The 7th International Electronic Conference on Medicinal Chemistry (ECMC 2021)
01–30 NOVEMBER 2021 | ONLINE

GC-MS profile and antibacterial potency of thermophilic *Bacillus licheniformis* LMB3701 isolated from Dbagh hot spring in Algeria

Nadia AISSAOUI [1]*, Fatima NAS[2], and Nihel Klouche-Khelil [2]

[1] LAMAABE Laboratory Tlemcen University, Algeria;
[2] LAMAABE Laboratory Tlemcen University, Algeria . Laboratory of Experimental Surgery, Dental Surgery Department, Faculty of Medicine, Tlemcen University, Algeria.

•Corresponding author: nada2389@gmail.com

GC-MS profile and antibacterial potency of thermophilic *Bacillus licheniformis* LMB3701 isolated from Dbagh hot spring in Algeria

Graphical Abstract

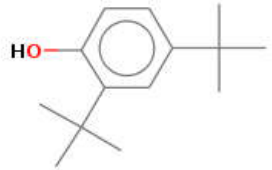
Dbagh's hot spring

Isolation of *B.licheniformis* LMB3701

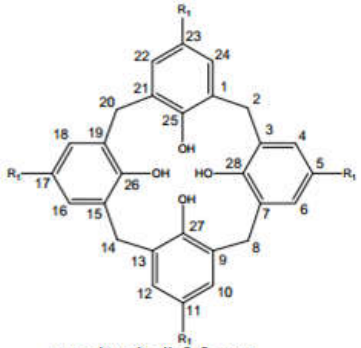
Radial Diffusion Assay

Activity against multidrug resistant bacteria

GC-MS analysis



2,4-di-tert-butylphenool



p-ter-butylcalix[4]arene

Abstract:
Bacillus genus is known as an auspicious reservoir of active compounds with diversity in chemical structure. Although, thermophilic Bacillus and their antimicrobial compounds are little explored. Based on this, the objective of this work is to determine GC-MS profile and evaluate the antibacterial potency of a strain designated as LMB3701 isolated from water of Dbagh's hot spring, Guelma, Northeast, Algeria. The LMB3701 strain was identified as *B.licheniformis* (Accession No.:KX100031) using 16S rRNA gene (98% of similarity). The ethyl acetate extract of thermophilic *B.licheniformis* showed an antibacterial activity against multidrug-resistant *Staphylococcus aureus* NR_075000.1 (6 mm ± 0 mm) and *Pseudomonas aeruginosa* NR_0754828.1 (8 mm ± 0.580 mm) using Radia Diffusion Assay. Five major groups [Polyphenolic macrocycle (34.087 %), Phenol (14.672 %), Stearic acid methyl ester (4.745 %), Fatty alcohol (4.082 %) and triterpene (1.177 %)] were identified using gas chromatography–mass spectrometry (GC-MS). To the best of our knowledge, this is the first report showing production of p-tert-butylcalix[4]arene and 2,4-di-tert-butylphenol as extremolytes compounds from thermophilic *Bacillus* licheniformis at 55 C.
Keywords: Antibacterial Potency; Bacillus licheniformis; Dbagh hot spring; GCMS profile.

Introduction

Thermophilic microorganisms are extensively studied and recognized as a source of secondary metabolites with great structural and functional diversity for biotechnological applications (Selvarajan et al. 2017).

Bacillus genus can isolate in a thermophilic environment and and was recognized to produce many clinically useful antibiotics. However, few studies were conducted on thermophilic *Bacillus* that has the potential to produce secondary compounds with antibacterial properties.

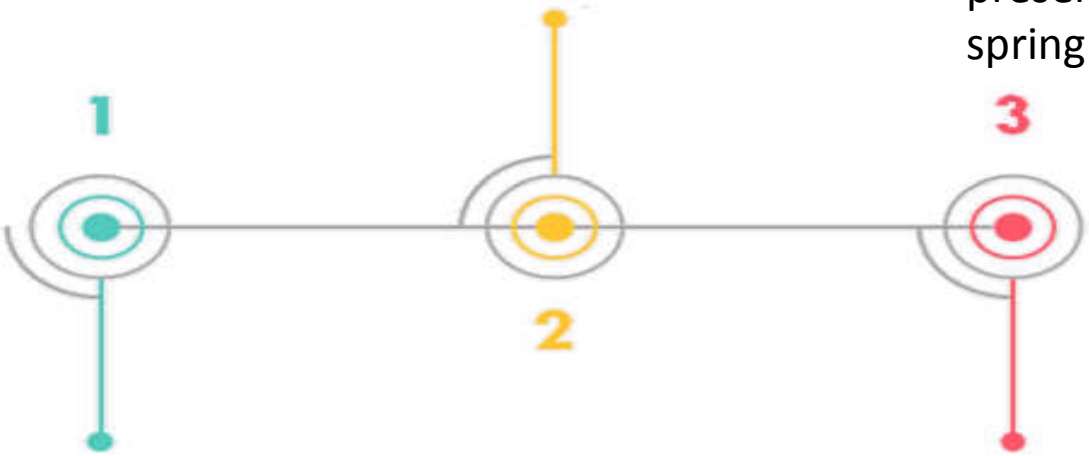
Introduction

The aim of this study was to evaluate the antibacterial potential of thermophilic bacilli isolated from mineral terrestrial hot spring in Algeria against two human pathogenic. GC–MS analysis was used to elucidate a structural and percentages of secondary metabolites. .

Results and discussion

16S rRNA gene identification :

98% of similarity with *B.licheniformis*



✓First identification of this species in Dbagh hot spring;

✓Adiguzel et al. (2011) and Baltaci et al. (2017) reported the presence of this species in the hot spring in Turkey .

Phenotypic characteristics:

Bacillus cluster

Thermophilic strain: optimal temperature growth at 55°C

Strain deposed in GeneBank under Accession No.: **KX100031**

Results and discussion

Table 1: Antibacterial activity of crude extract of LB3701 strain against multidrug resistance bacteria

Microorganisms	Inhibition zone(mm)
P. aeruginosa NR_0754828.1	8 ± 0.580[a]
S. aureus NR_075000.1	6 ± 0[a]

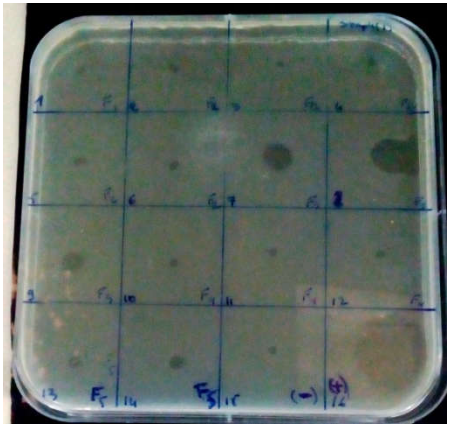


Figure 1: Activity against *S. aureus* NR_075000.1

✓Thermophilic *Bacillus* that has a potential to produce secondary metabolites with antibacterial properties (Mendoet al. 2004) .

Results and discussion

✓p-tert-Butylcalix[4]arene and 2,4-di-tert-butylphenol were present in high percents in crude extract;
✓no reports on the biological synthesis of p-tert-Butylcalix [4]arene;
✓this is the pioneer report linking the potential of thermophilic bacteria to biosynthetize, at 55 C, p-tert-Butylcalix [4]arene and 2, 4 DTBP.

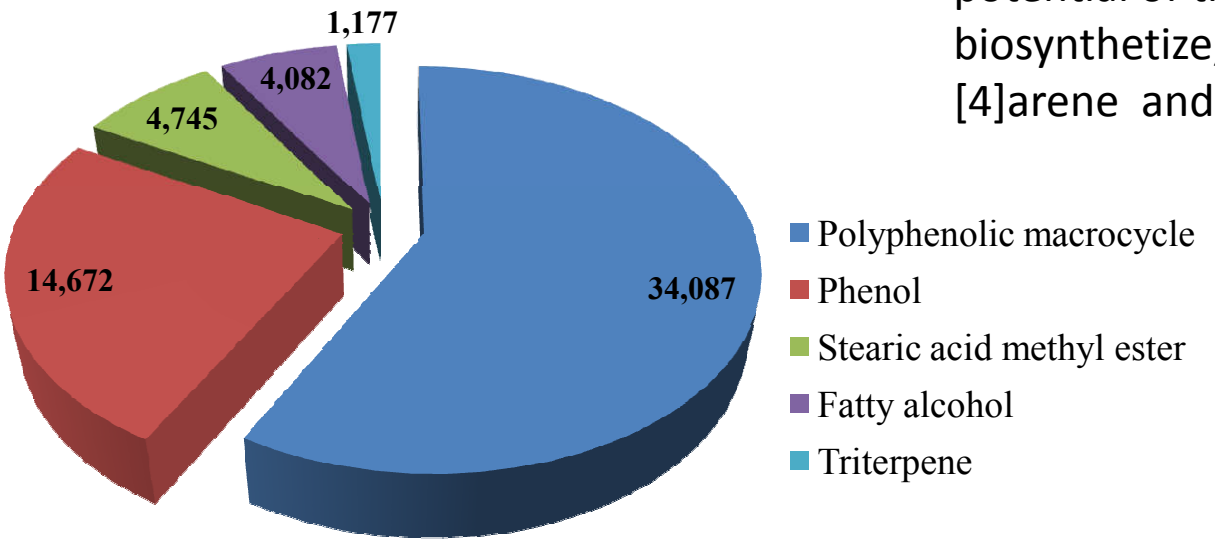


Figure 2: Principals compounds of crude extract of LMB3701 strains based on GC-MS analysis

Conclusions

In this study, we provided insights into the potential of thermophilic *Bacillus* as producers of drugs with efficacy against clinical strains suggested that hot spring is a valuable source of antibacterial compounds.

In addition, we demonstrate the importance of analytical chemistry tools like GC–MS to determine the chemical profile of crude culture extract.

In future investigation, we focuse on purified crudes extracts using HPLC to produce unique molecules used in pharmaceutical and medical industries.

Acknowledgments

Genotypic and GC-MS analysis were realized in Univ. Manouba, ISBST, BVBGR-LR11ES31, Biotechpole Sidi Thabet, 2020, Ariana (Tunisia).