Targeting *Echinococcus granulosus* thioredoxin glutathione reductase using a Dynamic Combinatorial approach.

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INTRODUCTION

Dynamic Combinatorial Chemistry (DCC) is a synthetic tool that merges chemical synthesis with biological tests in one pot.[1] The library is under thermodynamic control and introduction of a template could stabilize a compound through library redistribution *via* reversible bonds. This stabilization could result on an amplification of the best binder.

E. granulosus is a flatworm parasite responsible for cystic hydatid disease. Its thiol redox homeostasis is dependent on an essential single enzyme: thioredoxin glutathione reductase (TGR), which is a key drug target.[2]

RESULTS AND DISCUSSION

Exploring for new *E. granulosus* TGR inhibitors through site-specific DCC, we aimed to generate a thiol-disulfide dynamic library using TGR as a template. The library was prepared using TNB (**a**), commercially available thiols (**b**-**e**) and the synthetic thiol (**f**),[3] to afford 21 theoretical disulfides. Library distribution was compared before and after enzyme introduction. In presence of the template (TGR), a shift in the equilibrium towards a new stabilized compound (**a**S-S**f**) was observed by HPLC-MS.

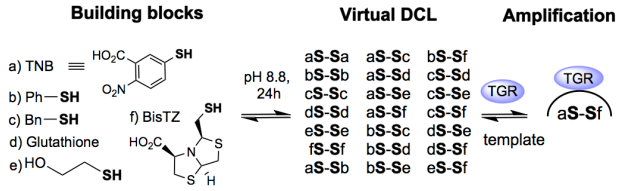
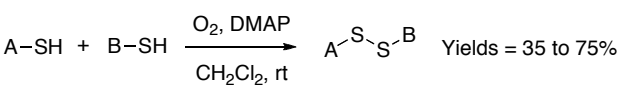


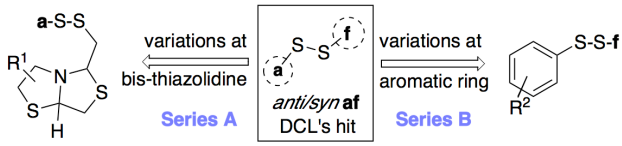
Figure 1. DCL: thiol/disulfide exchange introducing TGR as a template.

Building upon these promising results, we started an independent synthesis of the library-hit **a**S-S**f** and several analogs, see scheme 1.



Scheme 1. General preparation of **A**-S-S-**B** heterodimers.

The analogs were prepared by variations in both thiols: the bisthiazolidine moiety (series A) and the aromatic ring (series B):



The prepared disulfides were screened for TGR inhibition at [Inh] = 30 μM. Compounds with the highest inhibition values were selected for IC_{50} determination (**af**, **ah** and **ai**); see Figure 2.

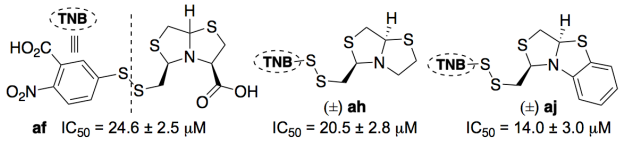


Figure 2. IC_{50} ± sd values of compounds af, ah, and aj.

A structure activity relationship study was performed and the TNB-tricyclic disulfide (**aj**) was identified as the best enzyme inhibitor (IC_{50}= 14 μM).

CONCLUSION

We have demonstrated the use of a thiol-disulfide DCL using *E. granulosus* TGR as a template for the first time. The disulfide (**af**) was identified as a library amplified compound, combining TNB (**a**) and bisthiazolidine (**f**). Independent synthesis of (**af**) and 14 further analogs were carried out for biological evaluation on TGR. Compound (**aj**) presents the best inhibition value: IC_{50} = 14μM.

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