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Identification of Some Mycromycetes Aiming at Collection Development

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Abstract: ***The collection of microorganisms is a background for improvement in disease diagnosis, and testing of bacterial antagonists intended for biocontrol and protection of plants. Thorough study on phenotypic properties, including pathogenicity, virulence, and genetic diversity of the essential cultures, is necessary for identification of disease resistant farm crops breeds and lines.***

Keywords: Trichoderma viride, Alternaria infectoria, Cladosporium carpophilum, plants, phytopathogens

1. Introduction

Presenting not only cultures for research, but also considerable information content for intended use of specific types and strains, collections of microorganisms with various purposefulness, including pathogenic microorganisms, are in the focus of many studies.

In the course of human development, plant diseases affected quality of our life in a variety of ways. Novel technologies and methods allowed reducing harmfulness of plant diseases; international trade and migration facilitated global spread of pathogens.

Collections of cultures are keepers for microbial resources for science and society. This is to be the significant contribution to progress of up-to-date and future basic and applied, medical and biotechnological studies. In 2009 12th session of CGFRA, Commission of Plant Genetic Resources for Food and Agriculture in Rome, recognized importance of microorganisms in food security and stable farming. Provision of services to food production section and farming is one of requirements outlined in the guidelines for development and function of microorganism culture collections devised by the World Federation for Culture Collections (WFCC).

Exclusive Collection of Phytopathogenic Microorganisms created in 1993 at the initiative of Professor Khasanov B.A. in the frames of grants from the International Soros Foundation and USAID, UN, USDA/ARS projects, collects, studies and maintains cultures of phytopathogens. In 2009 Unique State scientific Object status was assigned to the Collection.

In January 2005 the Collection was registered in WDCM, the WFCC World Data Center for Microorganisms with the number 862.

2. Materials and Methods

Samples of agricultural plants, such as cotton, wheat, tomatoes, potatoes, aubergine; those of conifer branches, such as spruce, juniper with signs of diseases, as well as collection cultures of phytopathogens for conservation and reprocessing, were materials for the study.

The study used classical approaches described elsewhere [1, 7]. The media were prepared in compliance with classical prescriptions [2, 4, 7]. The mycetic taxonomy was determined according to appropriate field guides [3, 5, 7, 8]. Presently, there are 951 cultures of microorganisms of 44 genera, 146 of those prepared by cool dehumidification, having scientific, medical, agricultural, industrial, ecological, taxonomic and educational values, aiming at preservation of biodiversity.

146 cultures prepared by cool dehumidification in 1993-2002 are kept in ampules, each object being stored in 5-15 ampules, in 2 ampules in individual cases. 35 cultures are temporarily stored in glass tubes by reinoculation. 860 cultures are preserved under mineral medicinal oil in glass tubes with cotton-gauze stoppers in the racks under serial numbers, in household refrigerators at +4-7°C; 139 of 146 lyophilized cultures are stored under mineral medicinal oil layer.

Annually, major efforts are made in preservation and reprocessing of the cultures. To control survivability and reproduction of gene pool in the collection of microbiological cultures visual inspection of 100 cultures in 2-3 tubes of each culture, 200-300 tubes with cultures in total, of those under mineral oil, has been performed. 120 cultures kept under oil have been reprocessed with preliminary plating on semisynthetic media.

The collection is constantly restocked. Thus, 59, 102, 92, 94 and 150 pure cultures of microfungi were placed in 2012, 2013, 2014, 2015 and 2016 years, respectively.

Pure cultures of an apple scab agent, *Cladosporium carpophilum* (Thiime) Oud. (*Venturia carpophila* Fisch teleomorph) belonging to *Hyphomycetales*, *Cladosporium oxysporum* order, were isolated in our region for the first time. Rare microfungi, such as *Cylindrocarpon mali* (All.) Wollenw. Fide Wollenw. (*Nectria galligena),* apple tree cancer (Nectria canker) agent, *Papularia sphaeroesperma* (Person) v. Hohnol, a saprotroph, *Alternaria infectoria*, affecting apple-tress and discharging toxins, were isolated.

For the first time, in Uzbekistan seven cultures of endophytic fungi, aulophytes, considered as potential resources of biological growth stimulation of new generation

were isolated from saksaul (*Haloxylon aphyllum*) and two types of tamarisk (*Tamarix hispida* and *T. ramossisima*) growing at Ustyurt plateau of the southern Aral Sea newly formed dry land. Among the newly identified microorganisms there are *Alternaria tenuissima* (*Kunze*) *Wiltshire, Trichoderma viride Pers., Ulocladium consortiale* (*Thiim*) *E.G. Simmons, Acremonium sp., Chaetomium sp., Stemphylium sp., Alternaria sp*. and others (Fig. 1,2).

Figure 1: *Trichoderma viride* fungus

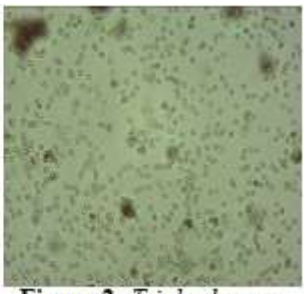

Figure 2: *Trichoderma viride* conidia (x600)

Among advances worth mentioning there are, industrial killer strains, potential agents for adverse vegetation's biocontrol, such as globally known *Pleospora papaveracea*, *Ascochyta chenopodii* Rostr., *Pyrenophora semeniperda* (Brittle.et *Adam*) Shoem,

Biological defense technologies *Trihoderma viridi, T.harzianum.*

Cultures to test disease resistance of essential farm crops, *Verticillium dahliae* Kleb.; *F. oxysporum* Schl. f.sp. *vasinfectum* (Atk.) Snyder & Hansen*; Xanthomonas camhestris* pv. *malvacearum; Rhizoctonia solani Kuhn.*; *Cochliobolus sativus* (Ito et Kurib. Drechs. Ex Dastur.; *F. graminearum* Schwabe.; *Oospora parasitica* and others.

Rare microorganisms, such as, *Nectria radicicola* Gerlach et Nilsson (anam. *Cylindrocarpon didymum* (Hart.) Wr. Boot.), cylindrocarpon destructive; *Rosellinia necatrix* (R. Hartig) Berl; (anam. *Dematophora necatrix* Hart.), dematophora pernicious; *Geotrichum candidum* Lk emend J. Carm.; *Alternaria yaponica* (*raphani).*

Figure 3: 1-healthy plant, 2-affected plant

Figure 4: *F. Oxysporum* fungus

Figure 5: *F.oxysporum conidia (x1000)*

Figure 6: *F.oxysporum mycelium (x1000)*

Pure cultures for agent of blotch of stone fruits, *Cladosporium carpophilum* (Thiime) Oud. (*Venturia carpophila* Fisch teleomorph), belonging to *Hyphomycetales* order, agent of apple-tree pink rot, - *Acladium conspersum, Acladium* Link (1809), *Botryobasidiaceae*, agent of apple-tree trunk rot, a pileate fungus, *Pleurotus pomeli*, were isolated in our region for the first time.

Available microbial genetic resources are unique; if lost, their restoration by repeated isolation form populations is associated with monumental challenges or practically impossible.

The collection of microorganisms is a background for improvement in disease diagnosis, and testing of bacterial antagonists intended for biocontrol and protection of plants. Thorough study on phenotypic properties, including pathogenicity, virulence, and genetic diversity of the essential cultures, is necessary for identification of disease resistant farm crops breeds and lines.

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