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Confocal laser scanning microscopy technique for the study of internal genitalia and external morphology of eriophyoid mites (Acari: Eriophyoidea)

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Abstract

Confocal laser scanning microscopy (CLSM) is a modern powerful technique that can be used for studying the external and internal anatomy of arthropods. CLSM has seldom been used in acarology and very rarely for studying eriophyoid mites. It allows the capture of precise digital images of the fine details of external and internal chitinous structures, which can be further analysed using various computer programs. CLSM can serve as an effective tool for comparing closely related and/or cryptic species, correcting diagnoses of poorly described taxa, studying immature instars, and particularly, for studying the structures and the functioning of the internal genitalia of adult females and males. In this paper, the potential use of CLSM for the study of eriophyoids is demonstrated using specimens of 13 mite species and eight genera from the families Phytoptidae Murray 1877 and Eriophyidae Nalepa 1898. This study showed that freshly mounted specimens on microscope slides appeared to be the most appropriate for CLSM as older specimens tended to have reduced autofluorescence. The best choice for studying the external morphology and internal genital apparatus of eriophyoid mites appeared to be the blue laser. Green and light blue wavelengths (488 nm and 532 nm) were found to be less useful. The quality of CLSM images depended on the slide-mounting medium used. Among those compared, Hoyer's medium was found to be the most appropriate whereas Heinze medium and media including Iodium gave poorer results. The empodia and proximal parts of setae were shown to have very weak autofluorescence signals, but they reflected red (635 nm) and blue (405 nm) laser light, which could be detected with CLSM.

Key words: CLSM, apodemes, spermatheca, Phytoptidae, Eriophyidae

Introduction

Eriophyoid mites are minute and highly specialized plant-feeding arthropods. In comparison to other groups of acariform mites, they have very simplified and relatively homogeneous morphologies that create serious difficulties for species identification and phylogenetic analysis. The set of characters used in eriophyoid mite taxonomy may seem extensive, but includes only a few anatomical structures that are undoubtedly apomorphic. At the same time, the anatomy of eriophyoid mites remains poorly understood. Detailed comparative studies of the structure of both the genital apparatus and gnathosoma would help to find new characters that are useful for eriophyoid systematics (C. Craemer 2010 *PhD thesis*; J. Amrine & E. Lindquist *pers. comm.* Oct. 2011 & Feb. 2012, respectively).

According to the last published classification of Eriophyoidea Nalepa 1898, this superfamily includes 3 families (Amrine *et al*. 2003). Lindquist (1996a p. 320) concluded that only one of them (Diptilomiopidae Keifer 1944) may be monophyletic. The family Eriophyidae includes evolutionary derived forms, whereas the mites possessing putative plesiomorphies (*ve, vi, c1, φ*) have been combined into the family Phytoptidae Murray 1877 (Amrine *et al*. 2003; Chetverikov *et al.* 2009). Towards the end of the twentieth century, a series of studies on phytoptid mites was initiated by a group of Russian acarologists led by Dr. V. G. Shevchenko (Shevchenko *et al.* 1991; Sukhareva 1994; Bagnjuk *et al.* 1995, 1998). Dr. Shevchenko's legacy has been carried on by his students and colleagues despite his retirement and death (Chetverikov *et al.* 2009; Chetverikov & Sukhareva 2009;

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