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Characterization and Taxonomy of Entomogenous Fungi, *Cordyceps* and Its Allies Isolated in Korea

Sung Hee Nam

Department of Agricultural Biology, National Institute of Agricultural Science Technology, RDA, Suwon 441-400, Korea.

Morphological characteristics of *Cordyceps* species and its allies collected in Korea were clarified and their phylogenetic relationships were also analyzed based on molecular data and the study further accomplished the artificial culture system for *Paecilomyces tenuipes.*

Through the survey conducted from June 1999 to October 2002 in 19 mountains in Korea, 667 samples of entomogenous fungi were collected. *Cordyceps* and its allies of 17 species of 5 genera were identified as *Cordyceps gracilioides, C. japonica, C. longissima, C. martialis, C. militaris, C. myrmecophila, C. nutans, C. pruinosa, C. sphecocephala, C. tricentri, Hirsutella nutans, Paecilomyces cicadae, P. farinosus, P. tenuipes, Paecilomyces* sp., *Shimizuomyces paradoxa, Tilachlidiopsis nigra.* The fungi with insect hosts have been collected mainly in the place of shade or mosses near brooks and streams that had high humidity. Overall the frequency of fungal infection in natural ecosystem was relatively low as few as 10 collections per each species. However, many species were found in terms of the few number of collection sites with seasonal limitations. Occurrence of the fungi in Jeju island remote from inland of the Korean peninsula was diverse in their species due to the varied weather of vertical distribution following the altitude. Three most common species were *C. nutans, P. tenuipes* and *C. militaris,* mainly found early in August when the relative humidity and temperature were high, of which *C. nutans* occupied the highest frequency consisting of 65% in total collections. Neither variation in ascomata arrangement in stromata nor development of secondary spores was recognizable, while the number, shape and colour of stromata varied with insect hosts and weather conditions.

ITS 1, ITS 2 and 5.8 rDNA regions amplified from 21 isolates produced a total of 468 to 498 base pairs. The delimitation of genera, *Cordyceps, Beauveria*, and *Paecilomyces* were inferred, but intraspecific differentiation was not achieved. Absence of variation in different collections of *C militaris, C. sinensis, B. bassiana, P. tenuipes* from different locality implied that environmental factors did not affect the genetic variety. The sequences of *C. militaris* showed only 81~83% similarity with those of the same generic species, *C. sinensis,* whereas 91% similarity was shared with those of *B. bassiana.* The results suggested that the genus *Cordyceps* may be the teleomorphic counterpart of mitosporic genus, *Beauveria.* Molecular technique proved to be useful in searching interspecific relationship, finding teleomorph/ anamorphic connection and classifying species of entomopathogenic fungi.

The growth of *P. tenuipes* in SLA-A medium showed two times than in common solid media and it was assumed that the improvement of nutrients which are analogous to the component of insect hosts was effective. The SL medium produced the hyphae as much as 12.2±0.88 mg in dried weight being the highest amount out of 11 different media. For the efficiency of culture, homogenization of 150 rpm with 6 ㎜ bead of 6 g resulted in the highest production in dried weight of 1.32 g/100 ㎖. The productivity of dried mycelial weight in SL medium reached to the highest of 1.360±0.067 g/100 ㎖ with addition of 0.24% silkworm powder, while it decreased above 0.48% of the concentration. Silkworm powder might be useful for the efficient cultivation of other strain of entomogenous fungi. Optimal conidial production condition in hulled rice medium was at pH 5.0, 20℃ following preculture at 24℃ for 7 days, which produced the conidia as much as $4.30 \pm 0.35 \times 10^8$ conidia/㎖. The alternative temperature at 20℃ lead the inside temperature of culture flask to the optimum level of 23~24℃ for the conidial germination due to the generation of heat through the course of conidial germination. Synnematal development of *P tenuipes* was sensitive to light and temperature and its optimal condition was in 300 lux at 20℃, while below 15℃ it similarly developed to stromata without ascomata. Although the teleomorphic state of *P. tenuipes* was known as *C. takaomontana*, there have been no reports on producing teleomorphs in artificial cultivation. Therefore, the findings in this study are expected to be applicable to induce the teleomorphic state as well as to resolve the taxonomic uncertainty.