#5211

A Broad Spectrum Novel Bacterial Topoisomerase Inhibitor (NBTI) Killing Kinetics, ADME and *in vivo* Efficacy

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Background

- Lead Optimization was carried out on a novel series of bacterial topoisomerase inhibitors. The series originated from a phenotypic screen and initially optimised using the Elude Platform to circumvent efflux liabilities
- Efflux mitigated early leads were optimised for spectrum, ADME, safety and efficacy.
- The key ADMET, PK and efficacy data for the potential lead candidate, BWC0977 is presented in this poster
- Refer Poster#4654 for target potency and bacterial spectrum of BWC0977

BWC0977: *in vitro* ADME Properties

Properties	Mean Values
MIC in M9 medium (µg/mL)	0.03
MIC (µg/mL) in M9 medium with 30 mg/mL BSA (µg/mL)	0.03
Human Plasma Protein binding % bound	94
Beagle dog Plasma Protein binding % bound	94
SD rat plasma Protein binding % bound	95
Cynomolgus Monkey plasma Protein binding % bound	88
Aqueous Solubility pH 7.4 (µM)	364
Aqueous solubility (simulated intestinal fluid) (µM)	895.5
Aqueous solubility (simulated gastric fluid) (µM)	902
IV formulation solubility: 10% ascorbic acid in water, pH 4.0; (millimolar)	55
Hu_Heps Clint (µl/min/million cells)	2.1
Dog_Heps Clint (µl/min/million cells)	4
Rat_Heps Clint (µl/min/million cells)	5.8
Gp_Heps Clint (µl/min/million cells)	6.6
Predicted Plasma Clearance - Human- CLp (L/hr/kg)	0.34
Predicted %Human LBF (1.24 l/h/kg)	28
CYP1A2 IC50 (µM)	>100
CYP2B6 IC50 (µM)	>100
CYP2C8 IC50 (µM)	>100
CYP2C9 IC50 (µM)	>100
CYP2C19 IC50 (µM)	>100
CYP2D6 IC50 (µM)	>100
CYP3A4 IC50 (Testosterone) (µM)	>100
CYP3A4 IC50 (Midazolam) (µM)	>100

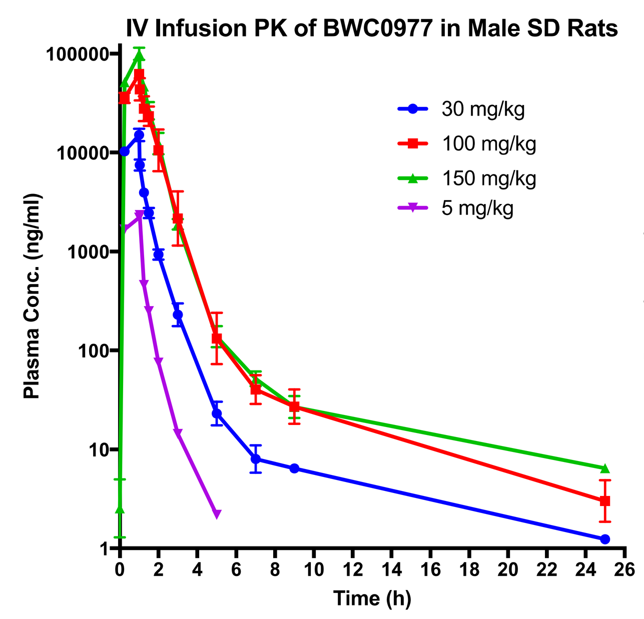
BWC0977: *in vitro* Safety

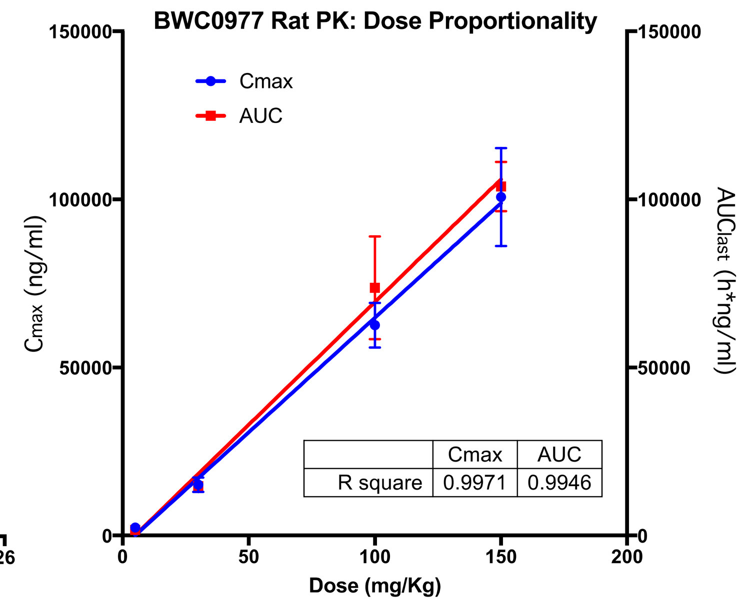
Properties	Mean Values
Cytotoxicity (HepG2) IC50 (µM)	>100
Cytotoxicity (A549) IC50 (µM)	>100
Cytotoxicity (HEK293) IC50 (µM)	>100
Cytotoxicity (Human Heps) IC50 (µM)	>100
Phospholipidosis (HepG2, HCA) at 100 µM fold induction	0.8
Cardiac Channels	
hERG IC50 (µM)	131
hNaV1.5 peak IC50 (µM)	>300
hNaV1.5 late IC50 (µM)	>300
hCaV1.2v IC50 (µM)	>300
hKCNQ1/E1 (Iks) IC50 (µM)	>300
Kv4.3 (Ito) IC50 (µM)	>300
Kir2.1 IC50 (µM)	>300
In vitro micronucleus assay	
Micronucleus (CHO - S9) (µM)	>250
Micronucleus (CHO + S9) (µM)	>1000

Conclusions

- BWC0977 is a novel broad spectrum NBTI
- Exhibits rapid killing in vitro
- ADME properties include good solubility, high metabolic stability in rat and human hepatocytes, and lack of inhibition of major human CYP450 enzymes
- In vitro safety: weak to lack of inhibition of cardiac channels, major cell lines & hepatocytes and negative in CHO-micronucleus assay
- Significant renal clearance observed
- PK/PD in rats: Exposures exceeded the range of MIC to yield significant effiacay in the rat thigh, lung and UTI infection models

Pharmacokinetics: Sprague Dawley Rats





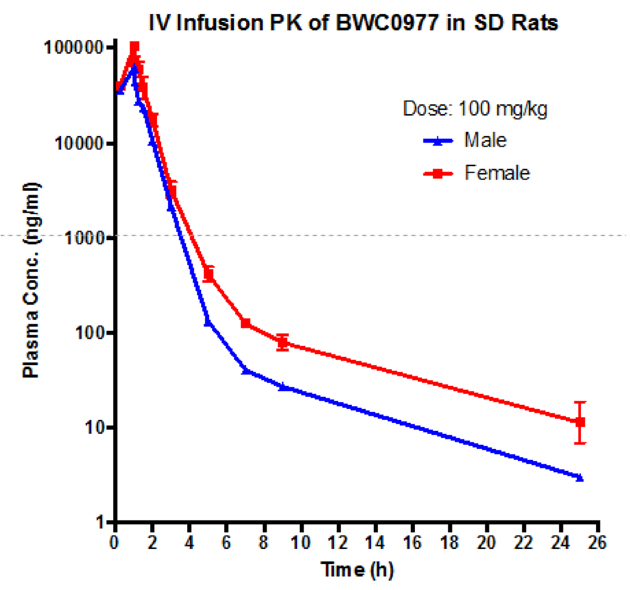
BWC0977 - IV NCA PK parameters[Mean ± SD]

Parameter	5 mg/kg	30 mg/kg	100 mg/kg	150 mg/kg
C_{max} (µg/ml)	2.4 ± 0.06	15.2 ± 2.2	62.6 ± 6.7	100.7 ± 145.5
T_{max} (h)	0.77 ± 0.01	1.0 ± 0.0	1.0 ± 0.0	1.0 ± 0.0
AUC_{inf}(h*µg/ml)	1.7 ± 0.05	14.7 ± 0.2	73.7 ± 15.3	103.9 ± 7.3
AUC_{last} (h*µg/ml)	1.7 ± 0.05	14.7 ± 0.2	73.7 ± 15.3	103.9 ± 7.3
CLp (L/h/kg)	2.92 ± 0.08	2.04 ± 0.03	1.40 ± 0.29	1.45 ± 0.11
Vss (L/kg)	0.91 ± 0.05	0.84 ± 0.06	0.90 ± 0.05	0.86 ± 0.04
$t_{1/2}$ (h)	0.53 ± 0.03	0.96 ± 0.02	4.93 ± 0.5	6.17 ± 0.42

Note: n=3 animals/group.

- Moderate Clearance
- Low Volume of Distribution
- Dose proportional increase in C_{max} and AUC

Pharmacokinetics: Male vs. Female SD Rats

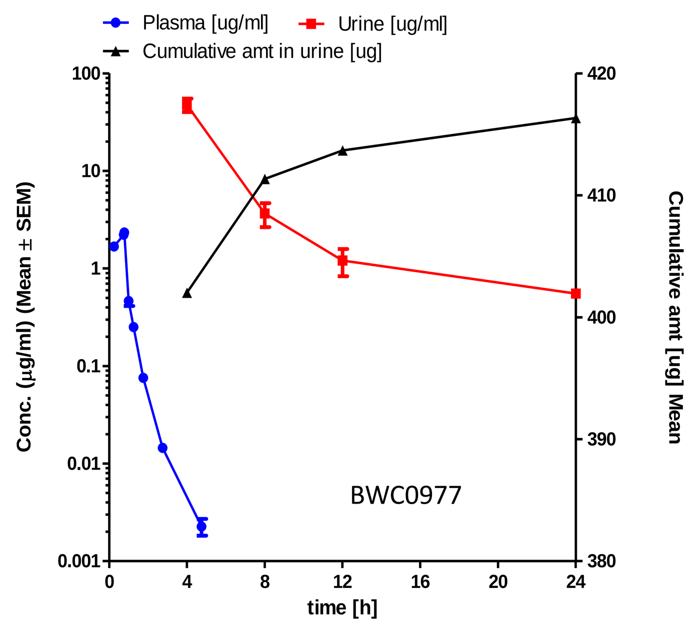


BWC0977 - IV NCA PK parameters [Mean ± SD]

Parameter	100 mg/kg [Males]	100 mg/kg [Females]
C_{max} (µg/ml)	62.6 ± 6.7	104.1 ± 7.01
T_{max} (h)	1.0 ± 0.0	1.0 ± 0.0
AUC_{inf}(h*µg/ml)	73.7 ± 15.3	114.4 ± 12.2
AUC_{last} (h*µg/ml)	73.7 ± 15.3	114.4 ± 12.1
CLp (L/h/kg)	1.40 ± 0.29	0.88 ± 0.09
Vss (L/kg)	0.90 ± 0.05	0.71 ± 0.06
$t_{1/2}$ (h)	4.93 ± 0.5	5.63 ± 1.45

Note: n=3 animals/group.

Pharmacokinetics in Rats: Renal Clearance



Dose: 5 mg/kg [1 hour infusion]

Parameter	Estimate
Fraction excreted in urine (fe)	0.27
Plasma clearance(L/h/kg)	2.92
Renal clearance (L/h/kg)	0.79
AUC_{last} (hr*ug/ml)	181

- 30% parent compound in urine over 24 he
- Renal clearance: could be significant route of elimination
- Potential for efficacy in UTI and cUTI patients
- Confirmatory studies in Dog in progress

References

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3. Hameed P, S. et al. **2018**. Nitrothiophene carboxamides, a novel narrow spectrum antibacterial series: Mechanism of action and Efficacy. *Scientific Reports.* Volume **8,** Article number: 7263

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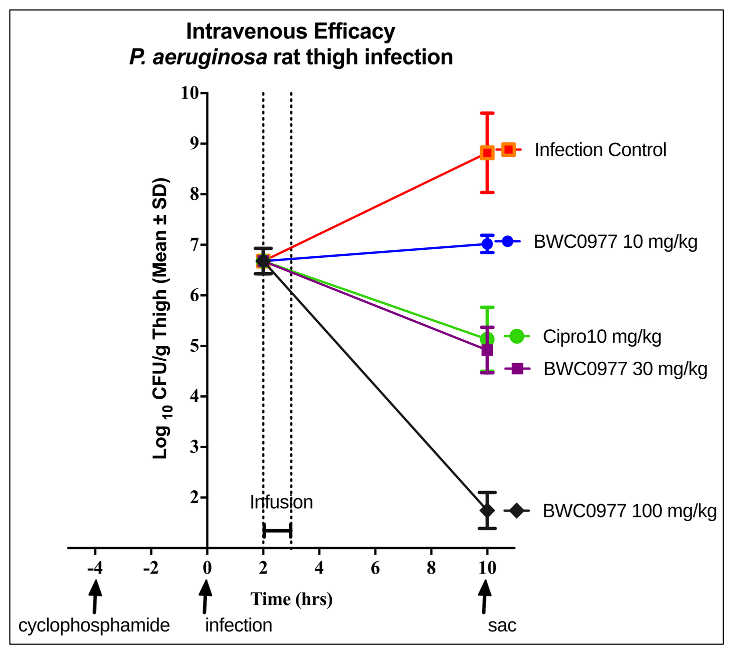
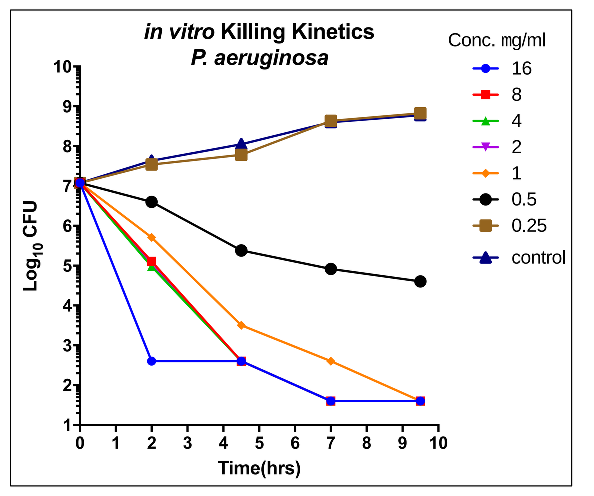
In vitro Killing Kinetics & *in vivo* Efficacy

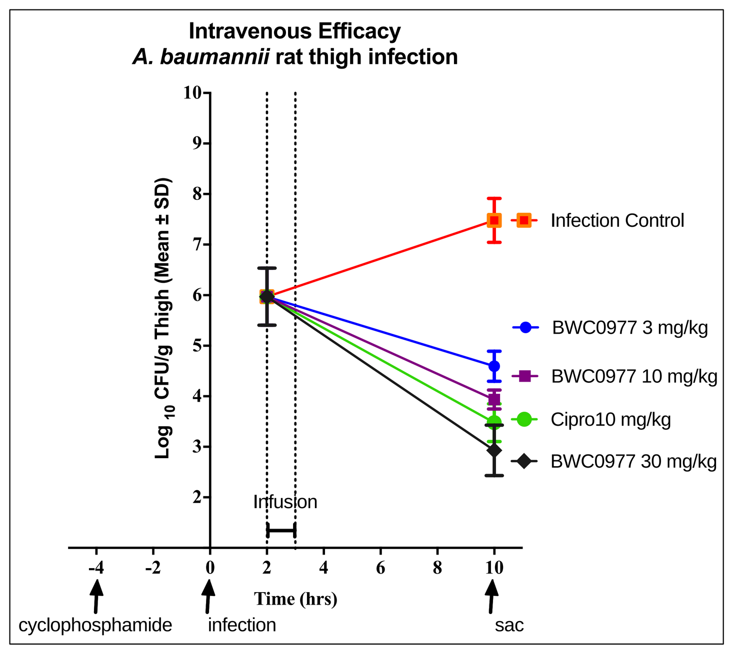
Pseudomonas aeruginosa



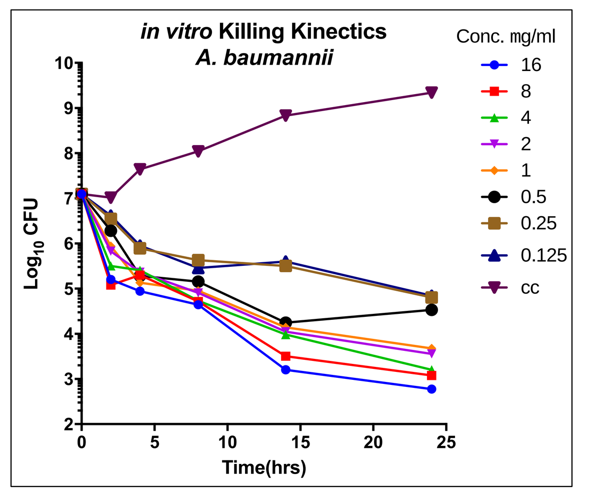
Concentration Dependent Killing
5 Log kill 8x MIC by 10h



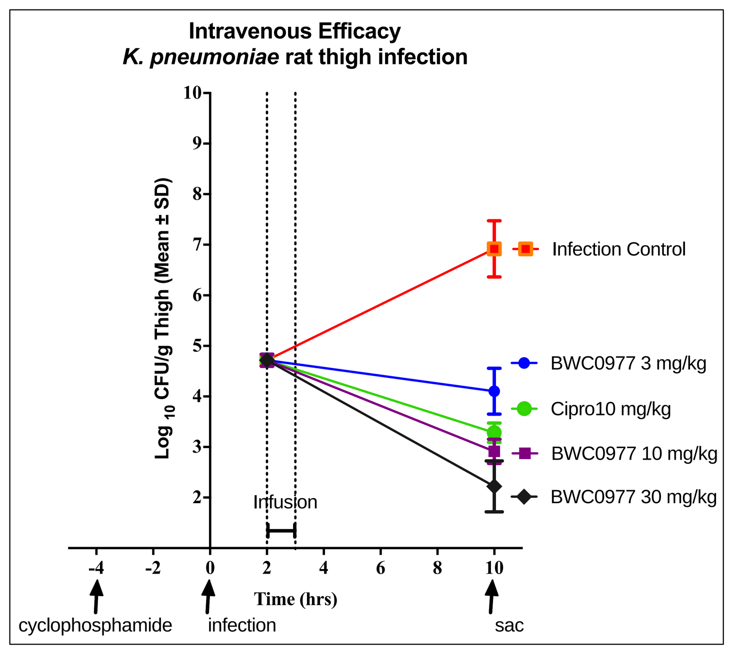
Acinetobacter baumannii



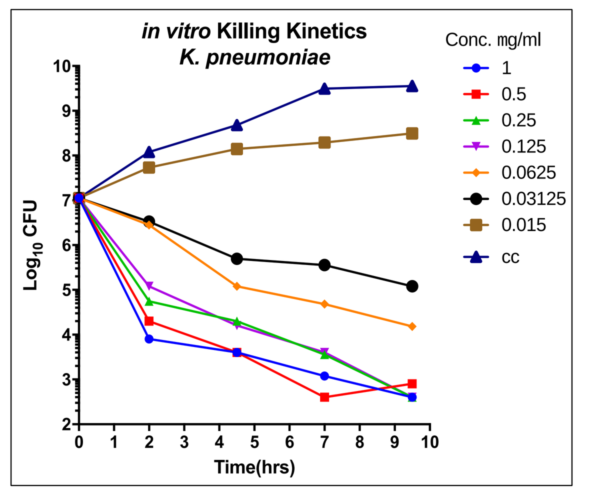
Concentration dependent Killing
4.5 Log kill at 64x MIC at 24h



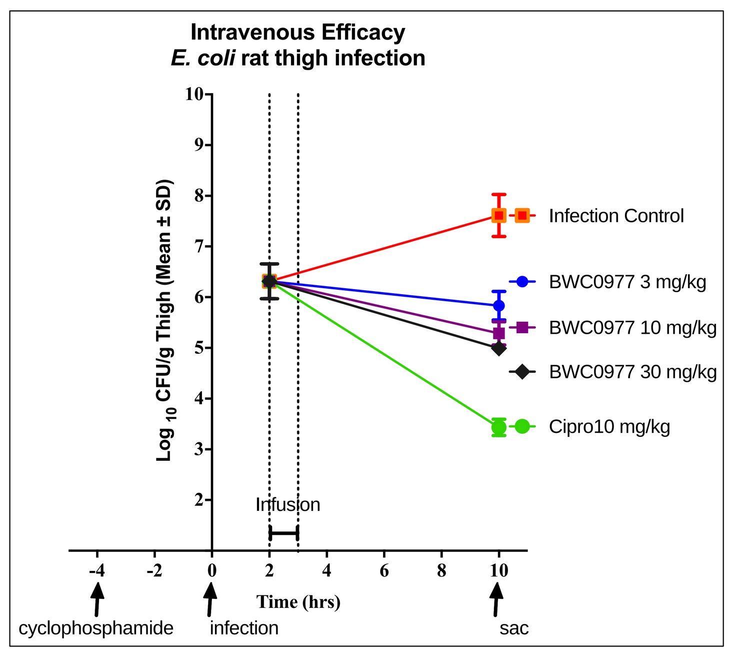
Klebsiella pneumoniae



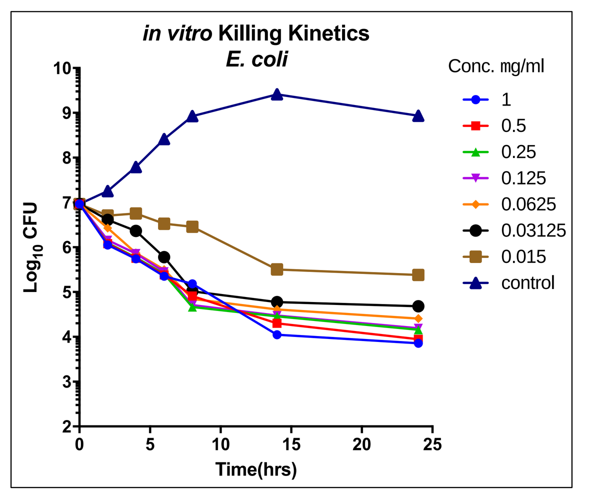
Concentration dependent Killing
4.5 Log kill at 64x MIC at 10h



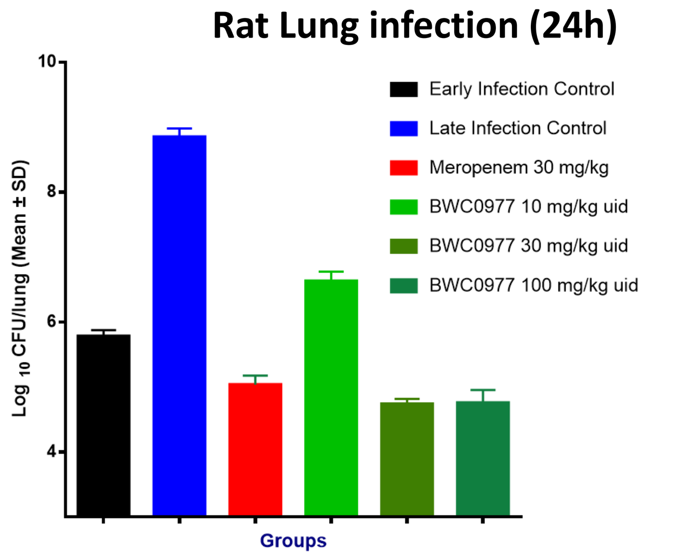
Escherichia coli



Concentration Independent Killing
3 Log kill at 64x MIC at 24h



Intravenous efficacy P. aeruginosa
Rat Lung infection (24h)



Intravenous efficacy E. coli
Rat UTI infection (24h)