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Short Communication

# Combined microbiological approach to screening of producers of proteases with hemostasis system proteins activity among micromycetes

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Highlights:

- Most of micromycetes are produced proteolytic enzymes which can affect human hemostasis proteins
- For determination of such activity special chromogenic peptide substrates are used
- Proteases secretion by micromycetes depends on medium composition

## Abstract

A scheme for screening of micromycetes - producers of proteases with the activity of hemostasis system proteins, based on their enzymatic indices determination (1) and the activity towards chromogenic peptide substrates for proteins of the hemostasis system (2) was developed. Depending on the ability of proteases producers to cleave such substrates, an enzymatic reaction in conditions containing human plasma is suggested, which makes it possible to identify the potentiality of the target plasma hemostasis proenzymes activation.

## Introduction

One of the main challenges of modern medicine is the diagnosis and treatment of thrombotic complications. Various thrombolytic agents, which are used in therapy, are proteolytic enzymes with the activity similar to the one of hemostasis system proteases (1). Such activity was also found in proteases present in venom of snakes and in cultures of microorganisms. The preparations obtained on their basis are used as a part of diagnostic kits and as therapeutic agents for the detection and treatment of such complications (2-5). Extracellular proteolytic enzymes of micromycetes from different ecological groups have the ability to act on hemostasis system proteins as activators (triggering procoagulant and anticoagulant processes) or direct fibrinolytic agents (breaking down already formed thrombi) (4, 6). But there are no unified ways to determine their ability to produce such proteases.

## Materials and Methods

Micromycetes for screening were supplied by the Department of Microbiology, Moscow State University. The strains were maintained in tubes with slant agar. Agar-plate cultivation of the fungi was carried out on Czapek's media with 1% casein or fibrin addition as the single source of nitrogen. After 5 days of cultivation at 28°C, 5 ml of 0.08% Coomassie brilliant blue