Metabolomics guided the selection of culture conditions for the production of phomactins by the fungus *Biatriospora* sp. CBMAI 1333

LEANDRO S. OLIVEIRA [1], CAMILA M. CRNKOVIC [1,2] , ROBERTO G. S. BERLINCK [1]
[1]Instituto de Química de São Carlos, Universidade de São Paulo
[2]Faculdade de Ciências Farmacêuticas, Universidade de São Paulo
leandro.oliveira10@usp.br / rgsberlinck@iqsc.usp.br

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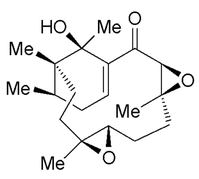
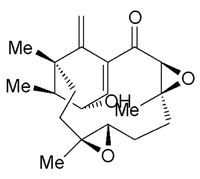
INTRODUCTION

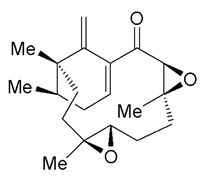
Phomactins are natural diterpene products that display inhibitory activity on the platelet activating factor (PAF) receptor. These compounds reduce the repopulation of tumor cells after radiation with γ rays, an activity that can be exploited as an adjuvant in the treatment of cancers.[1,2] Recently several phomactins were isolated from the growth medium of the marine fungal strain *Biatriospora* sp. CBMAI 1333.[2]



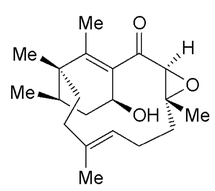
Phomactin I



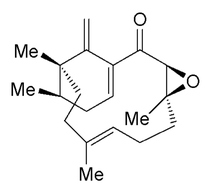
Phomactin F



Phomactin P



Phomactin Q



Phomactin R

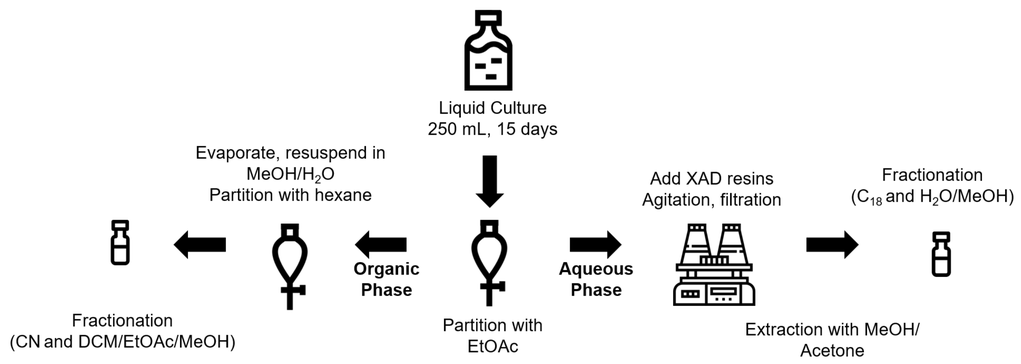
OBJECTIVES

The present investigation aimed to increase the diversity and yield of phomactins produced by the marine fungal strain *Biatriospora* sp. CBMAI 1333.

EXPERIMENTAL SECTION

For the production of phomactins, different culture media and growth conditions were evaluated. Liquid culture media of 2% malt broth (M2), 3% malt broth (M3), minimal medium (MM), glucose peptone yeast extract (GPY) , potato dextrose broth (PDB), sabouraud dextrose (SBD) and sabourad dextrose with yeast extract (SBD + YE) were prepared with distilled and artificial sea water (ASW).

Figure 1: Workflow used to obtain the fractions of Biatriospora sp. CBMAI 1333.



LC-MS analysis: UPLC-HRMS/MS (Waters® Xevo® G2-XS QToF)

Data processing: MSConvert and MZMine 2

Metabolomic analysis: Global Natural Product Social Molecular Networking (GNPS) and XCMS Online.

Data visualization: Gephi and XCMS.

CONCLUSION

Evaluation of different culture media and growth conditions combined with the application of GNPS and XCMS platforms allowed us to increase the diversity and yield of phomactins produced by the marine fungal strain *Biatriospora* sp. CBMAI 1333. A large-scale experiment using the optimized growth conditions led to the isolation of two possible new compounds, which are under identification.

ACKNOWLEDGMENTS

CAPES
Process 88887.372014/2019-00

BIOTA/BIOprospecTA #2013/50228-8

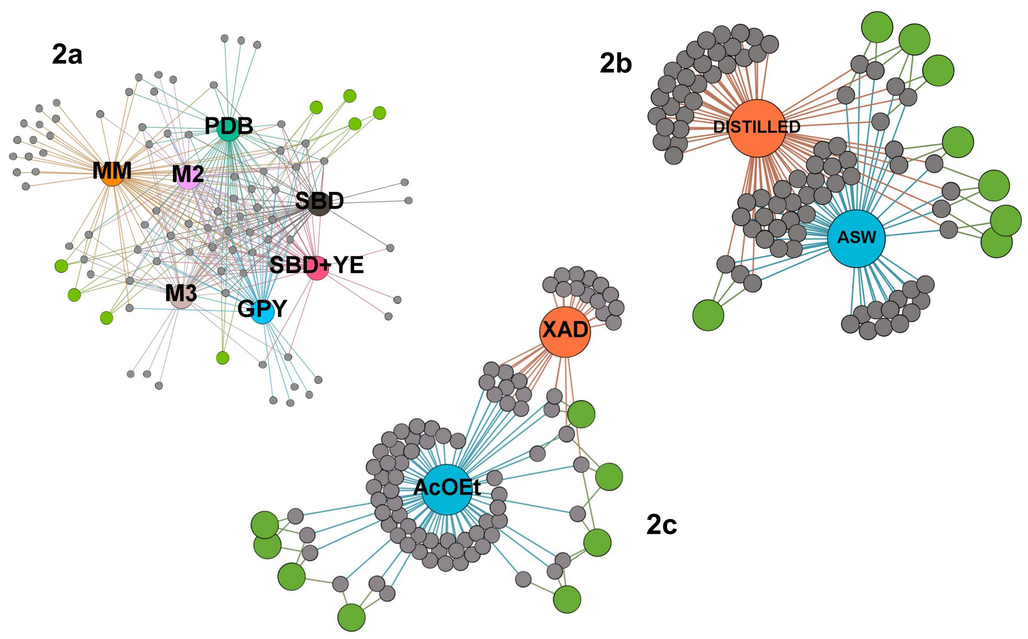
RESULTS and DISCUSSION

GNPS molecular networking was used to identify phomactin-like compounds present in the fractions based on their MS/MS spectral similarity to known phomactin standards. Results were formatted for visualization on Gephi as metabolite association network (Figure 2).

Figure 2a: Diversity induced by different culture media composition.

Figure 2b: Phomactin diversity induced by media prepared with distilled water and artificial sea water (ASW).

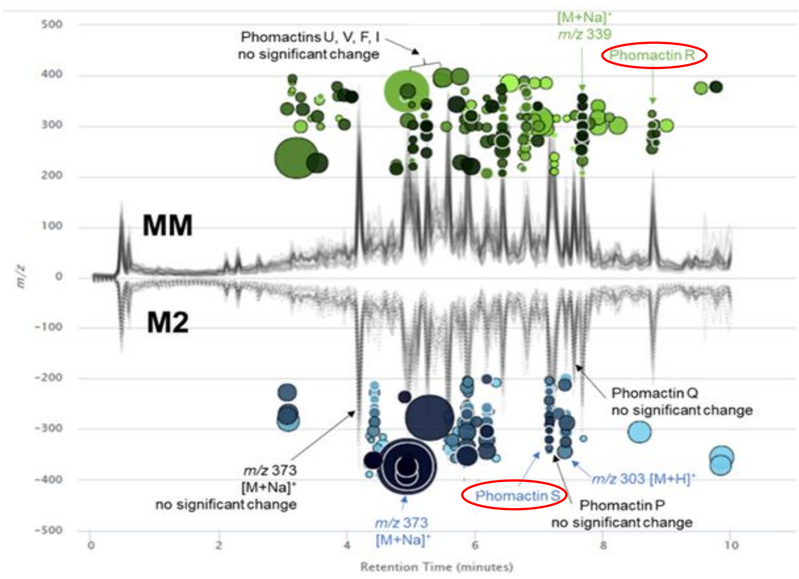
Figure 2c: Phomactin diversity in the organic (AcOEt) and aqueous (XAD) fractions.



MM produced the highest number of unshared nodes (unique features) and it is also the most diverse medium in terms of the content of phomactin-like compounds.

The greatest diversity of known phomactins and possibly new analogues was observed in the organic fraction of the minimal medium (MM) prepared in distilled water. In addition, phomactins were also detected in higher intensities in MM when compared to the previously studied culture medium (M2) as shown in figure 3. Since MM demonstrated to be the best culture medium to increase the diversity and yield of phomactin compounds, it was chosen for large scale experiments using optimized growth conditions. After culture medium extraction, the organic fraction was subjected to several chromatographic separation steps, resulting in the isolation of two possible new compounds, yet not identified.

Figure 3: Comparison between feature intensities in MM and M2 (control) visualized on XCMS.



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