# Rethinking the pharmacologic principles of PZA: a metabolomic perspective

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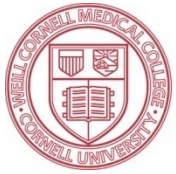
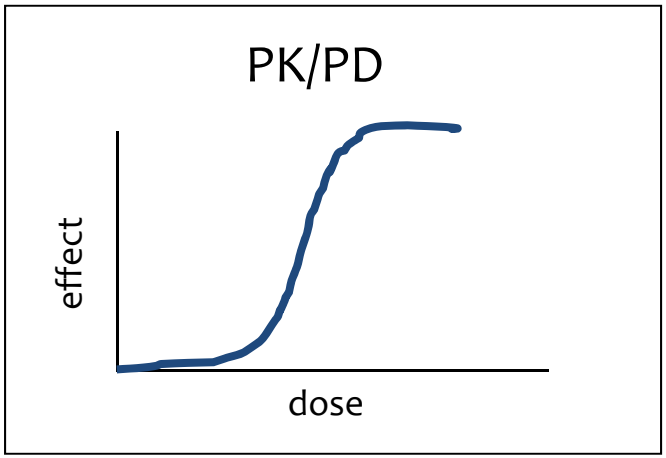
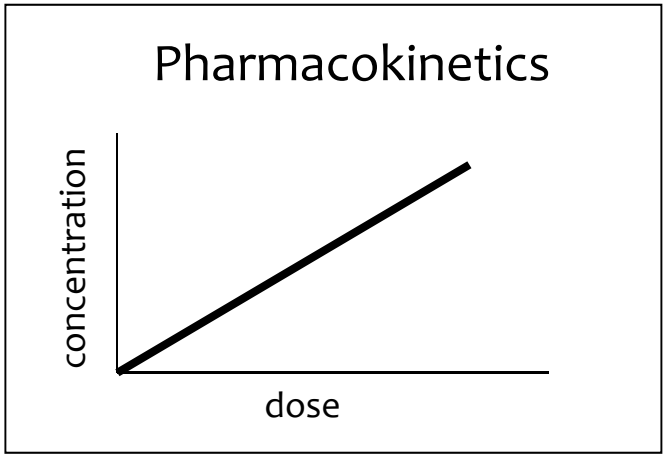
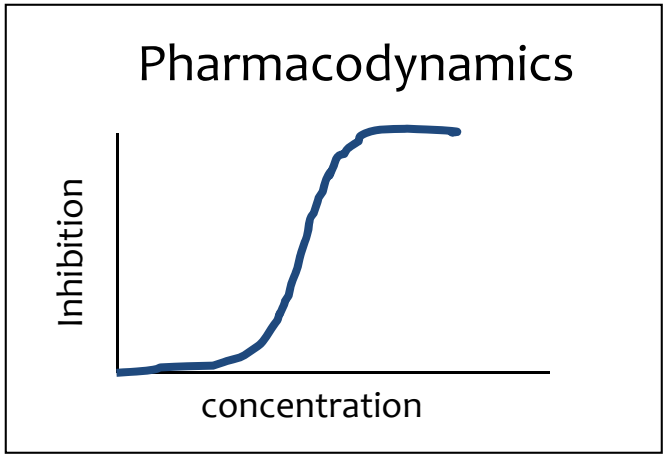
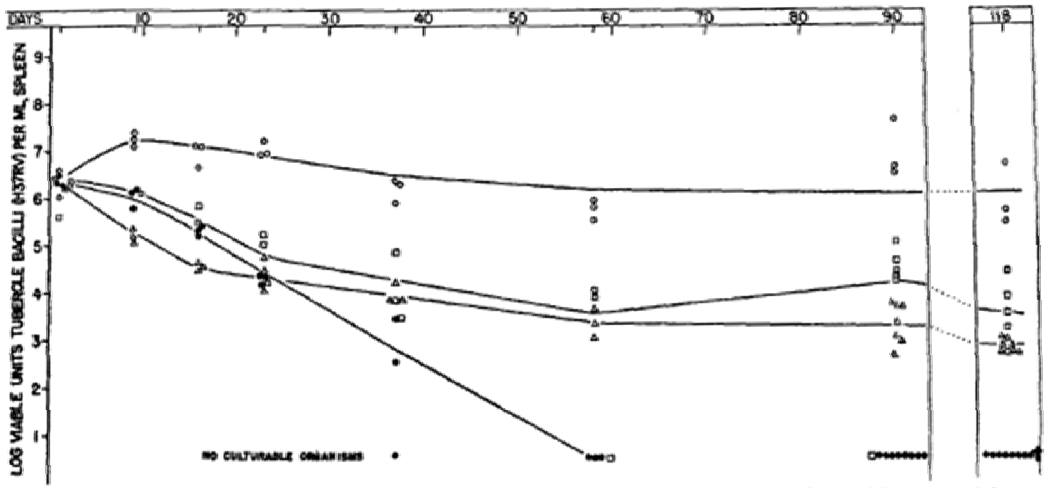


Weill Cornell Medical College

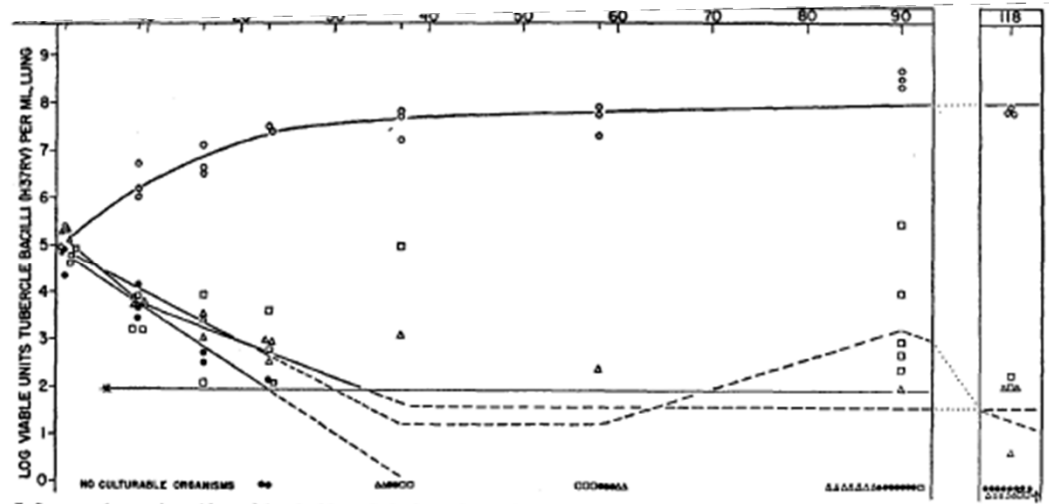

PK/PD
effect
dose


Pharmacokinetics
concentration
dose


Pharmacodynamics
Inhibition
concentration




TEXT-FIG. 2. Influence of pyrazinamide and isoniazid used singly and together on populations of tubercle bacilli (H37Rv) in spleens of the same animals whose lung populations are shown in Text-fig. 1. ○, control; □, pyrazinamide; △, isoniazid; ●, pyrazinamide-isoniazid.

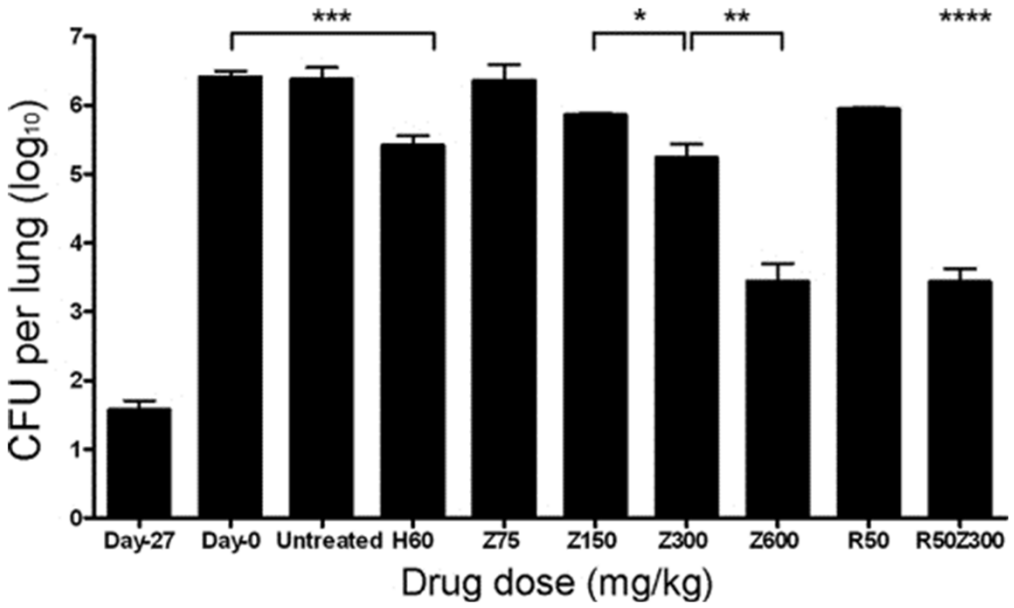



TEXT-FIG. 1. Influence of pyrazinamide and isoniazid used singly and together on populations of tubercle bacilli (H37Rv) in mouse lungs during 118 days of therapy. Infecting inoculum; $2.0 \times 10^6$ culturable units tubercle bacilli. ○, control; □, pyrazinamide; △, isoniazid; ●, pyrazinamide-isoniazid.

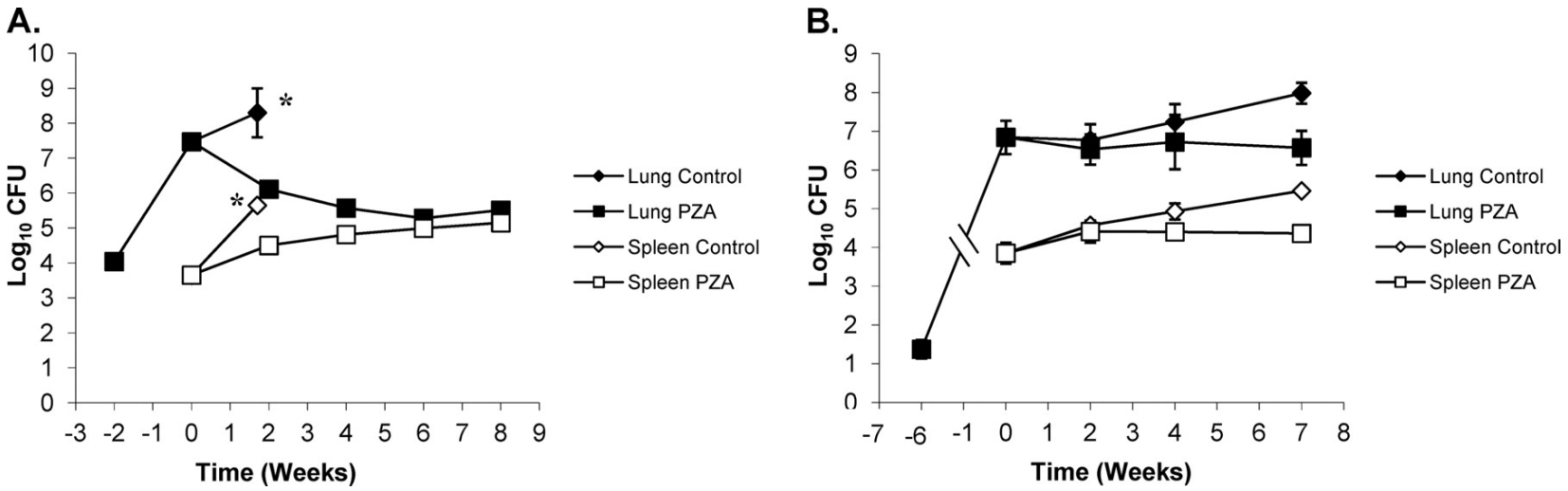
* The techniques used during the first 90 days of the experiment permitted detection of 70 to 90 culturable units of tubercle bacilli per lung.

† The techniques used on day 118 permitted detection of 1 to 3 culturable units of tubercle bacilli per lung.

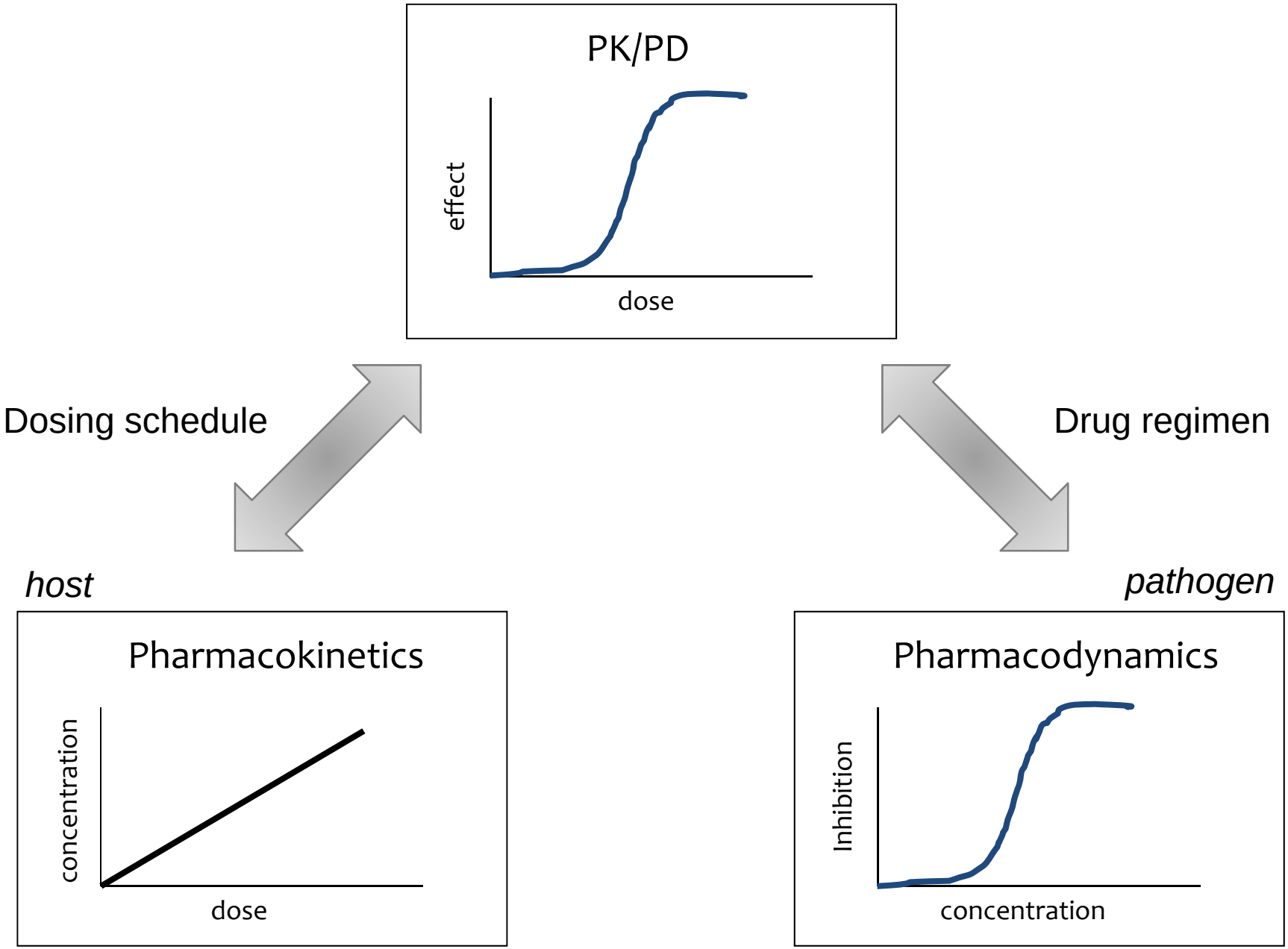
McCune et al. 1956. JEM. 104: 763.

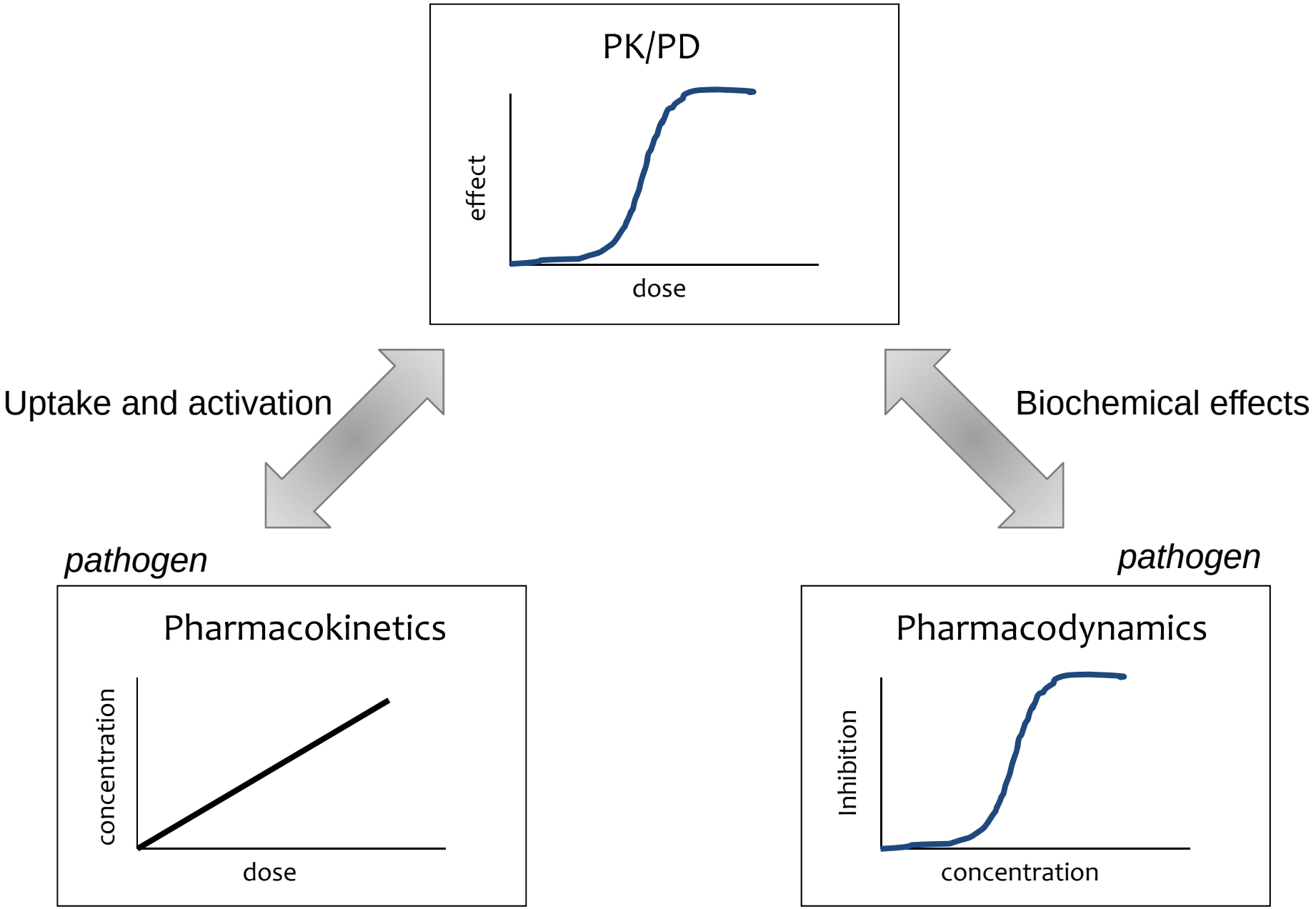



Ahmad Z et al. 2011. AAC. 55: 1527




Driver E R et al. 2012. AAC. 56:3181-3195


PK/PD
effect
dose
Dosing schedule
Drug regimen
host
pathogen
Pharmacokinetics
concentration
dose
Pharmacodynamics
Inhibition
concentration

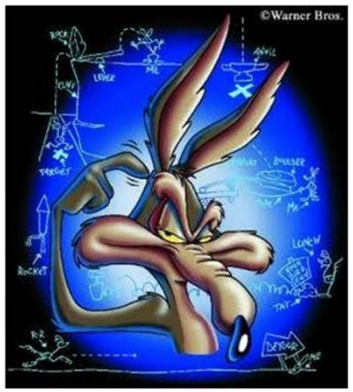

PK/PD
effect
dose
Uptake and activation
Biochemical effects
pathogen
pathogen
Pharmacokinetics
concentration
dose
Pharmacodynamics
Inhibition
concentration

# In vitro pharmacologic properties…

- Conditional
  - PncA
  - acid pH
- Synergistic
- Multiple reported biochemical targets
- AUC/MIC driven

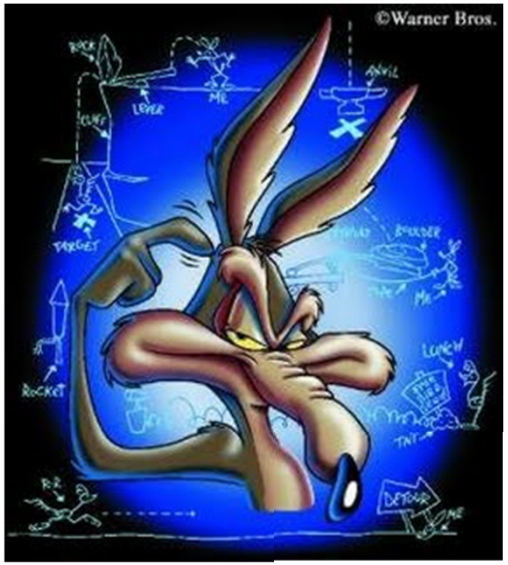
# PK/PD ambiguities…

- One drug, one target?
- One drug, many targets?
- Many drugs, many targets?
- Moving targets?

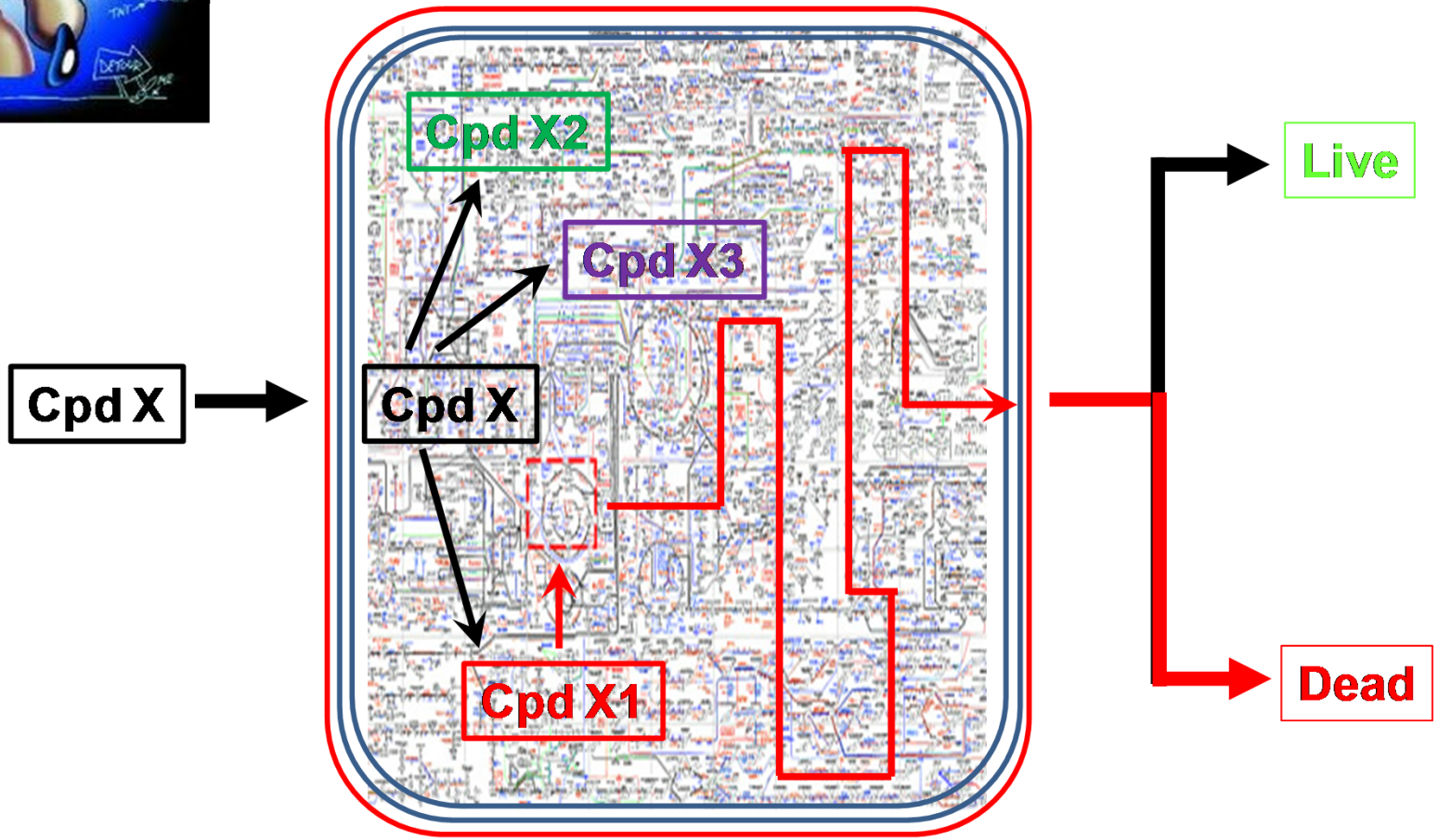



# TB Drug Development 2012

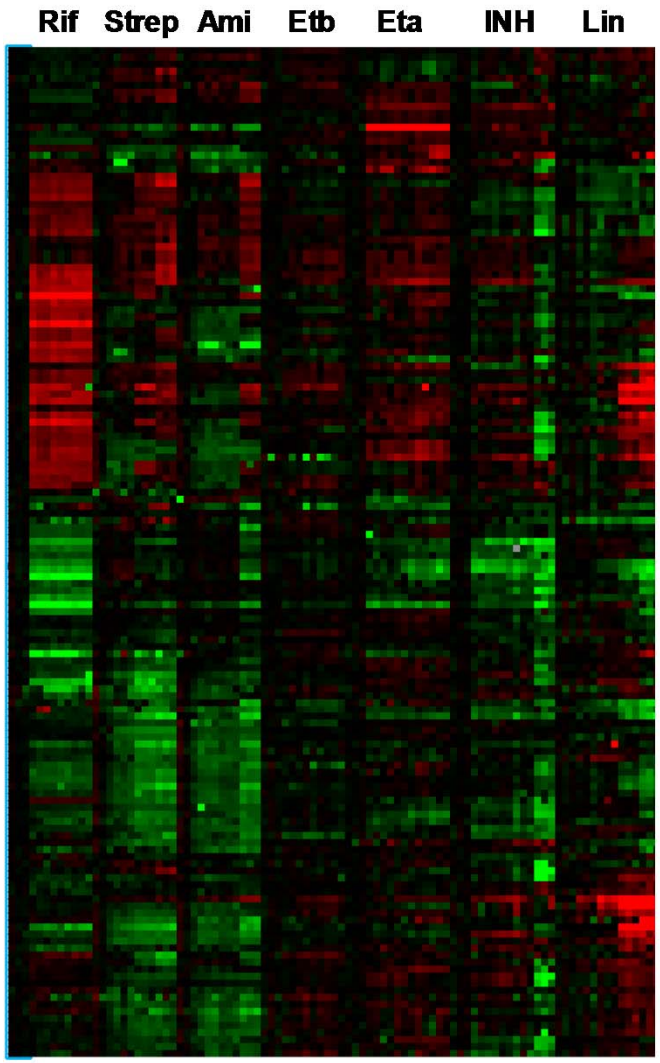
DRUG → **Mtb** → live / dead

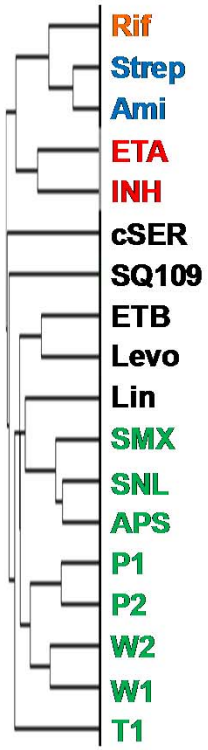


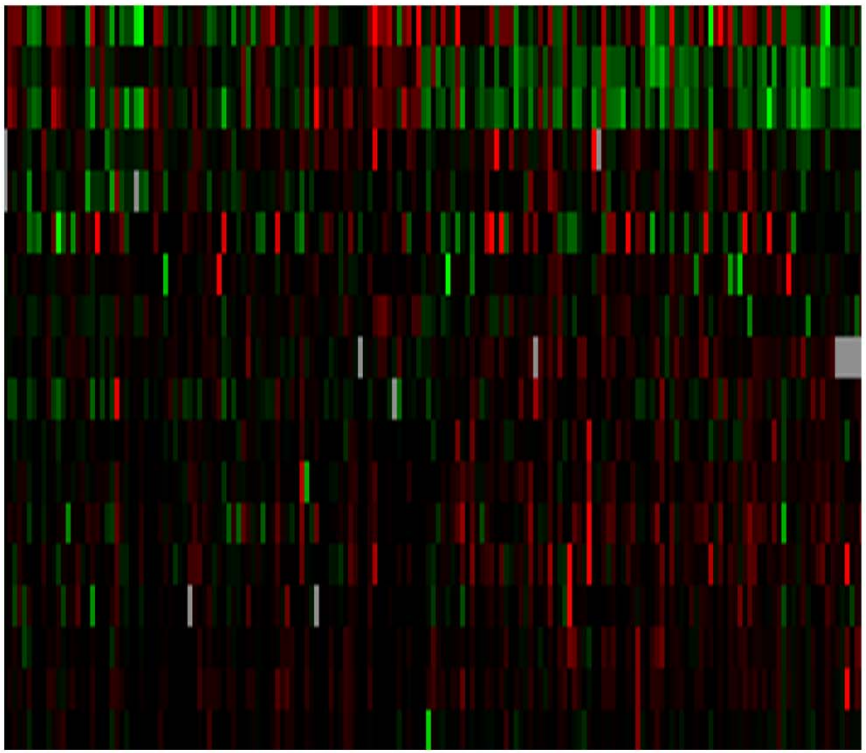

# Metabolomic pharmacology



# Compound profiles: Discriminatory and diagnostic



# PK profiling of PZA

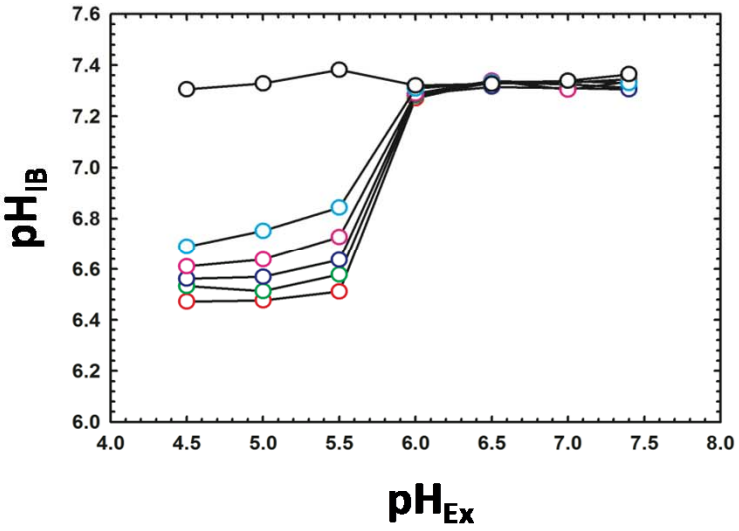
- Enzyme-catalyzed conversion of PZA into POA (pyrazinamidase - *pncA*) is a mediator of both sensitivity and resistance to PZA (classic activation mechanism)

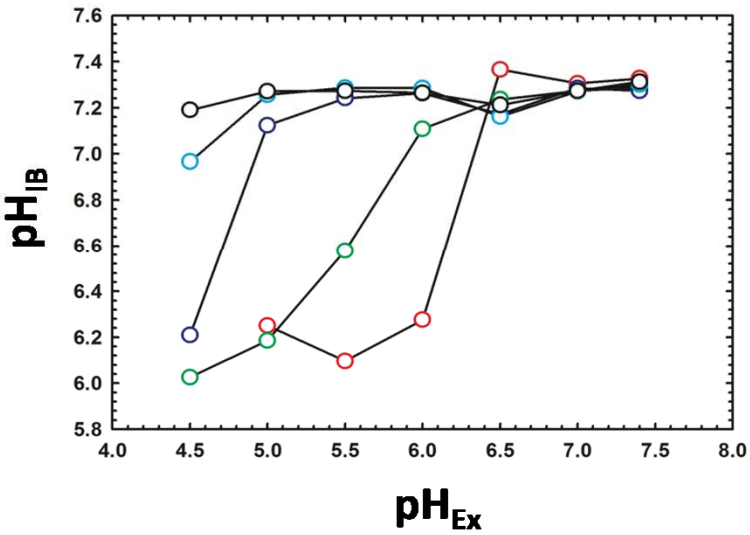
TABLE 2. Effect of PncA and PzaA expression on PZA and 5-Cl PZA turnover and MIC in tuberculous bacilli

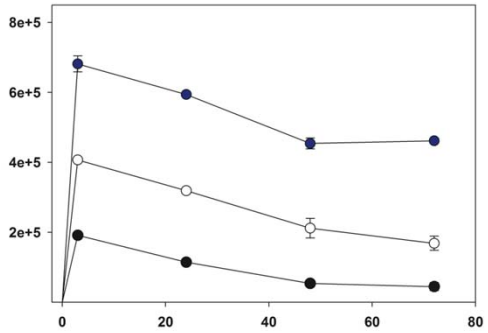
| Strain | Characteristic[a] | 5-Cl PZA | | | PZA | | | Reference |
|---|---|---|---|---|---|---|---|---|
| | | Turnover (nmol/min/ml of cells [$OD_{600}$]) | MIC μg/ml | | Turnover (nmol/min/ml of cells [$OD_{600}$]) | MIC μg/ml | | |
| | | | pH 6 | pH 6.8 | | pH 6 | pH 6.8 | |
| *M. tuberculosis* H37Ra | Attenuated mutant of H37Rv | 0.15 ± 0.01 | 25 | 25 | 0.23 ± 0.01 | >1,000 | 50 | |
| mc$^2$7092 | H37Ra *attB*$_{1.5}$::$P_{Tc}$::*pzaA*$_{Mmarg}$ | 27 ± 1 | 200 | 100 | 20 ± 1 | 62.5 | 25 | This work |
| mc$^2$7093 | H37Ra *attB*$_{1.5}$::$P_{Tc}$::*pncA*$_{Mtb}$ | 0.096 ± 0.002 | 25 | 25 | 2.9 ± 1 | 62.5 | 50 | This work |
| *M. bovis* BCG-Pasteur | Attenuated mutant of *M. bovis* | 0.11 ± 0.01 | 12.5 | 12.5 | 0.015 ± 0.001 | >1,000 | >1,000 | |
| mc$^2$7091 | BCG-Pasteur *attB*$_{1.5}$::$P_{Tc}$::*pzaA*$_{Mmarg}$ | 24 ± 1 | 200 | 50 | 16 ± 1 | 12.5 | 62.5 | This work |
| mc$^2$7099 | BCG-Pasteur *attB*$_{1.5}$::$P_{Tc}$::*pncA*$_{Mtb}$ | 0.080 ± 0.01 | 12.5 | 12.5 | 3.5 ± 0.1 | 12.5 | 62.5 | This work |

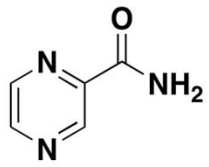
[a] *Mmarg*, *M. marginatum*; *Mtb*, *M. tuberculosis*.

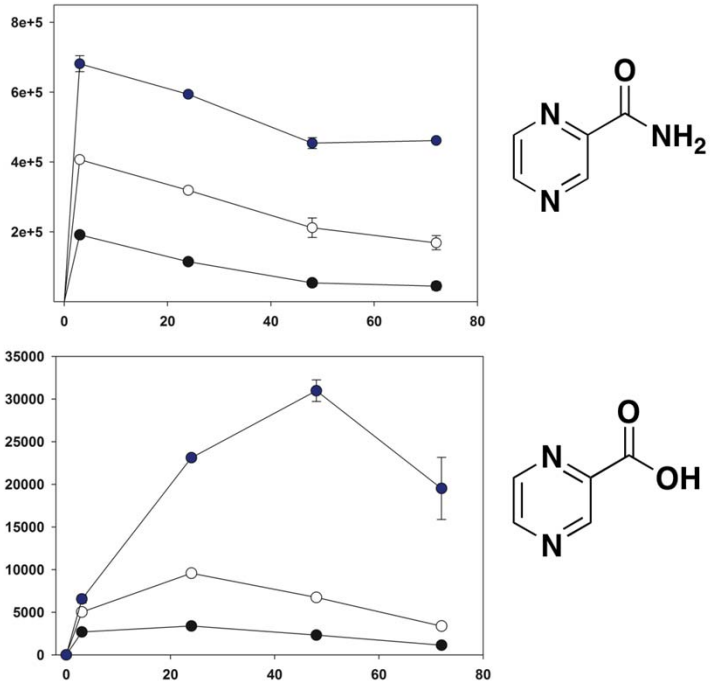
Baughn et al. 2010. AAC. 54: 5323


7.6
7.4
7.2
7.0
6.8
6.6
6.4
6.2
6.0
4.0
4.5
5.0
5.5
6.0
6.5
7.0
7.5
8.0
pH_IB
pH_Ex


7.6
7.4
7.2
7.0
6.8
6.6
6.4
6.2
6.0
5.8
4.0
4.5
5.0
5.5
6.0
6.5
7.0
7.5
8.0
pH_IB
pH_Ex

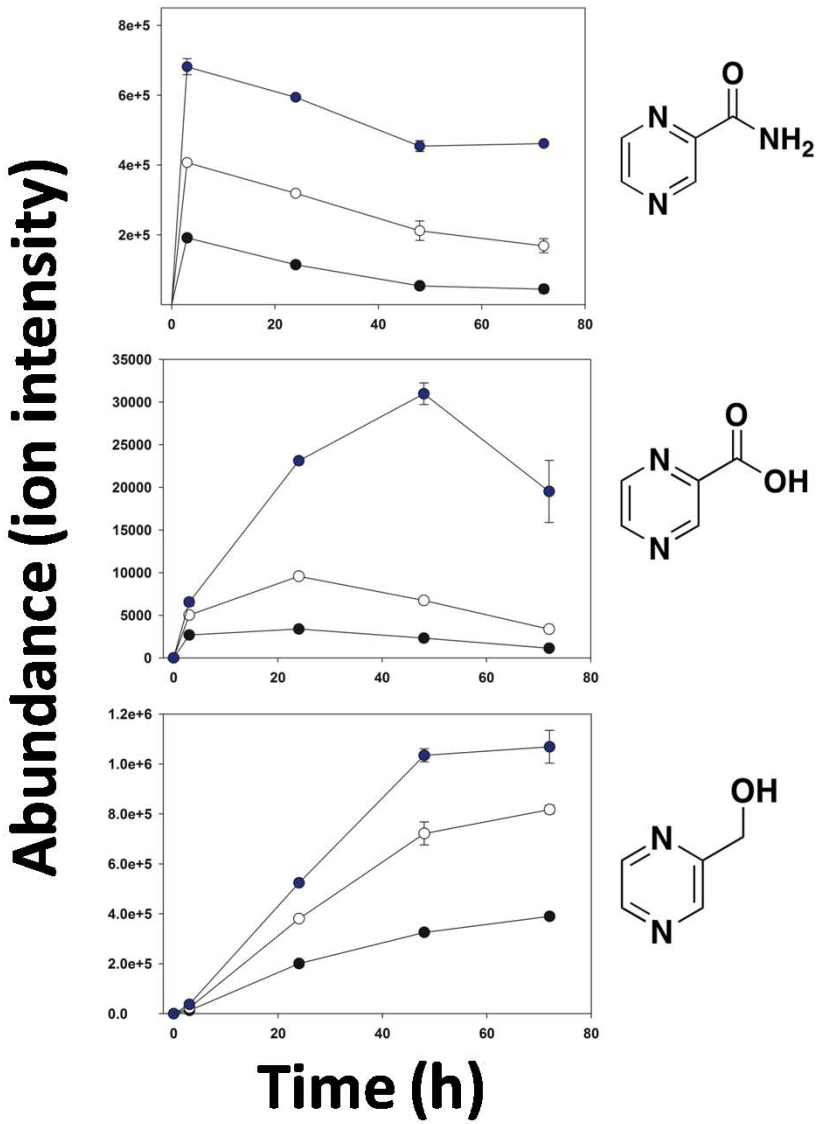

8e+5
6e+5
4e+5
2e+5
0
20
40
60
80


O
N
NH2
N

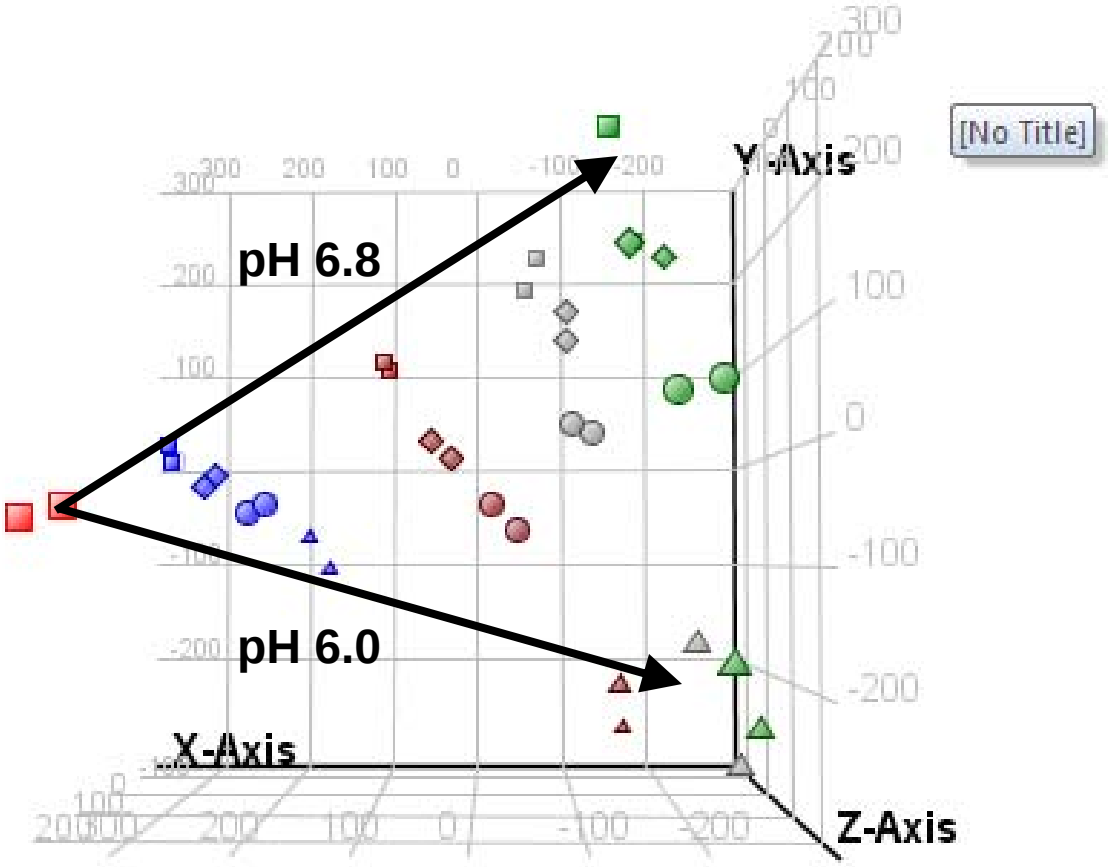

8e+5
6e+5
4e+5
2e+5
0
20
40
60
80
35000
30000
25000
20000
15000
10000
5000
0
0
20
40
60
80


O
N
NH2
N


O
N
OH
N


Abundance (ion intensity)
8e+5
6e+5
4e+5
2e+5
0
20
40
60
80
35000
30000
25000
20000
15000
10000
5000
1.2e+6
1.0e+6
8.0e+5
6.0e+5
4.0e+5
2.0e+5
0.0
Time (h)
O
N
NH2
OH

# Metabolomic profiles of PZA-treated Mtb




X-Axis Component 1 ... Y-Axis Component 2 ... Z-Axis Component 3 ...

# Goals

- Near term
  - Drug inventory
- Long term
  - Activity inventory
  - Target inventory
- Wishful thinking
  - Tissue/lesion inventory
  - New strategies and combinations

# Acknowledgements


- Sumit Chakraborty, Madhumitha Nandakumar
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- ID division

**William Randolph Hearst Foundation**