CHAGAS DISEASE TARGET CANDIDATE PROFILE AND SUGGESTED SCREENING CASCADE

Resynthesized and confirmed hits:
- IC50 <5uM • Max activity >90%
- Selectivity >10 • Small chemical expansion and early SAR

HIT ASSESSEMENT

Initial HTS / HCS hit

Chemical- & target-based prioritization:
- Prioritize novel chemical scaffolds
- Removal of CYP51 inhibitors
- Attention to other highly prevalent MoAs

***In vitro* assessment:**
- IC_{50} <1uM • Max activity >90%
- Selectivity >50 • Stable activity (10x) among *T. cruzi* strains • Fast acting and cidal preferred
- Measured *in vitro* metabolism, lipophilicity, permeability, and solubility

***In vivo* PK and Tolerability:**
- Free C_{min} above IC_{50}/IC_{90} for at least 8h
- No adverse effects at meaningful exposure

***In vivo* Chagas BLI acute mouse model:**
- 5 days QD/ BID 50 mg/kg PO • Significant reduction in parasitemia at EOT

HIT-TO-LEAD

Cytotoxicity host cells

***T. cruzi in vitro* assays**

ADME Tier 1

Mouse PK and tolerability

Chagas efficacy model Acute BLI

Compound design and synthesis

Complementary assays

Secondary *in vitro* parasitology assays:
- Trypomastigotes • Time-to-kill
- Washout • Panel of strains
- Persisters

Target/MoA deconvolution:
- Genetic & chemical validation

***In vivo* Chagas BLI chronic mouse model:**
- 100% parasite burden reduction and no relapse after immunosuppression

Exploratory toxicology & Safety pharmacology:
- No major issues identified

In-depth understanding of PKPD relationship:
- Human dose prediction <10 mg/kg/day

LEAD OPTIMIZATION

ADME Tier 2

***T. cruzi in vitro* assays**

Safety profiling

PKPD

Chagas efficacy model Chronic BLI

Pre-clinical candidate

Adapted from Kratz JM, Gonçalves KR, Romera LMD, Moraes CB, Bittencourt-Cunha P, Schenkman S, Chatelain E, Sosa-Estani S. *Memorias do Instituto Oswaldo Cruz 2021*; 116: e200501. doi: 10.1590/0074-02760200501.

DNDi Best Science for the Most Neglected

OPEN CHAGAS