VT-1161 Protects Immunosuppressed Mice from *Rhizopus oryzae* Infection

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INTRODUCTION

- Mucormycosis (zygomycosis) is a rare but life-threatening fungal infection which are mainly caused by *Rhizopus* species (1).
- It occurs mostly in immunocompromised hosts such as neutropenic patients (1).
- VT-1161 is a novel antifungal CYP51 selective inhibitor with broad spectrum activity against fungi (2).
- Here we assessed the *in vitro* and *in vivo*activity of ISA against *Rhizopus oryzae*, the most common cause of mucormycosis.

METHODS

- The in vitro susceptibility of VT-1161 against *R. oryzae* clinical isolates was evaluated using the Clinical Laboratory and Standards Institute (CLSI) M38-A2 method.
- ICR mice were immunosuppressed by cyclophosphamide (200 mg/kg) and cortisone acetate (500 mg/kg) on days -2 and +3, relative to infection (3).
- Immunosuppressed mice were intratracheally infected with *R. oryzae* susceptible strain.
- For survival studies, treatment with oral VT-1161 given once daily started 16 h post infection and continued through day +7. Treatment with LAmB started 16 h post infection and continued through day +4.
- For tissue fungal burden, treatment with either drug started 8 h post infection and continued through day +3. Mice were sacrificed on day +4 and fungal burden in target organs was determined by qPCR (4).
- Placebo mice received vehicle 0.5% carboxymethyl cellulose given by oral gavage.
- Statistical analysis was carried out by the non-parametric Wilcoxon Rank Sum test for the tissue fungal burden and by Log Rand Sum test for the survival studies with *P* values of <0.05 being significant.

REFERENCES

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RESULTS

Table 1. *Rhizopus oryzae* is susceptible to VT-1161. We have tested 12 clinical isolates of *R. oryzae* Type I or *R. oryzae* Type II (aka *R. delemar*) for their susceptibility to VT-1161.

Species	Isolate Number	VT-1161 MIC (mg/L)
R. oryzae	RO1	0.5
R. oryzae	RO2	2
R. oryzae	RO5	0.5
R. oryzae	RO8	0.5
R. oryzae	RO9	0.25
R. oryzae	RO10	1
R. oryzae	RO12*	1
R. delemar	RO3	>32
R. delemar	RO4	>32
R. delemar	RO6	8
R. delemar	RO7	>32
R. delemar	RO11	16

*isolate used in infection model

Table 2. Mouse plasma levels of VT-1161. Mice were treated with 3 daily doses of VT-1161 at 7.5 mg/kg or 15 mg/kg. A day after the last treatment, plasma was collected and drug levels were measured by LC/MS-MS. Data are presented in mg/L.

Mouse #	VT-1161 (7.5 mg/kg)	VT-1161 (15 mg/kg)
1	3.1	12
2	5.2	11
3	7.9	10
4	8.2	10
5	5.4	12
6	4.3	10
7	3.8	10
8	4.8	12
9	12	9.6
10	3.5	11
Mean (SD)	6 ± 3	11 ± 0.8

Vehicle control animals had undetectable levels of VT-1161.

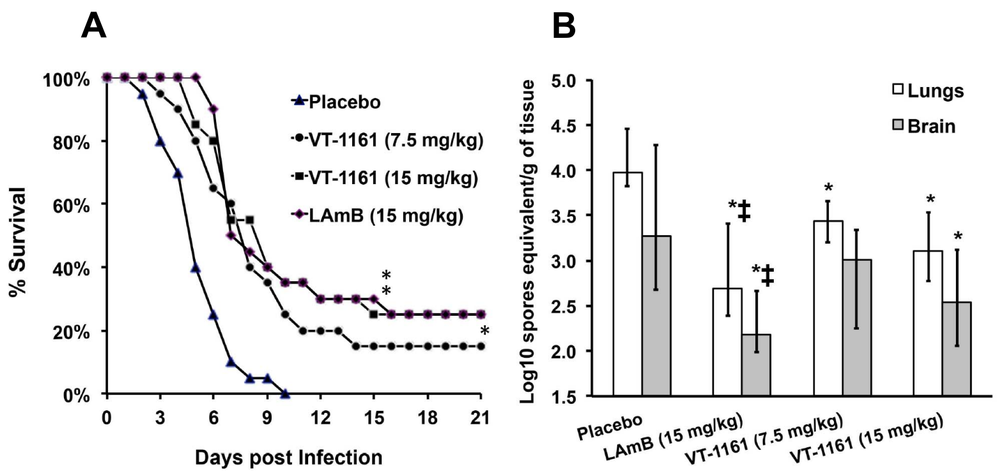

Fig 2. VT-1161 is as effective as high dose LAmB in improving survival (A) and reducing fungal burden (B) of neutropenic mice with mucormycosis. Mice (n=20 per arm from two independent experiments with similar results) were infected intratracheally with *R. oryzae* (average inoculum of 6.9 x 10^3). (A) **P* < 0.001 of all treated arms compared to placebo by Log Rank test; (B) Lungs and brains mice (n=20 per arm) were harvested on day +4 post infection. **P* < 0.002 compared to placebo while ‡*P* < 0.02 versus VT-1161 at 7.5 mg/kg by Wilcoxon Rank Sum test.

SUMMARY/CONCLUSIONS

- VT-1161 has better MIC activity against *R. oryzae* versus *R. delemar.*
- VT-1161 was significantly efficacious in the neutropenic murine model of pulmonary mucormycosis.
- VT-1161 efficacy was equivalent to the positive comparator LAmB.
- At the doses tested, plasma levels were above most of the MIC values vs. *Rhizopus* species.
- No drug-related adverse effects were observed with the two doses tested.
- These data warrant further examination of VT-1161 in the treatment of mucormycosis.

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