Arachidonic acid is a potential inhibitor of Abc1 transporter of *Candida krusei*

Abdullahi T. Jamiu*, Jacobus Albertyn, Olihile Sebolai, Carolina H. Pohl

Pathogenic Yeast Research Group, Department of Microbiology and Biochemistry, University of the Free State, PO Box 339, Bloemfontein 9300, South Africa
*E-mail: abdullahijamiu45@gmail.com

Introduction

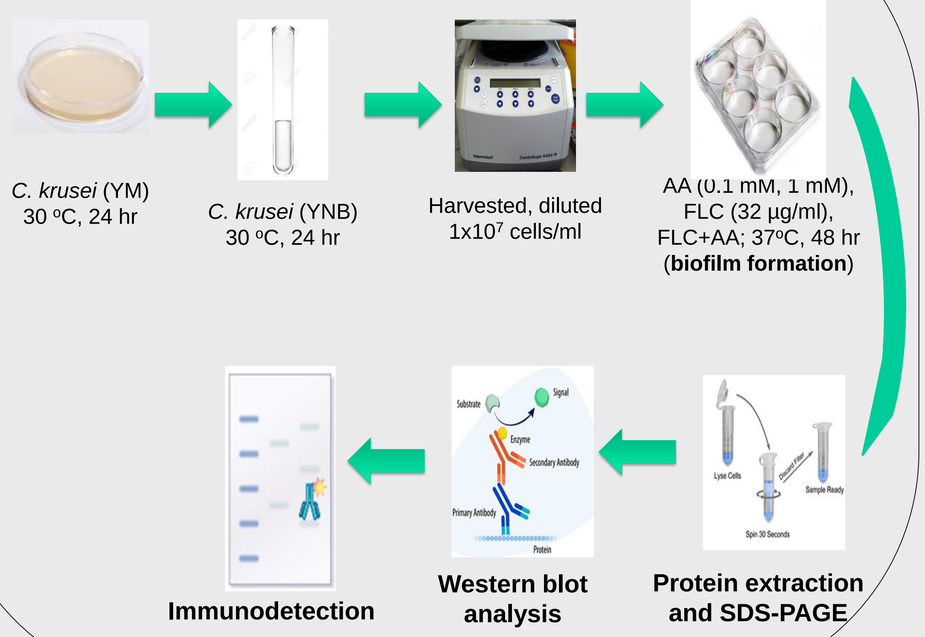
Candida spp. remain the most common cause of invasive mycoses and the number of life-threatening candidiasis has markedly increased over the past decade due to a growing population of immune compromised individuals [1]. *Candida albicans*, no doubt, remains the major cause of candidiasis, however, the epidemiology has changed in recent years with a significant number of infection been attributed to non-*albicans Candida* (NAC) species, including *Candida krusei* [2]. Fluconazole (FLC) is the most widely used antifungal drug for the treatment of candidiasis due to its low toxicity and great efficacy [3]. However, *C. krusei* exhibits intrinsic resistance to FLC with more than 97% isolates displaying resistance [4]. The mechanism for this resistance has been, partly, attributed to constitutive expression of ATP-binding cassette 1 (Abc1p) [5]. However, inhibitors of this transporter are lacking. Polyunsaturated fatty acids (PUFAs, e.g. arachidonic acid (AA) which are known disruptors of microbial cellular membranes might function well as effective inhibitors of Abc1p, since this protein localises in the cell membrane. However, this needs to be investigated.

Aim

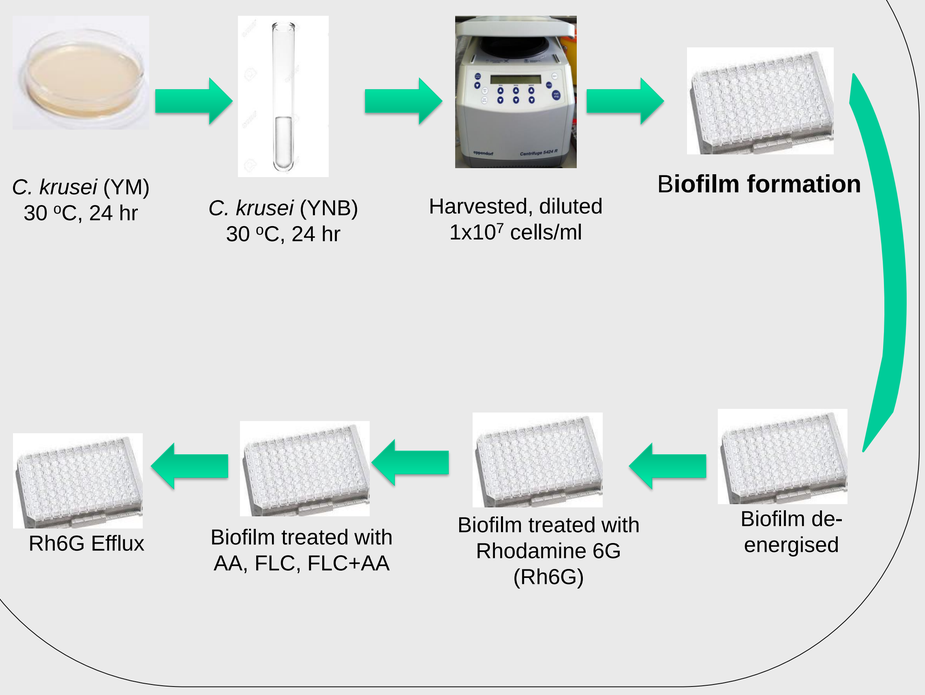
To investigate the influence of AA and FLC on the expression and activity of Abc1p in *C. krusei*

Materials & Methods

Influence of AA on Abc1p expression



Influence of AA on Abc1p activity



Results

Influence of AA and FLC on the expression of Abc1p

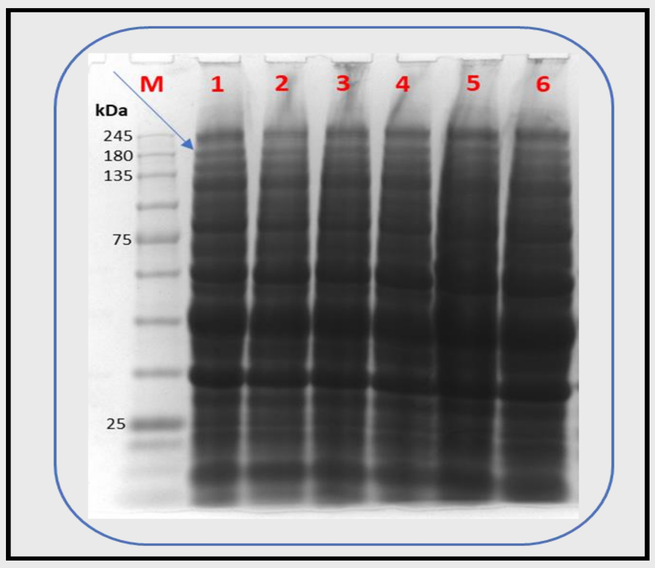


Fig. 1 SDS-PAGE profile of proteins from *Candida krusei* biofilms following exposure to various treatments. **1**: DE (DMSO and ethanol) control, **2**: 32 µg/ml fluconazole (FLC), **3**: 0.1 mM Arachidonic acid (AA), **4**: combination of 0.1 mM AA and FLC, **5**: 1 mM AA, **6**: combination of 1 mM AA and FLC. M represents SDS-PAGE Pre-stained Protein Ladder (3.5 to 245 kDa). Arrow indicates a band corresponding to expected protein size of 172 kDa.

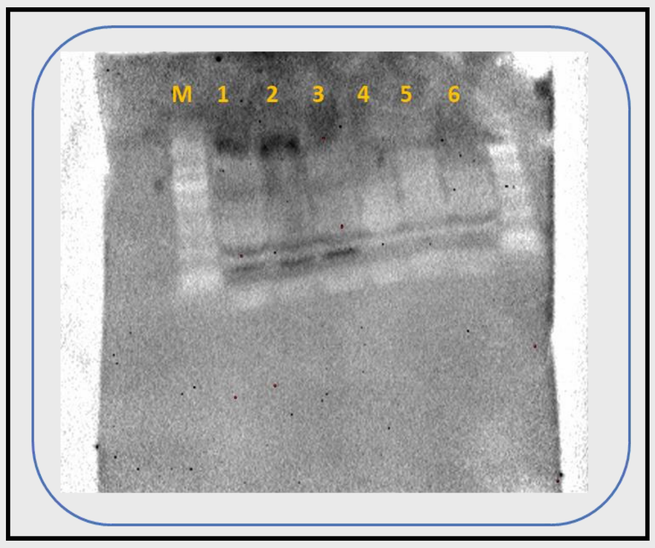


Fig. 2 Confirmation of Abc1p transporter and assessment of its expression level with western blot analysis following exposure to various treatments. **1**: DE (DMSO and ethanol) control, **2**: 32 µg/ml fluconazole (FLC), **3**: 0.1 mM Arachidonic acid (AA), **4**: combination of 0.1 mM AA and FLC, **5**: 1 mM AA, **6**: combination of 1 mM AA and FLC. Lanes 1 and 2 show expected band. M represents SDS-PAGE Pre-stained Protein Ladder (3.5 to 245 kDa).

Influence of AA and FLC on the activity of Abc1p

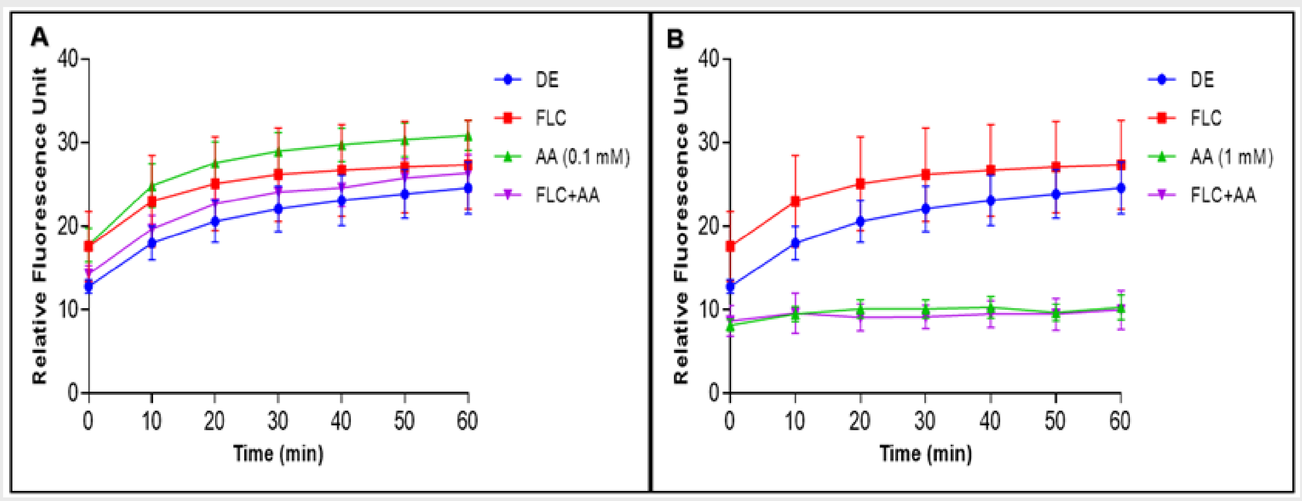


Fig. 3 Rhodamine 6G efflux in *C. krusei* biofilms after treatment with 0.1 mM arachidonic acid (**A**) and 1 mM arachidonic acid (**B**) in the presence or absence of fluconazole (32 µg/ml). Values are means of three independent experiments and error bars indicate standard deviation. **DE**: DMSO+Ethanol (control), **FLC**: fluconazole, **LA**: linoleic acid, **GLA**: gamma-linolenic acid.

Discussion and conclusions

- In this study, the influence of AA and FLC on the expression and activity of Abc1p was examined using western blot analysis and Rh6G efflux assay, respectively
- Although Abc1 transporter was overexpressed in the presence of FLC, its expression was abrogated and undetected in all treatments with AA
- Additionally, the functionality of Abc1p was enhanced in the presence of FLC; however, this was severely extenuated and diminished upon exposure to 1 mM AA, either alone or in combination with FLC.
- Although the mechanism by which AA diminishes the activity of Abc1 transporter was not investigated, our results do indicate that AA inhibits the expression of Abc1p
- Taken together, these findings demonstrate AA (1 mM) as a potential inhibitor of Abc1p expression and subsequent activity, and lent credence to the role of this transporter in FLC resistance

Future research

- Delineate the mechanism through which AA influences Abc1p
- Evaluate if other unsaturated fatty acids will produce similar effect

References

References can be obtained from the authors

Acknowledgements

National Research Foundation

ECA 2021 The 1st International Electronic Conference on Antibiotics The Equal Power of Antibiotics and Antimicrobial Resistance 08–17 MAY 2021 | ONLINE

UNIVERSITY OF THE FREE STATE
UNIVERSITEIT VAN DIE VRYSTAAT
YUNIVESITHI YA FREISTATA

UFS
NRF SARCHI CHAIR IN PATHOGENIC YEASTS