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SHORT COMMUNICATION

Isolation and molecular identification of actinomycete microflora, of some saharian soils of south east Algeria (Biskra, EL-Oued and Ourgla) study of antifungal activity of isolated strains

Isolement et identification moléculaire d'actinomycètes de quelques sols sahariens du Sud-est de l'Algérie (Biskra, El-oued et Ourgla). Etude de l'activité antifongique des souches isolées

A. Boudemagh ^{a,b,*}, M. Kitouni ^{a,b}, F. Boughachiche ^a, H. Hamdiken ^a, L. Oulmi ^a, S. Reghioua ^a, H. Zerizer ^a, A. Couble ^b, D. Mouniee ^b, A. Boulahrouf ^a, P. Boiron ^b

^a *Laboratoire de Microbiologie Appliquée, Département des Sciences de la Nature et de la Vie, Université Mentouri, Constantine, Algeria*

^b *UMR CNRS 5557, Ecologie Microbienne (Center for Microbial Ecology), Groupe de Recherche « Pathogènes opportunistes et environnement », Laboratoire de Mycologie Fondamentale et Appliquée aux Biotechnologies Industrielles, Faculté de Pharmacie, Université Claude Bernard Lyon 1, 8, avenue Rockefeller, 69373 Lyon cedex 8, France*

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Abstract As part of a research program whose aim is to identify new antifungal metabolites from rare actinomycetes, three Saharan soils from south east Algeria were analyzed. Twenty-seven (27) strains of actinomycetes were isolated and tested for their antifungal activity. The soil from the region of Biskra gave the highest number of actinomycetes, i.e. 52% versus 18% and 30% for the soils from EL-Oued and Ourgla, respectively. The results of this study showed the GLM medium to be the most favorable for the isolation of actinomycetes from these ecosystems, on its own providing 17 strains out of the total number of actinomycetes isolated. Two strains presented very important antifungal activity against most of the filamentous fungi and test yeasts used. Molecular identification by polymerase chain reaction using universal 16S rDNA primers allowed the two active strains to be classified in the genus *Streptomyces*.

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* Corresponding author.

MOTS CLÉS

Activité antifongique ;
Actinomycète ;
Sol du Sahara ;
Identification
moléculaire ;
Amorce universelle

Résumé Lors d'un programme de recherche de nouveaux métabolites antifongiques à partir des actinomycètes rares, trois sols sahariens du sud-est algérien ont été explorés. Vingt-sept (27) souches d'actinomycètes ont été isolées et testées pour leurs activités antifongiques. Le sol de la région de Biskra a donné le plus grand nombre d'actinomycètes soit 52 contre 18 et 30 % des sols d'El-Oued et de Ourgla. D'après cette étude, le milieu GLM est le plus favorable pour l'isolement des actinomycètes à partir de ces écosystèmes, il offre à lui seul 17 souches sur le total des actinomycètes isolés. Deux souches présentent une activité antifongique très importante contre la plupart des champignons filamenteux et levures tests utilisés. L'identification moléculaire par réaction de polymérase en chaîne (PCR), en utilisant des amorces universelles de l'ADNr 16S a permis de classer les deux souches actives dans le genre *Streptomyces*.

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Introduction

The frequency of fungal infections has increased in a dramatic way during the last two decades. This is mainly due to increasing number of immunodepressed patients, such as persons suffering from cancer, infected by the human immunodeficiency virus (HIV) or having undergone a graft of solid organs. In addition, the more and more invasive procedures such as chemotherapy and the graft of hematopoietic strain cells, the excessive use of vascular probes and the extended use of large spectra antibiotics, constitute other predisposing factors for developing a deep mycosis [28]. The emergence of new pathogen fungal agents and the development of resistance are equally important factors [20]. The situation is similar in animal therapy, in agriculture and rearing where fungal diseases are making ravages particularly in developing countries and against which there exist no efficient antifungals or non toxic fungicides, non pollutant and if possible biodegradable. The demands from leather, wood, and textiles as well as food industry to obtain active compounds against mouldiness and little toxic for human beings and animals are increasing [15].

Despite the discovery of numerous synthetic antifungal substances, such as caspofungine and Terbinafine, which are efficient but very expensive, all the best antifungal agents available are fermentation products [20].

The actinomycetes represent the main sources of secondary metabolites with anticellular activity. The specie belonging to the genus *Streptomyces* constitute 50% of the total population of soil actinomycetes and 75% of molecules with antibiotic activity are produced by this genus [21]. The actinomycetal bacteria called rare constitute an important potential source and little explored, of new secondary metabolites with antibacterial and antifungal activity [6,8,11,19,24]. Their selective isolation led us to discover new interesting substances

among the genus *Micromonospora*, *Actinomadura*, and *Streptosporangium* [26]. The screening has always been the main way to reach new antimicrobial molecules despite its reduced efficiency. It is practiced by several laboratories. They tried to diversify the sources of micro-organisms by taking samples coming from the most extreme habitations [11,15-17] and realized methods, which favor the rare specie [8,10].

This work allowed to isolate, select and identify by molecular methods actinomycetes strains and to screen antifungal activity of strains coming from saharian soils of south east Algeria. The agar diffusion method has been realized by means of two test medias (Yeast Malt Agar (YMA) and casitone) to screen antifungal activity of 27 actinomycete strains.

Materials and methods

Samples selection and used strains

The samples collected from several saharian ecosystems of south east Algeria: soil of Ourgla, Biskra and El-Oued. The samples are set apart according to Pochon and Tardieu technic [22]. They are conveyed to the laboratory for analysis as quickly as possible.

The target strains used for screening antifungal activity come from the American Type Culture Collection (ATCC) and the Mycology Unit of the Pasteur Institute (UMIP) and are: *Candida albicans* UMIP 48.72, *C. albicans* UMIP 884.65, *Candida tropicalis* R2 UMIP 1275.81, *Aspergillus fumigatus* UMIP 1082.74, *Aspergillus niger* ATCC 16404 and *Fusarium oxysporum* UMIP 625.72.

All the strains are maintained at 28 °C on Sabouraud's medium. The control strains used and obtained from the collection NRRL of the USDA/Agricultural Research Service (National Center for Agricultural Utilization Research, Microbial proper-

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