Novel natural and synthetic anticandidal therapeutic peptides to combat drug resistant infection

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INTRODUCTION

- Among the fungal genera, members of the genus *Candida* are most common pathogens causing skin, vaginal and oral infections.
- Extensive use of antifungal agents increased drug resistance in pathogenic strains of *Candida.*
- Antimicrobial peptides (AMPs) are considered as new generation antibiotics due to their diverse mechanisms of action.
- Present study is focused on natural and synthetic anticandidal peptides having therapeutic properties to combat drug resistant infections

RESULTS

- The physicochemical properties of synthetic peptides were predicated using Antifp software (Raghav et al., 2018) and their helical wheel projections were studied (Fig. 1).
- The MIC values of peptide SK08 was found to be 200 µg/ml and it was found to be non-haemolytic in nature (Fig. 2).

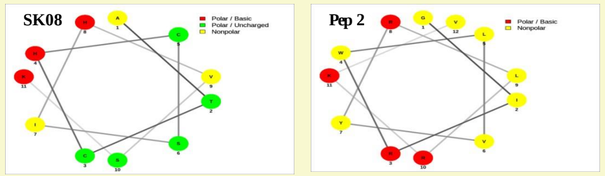

Fig. 1 Synthetic peptide helical wheel projections

Table 1 Physicochemical properties of synthetic peptides

S.No	Synthetic peptide	Hydropho--bicity	Amphipat-hicity	Hydrophili-city	charge	Mol. Wt.	structure
1	SK08	-0.12	0.65	-0.06	2	1937	defensin-like beta
3	Pep2	-0.25	0.92	-0.17	4	1559	Helical

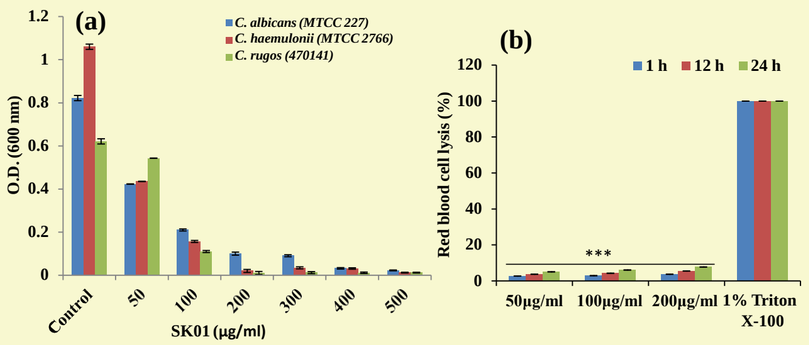

Fig. 2 (a) SK08 minimum inhibitory concentration (b) SK08 hemolysis assay

- *Bacillus subtilis* strain A52 and *Brevibacillus* sp. SVDS-15 produced natural anticandidal peptides with low MIC values 12- 30 µg/ml (Fig 3).
- MALDI mass spectrometry of HPLC purified peptides A52 and SVDS-15 revealed molecular mass as 1061 Da and 1296 Da, respectively (Fig. 3 & 4).

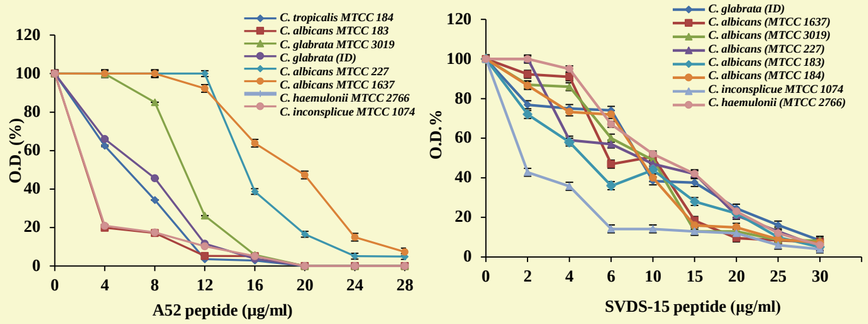

Fig. 3 MIC values of natural peptides against *Candida* strains

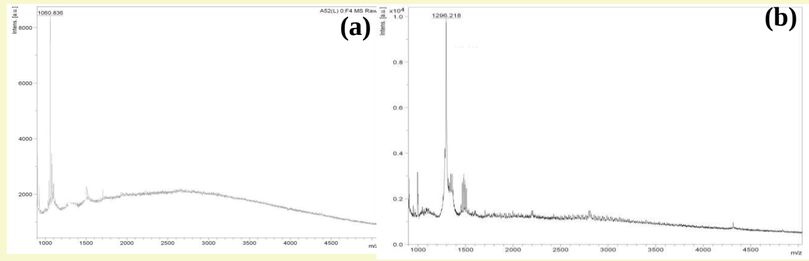

Fig. 4 MALDI mass spectrum (a) A52 (b) SVDS-15

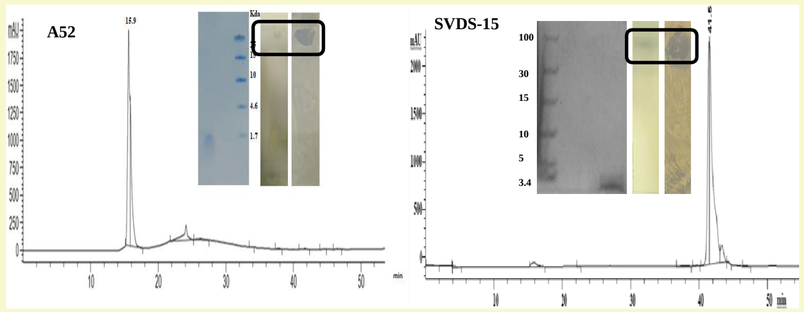

Fig. 5 RP-HPLC profile of peptides
(inset showing Tricine–SDS-PAGE of peptides, TLC stained with phosphomolybdic acid and bioautography demonstrating a clear inhibition)

- Strain A52 genome sequence showed presence of surfactin biosynthetic cluster producing novel low mol weight cyclic lipopeptide (Sharma et al., 2020).
- Peptides found to be pH (4.0 -10.0) and temperature stable.
- Killing kinetic studies of A52 showed complete killing within 7 h of treatment whereas SVDS-15 showed complete killing at 90 minutes (Fig. 5).
- SEM images showed peptides caused cell wall lysis (Fig. 6).

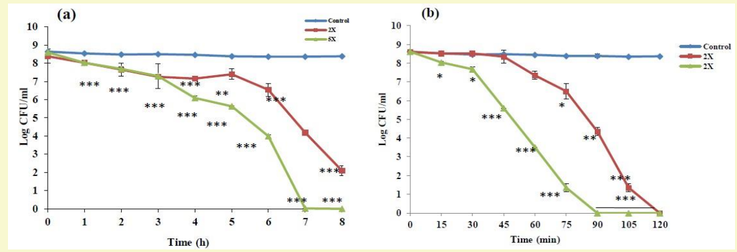

Fig. 6 Killing kinetics against *C. albicans* (a) A52 (b) SVDS-15

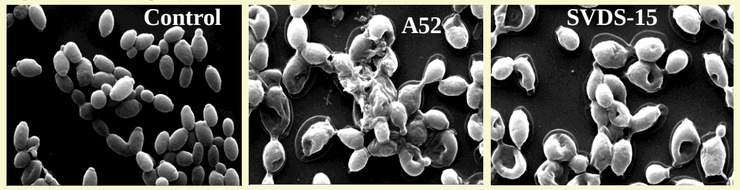

Fig. 7 SEM images of *C. albicans* treated with peptide

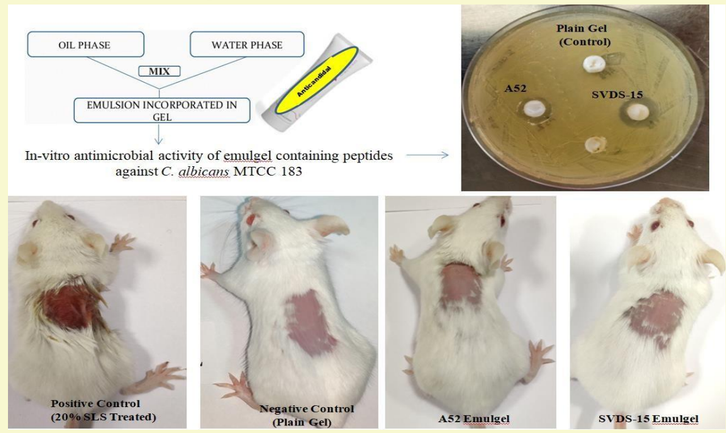

Fig. 8 Skin irritation studies, application of peptide for 72 h

CONCLUSIONS

- A52 and SVDS-15 peptides showed potent anticandidal activities.
- Emulgel formulation of peptides showed in vitro antifungal activity
- Emulgels did not caused irritation on BALB/c mice skin.
- Peptides have potential for external therapeutic applications.

ACKNOWLEDGEMENTS

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