relation to the gynostemium. Consequence of the differences is another place of pollinia deposition on a pollinator body. It preventing effective pollination of flowers of one subtribe by insects effectively pollinating the flowers of the other subtribe.

**Key words:** Liparidinae, Malaxideae, Malaxidinae, Orchidales, taxonomic divergency, pollination isolation

### INTRODUCTION

Tribe Malaxideae with its over 1700 taxa (including synonyms) is undergoing an extremely active phase of speciation as is evidenced by e.g. a high degree of morphological variability (inclusive of ontogenetic variation) (Margońska *et al.*, 2013).

Crucial in the process of divergence of tribe Malaxideae (subtribes Malaxidinae and Liparidinae) are differences in the landing places for insects on flower and the visiting position of their pollinators in relation to the gynostemium. All this results in a different place of deposition of pollinia on a pollinator body. The increase in the specialization of the floral structures form therefore promotes reproductive isolation between representatives of the subtribes.

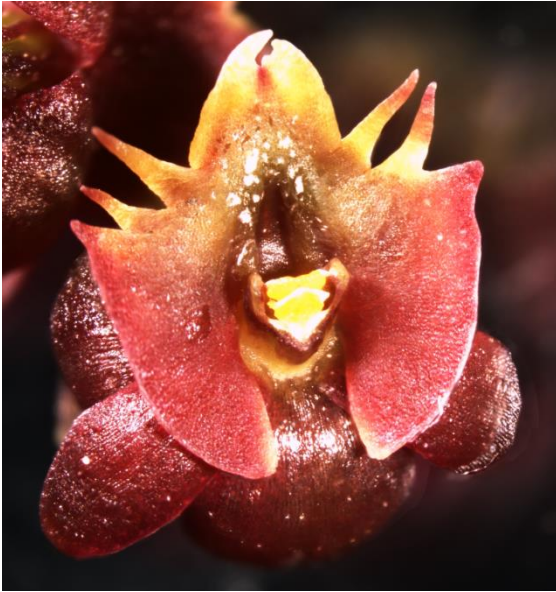
### MATERIALS AND METHODS

The object of the studies was representatives of both subtribes Malaxidinae and Liparidinae (Malaxideae). Observation of pollination strategy, anthesis of the orchids etc. were conducted in natural stands (*in situ*) and in glasshouses conditions (*ex situ*). Researches were performed on the basis of preserved (dried, conserved in Copenhagen mixture etc.) and live materials. Flowers structures examination was carried out with using

standard morphological and anatomical observation (light stereoscopic microscope), scanning (SEM) and transmission electron microscopy (TEM) analysis, also cytochemical tests (CYTO). Olfactory and secretory emissions were also subjected to chemical analysis.

### RESULTS

Subtribe **Malaxidinae** (“*Malaxeae*” Benth. & Hook.f., Gen. Pl. 3: 463, 465. 1883.) comprises species with flowers 360° resupinate, lip directed up (except only *Micr. monophyllos* subsp. *brachypoda*, *Micr. muscifera* (Lindl.) subsp. *stelostachya* (Tang&Wang) Marg., *Micr. yunnanensis* and *Tamayorkis*). Lip is parallel to gynostemium, with a distinctly reduced hypochile, while the epichile can be 3-lobed (middle and 2 lateral lobes) or 1-lobed. Epichile contains differently formed and ornamented 2-3 chambered (never globular) concavity. Gynostemium is column short up to 2-3 times as long as the anther. Anther is erect, parallel to the column and stigma (except only the mountain genus *Tamayorkis*), its locules opening ventrally or apically (never laterally). Stigma opening apically and situated inside a deep pocket. All mentioned morphological characters of Malaxidinae flowers make gynostemium and/or partly dorsal sepal the place of pollinating insects landing (Figure 1).



**Figure 1.** *Crepidium hoi*, flower: pollinia will deposited dorsally at pollinator body.

Subtribe **Liparidinae** (“*Liparidae*” Lindl. ex Miq., Fl. Ind. Bat., 3: 618, 621. 1855. emend Margońska *et al.* 2012 (2013)) flowers are 180° resupinate with lip directed down (except of hanging down epiphytes such as *Alatiliparis*, *Platystyliparis* and *Crossoglossa*). Lip is distinctly divided on the well-developed hypochile and epichile. Nectary are present usually in a form of smooth area around the lip base and/or its basal callus/calli/lamellae/globular structure (*Alatiliparis*, *Disticholiparis*, *Platystyliparis*) if they exist. Well visible is usually darker coloured and shiny stripe (central thickening, sometimes called as pseudonectary) reaching from the lip base to distal part of lip epichile. Gynostemium column is elongated, from 2-3 times or more as long as the anther (except *Crossoglossa* and *Crossoliparis* where the gynostemium length is similar to the anther length). Anther is always orthogonal to the column, staminodes and stigma and its locules opening ventrally or laterally (never apically). Stigma opening ventrally (inside a deep concavity not a pocket). The most exposed element of the flower is here epichile of the lip which is landing place for pollinating insect (Figure 2).

Additionally within the Malaxideae the nectar, if any, is secreted in very limited amounts, which probably forces the insects to visit many flowers before they satisfy their hunger – constituting an undeniable advantage for the orchids.



**Figure 2.** *Liparis (Stichorkis) crenulata*, flower: pollinia will deposited ventrally at pollinator body.

## DISCUSSION AND CONCLUSION

The concavity of Malaxidinae seems to lure insect by imitating the secrecy of attractants. Its epidermis for example is smooth and shiny, simulating the presence of a sticky liquid. Little amount of very minute droplets of secrecy was observed only above of the concavity border. The concavity and its surrounding is available for visiting insects only after landing on just gynostemium and/or partly on dorsal sepal. Reaching for the concavity, the insect from above must force itself between the concavity and the gynostemium, its position exactly coinciding with the apically opening pocket-like stigma. In this way the insect can deposit on the stigma the pollinia if have brought from a previously visited flower. When the insect withdraws, it unhooks the anther, which, upon bending, attaches the pollinia to the underside of the animal.

Within Liparidinae flowers the secretory area, if its presence was confirmed, is located below the lip basal callus/calli/lamellae/globule. The structure of the epidermis cells of the lip's suggests possibility of the dripping/exudation of the secretions towards the central part of the lip, than in the direction of central thickening and finally down the lip. Liparidinae secretory area is reachable for visiting insects after landing on epichil and following along the central thickening as nectar-guide. The insect must force itself

between the canalculated hypochile and the gynostemium, its position exactly coinciding with the ventrally opening concaved stigma. In this way the insect can deposit on the stigma the pollinia if have brought from a another flower. When the insect withdraws, it unhooks the anther, which, upon bending, attaches the pollinia to the back of the animal.

Therefore, the probability of crossed effective pollination between representatives of both subtribes becomes impossible.

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