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The application of the entomopathogenic fungus *Akanthomyces muscarius* modified GFP to study endophytization

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Entomopathogenic fungi (EPF) of the genus *Akanthomyces* (formerly *Lecanicillium*) are one of the most common and important fungal entomopathogens, infecting sucking insects of the order Hemiptera mainly. The fungi can also parasitize on phytopathogenic fungi (rust, powdery mildew). The entomopathogens from these genera reported as endophytes in various plants under natural conditions [1–2], contributing to an increase in plant immunity to pathogens, as well as a decrease in plant colonization by pests. Endophytic colonization of plants by the fungus *Akanthomyces lecanii* can suppress the growth of the peach aphid [3]. *Akanthomyces muscarius* strains caused the death of moth when feeding on cabbage colonized by the fungus [4]. Endophytic properties were assessed using the *A. muscarius* (= *Lecanicillium muscarium*) strain Vl 72-GFP fluorescently labeled with GFP [5]. The transformation was done by electroporation of germinated conidia of the high-virulent "wild" strain Vl 72 by the pBARGPE1 vector harboring an eGFP gene, showed an expression of fluorescent protein without affecting fungal growth and virulence. The influence of the fungus on the growth rates of beans was revealed when leaves, sterile soil and seeds were treated with a suspension of conidia of 10^8 spores/ml. On the 7th day, stimulation of the growth of the stems and roots of the beans was observed when the seeds were soaked in a spore suspension of the fungus. When spraying the leaves, only the stem's elongation was observed. The studied strain colonizes beans irregularly. When treating the seeds, the fungus was isolated in greater quantities from the roots (26%), when spraying the leaves — from the stem (36%), when watering the soil — also from the stem (43%). Infection of *A. muscarius* plants by spilling the soil was most effective. No effect of endophytization was found on the number of aphids after 14 days of aphid plant colonization. As a result of the introduction of the spores of Vl 72-GFP strain by shedding the soil under flower crops (lantana, gerbera, acanthus) in the greenhouse of Saint Petersburg Botanical Garden, this strain was isolated from the leaves of the *Acanthus mollis* L. after one month, which confirms the ability of this species to endophytic colonization of plants in greenhouse conditions. Analysis of hyphae Vl 72-GFP in the plant performed on an AxioImager M1 fluorescent microscope demonstrated the same level of fluorescence as in *A. muscarius* hyphae growing on the media.

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Keywords: *Akanthomyces muscarius*; endophytization; GFP.

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