Intracellular accumulation of novel and clinically used TB drugs potentiates intracellular synergy

Running title: Intracellular accumulation and efficacies of novel TB active compounds

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Supplementary figures and tables:

Figure S1: Murine alveolar macrophage cells treated with PhX1 retrieved from BALF sampling. Light microscopy (40X) of murine cells from BALF sampling, demonstrating the predominance of alveolar macrophages.

Table S1: MS/MS settings

| Analyte | Transition (m/z) | Dwell time (ms) | Declustering potential (V) | Entrance potential (V) | Collison energy (eV) | Cell exit potential (V) |
|---------|------------------|-----------------|----------------------------|------------------------|----------------------|------------------------|
| RMB041  | 505.2→418.0      | 150             | 81                         | 10                     | 43                   | 10                     |
| WHN296  | 489.2→223.2      | 150             | 86                         | 10                     | 55                   | 18                     |
| PhX1    | 395.2→351.2      | 150             | 66                         | 10                     | 49                   | 22                     |
| PhX2    | 428.93→370.4     | 150             | 68                         | 10                     | 54                   | 21                     |
| PhX6    | 395.47→341.46    | 150             | 76                         | 10                     | 59                   | 12                     |
| PhX8    | 395.48→351.60    | 150             | 66                         | 10                     | 49                   | 22                     |
| PhX10   | 396.46→352.40    | 150             | 72                         | 10                     | 48                   | 21                     |
| PhX14   | 408.52→350.20    | 150             | 76                         | 10                     | 41                   | 24                     |
| PhX15   | 424.52→351.9     | 150             | 68                         | 10                     | 44                   | 20                     |
### Table S2: Electrospray ionization settings

| Analyte                          | Ion spray voltage (V) | Nebulizer gas (AU) | Curtain gas (AU) | Turbo gas (AU) | Source temperature (°C) |
|----------------------------------|-----------------------|--------------------|------------------|----------------|------------------------|
| RMB041                           | 4500                  | 40                 | 20               | 20             | 400                    |
| WHN296                           | 4500                  | 50                 | 25               | 25             | 400                    |
| PhX1; PhX2; PhX6; PhX8; PhX10; PhX14; PhX15; Carbamazepine BDQ | 4500                  | 40                 | 20               | 20             | 400                    |
| BDQ                              | 4000                  | 40                 | 20               | 20             | 400                    |
| MXF                              | 4500                  | 40                 | 25               | 25             | 400                    |
| RIF                              | 4000                  | 30                 | 10               | 10             | 400                    |
|                                  | 4500                  | 45                 | 15               | 15             | 400                    |
| Analyte | Column specifications | Mobile Phase A | Mobile Phase B | Flow rate (μL/min) | Gradient profile |
|---------|-----------------------|----------------|----------------|-------------------|------------------|
| RMB041  | Gemini-NX, 5μm, C18, 50 x 2.0 mm | 0.1% FA in H₂O | 0.1% FA in ACN | 300               |
|         |                       |                |                |                   | %A | %B     |
|         |                       |                |                |                   | 0.25 | 90 | 10 |
|         |                       |                |                |                   | 2.75 | 10 | 90 |
|         |                       |                |                |                   | 3.4 | 10 | 90 |
|         |                       |                |                |                   | 3.45 | 90 | 10 |
|         |                       |                |                |                   | 8   | 90 | 10 |
| WHN296  | Gemini-NX, 5μm, C18, 50 x 2.0 mm | 0.1% FA in H₂O | 0.1% FA in ACN | 300               |
|         |                       |                |                |                   | %A | %B     |
|         |                       |                |                |                   | 0.25 | 95 | 5  |
|         |                       |                |                |                   | 2.0 | 5  | 95 |
|         |                       |                |                |                   | 3.4 | 5  | 95 |
|         |                       |                |                |                   | 3.45 | 95 | 5  |
|         |                       |                |                |                   | 6   | 95 | 5  |
| PhX1; PhX2; PhX6; PhX8; PhX10; PhX14; PhX15; Carbamazepine | Gemini-NX, 5μm, C18, 50 x 2.0 mm | 0.1% FA in H₂O | 0.1% FA in ACN | 400               |
|         |                       |                |                |                   | %A | %B     |
|         |                       |                |                |                   | 0.25 | 90 | 10 |
|         |                       |                |                |                   | 1.75 | 10 | 90 |
|         |                       |                |                |                   | 4.7 | 10 | 90 |
|         |                       |                |                |                   | 4.75 | 90 | 10 |
|         |                       |                |                |                   | 8   | 90 | 10 |
| BDQ     | Atlantis T3; 5μm, C18; 100 x 2.1 mm | 0.1% FA in H₂O | 0.1% FA in ACN | 300               |
|         |                       |                |                |                   | %A | %B     |
|         |                       |                |                |                   | 0.25 | 60 | 40 |
|         |                       |                |                |                   | 1.75 | 0  | 100 |
|         |                       |                |                |                   | 2.7 | 0  | 100 |
|         |                       |                |                |                   | 2.75 | 60 | 40 |
|         |                       |                |                |                   | 6   | 60 | 40 |
| Column | Description | Mobile Phase A | Mobile Phase B | Time (min) | %A | %B |
|--------|-------------|----------------|----------------|------------|----|----|
| MXF    | Gemini-NX, 5μm, C18, 50 x 2.0 mm | 0.1% FA in H2O | 0.1% FA in ACN | 400 | | |
| RIF    | Discovery, 5µm, C18, 50 x 4.6 mm | 0.1% FA in H2O | 0.1% FA in MeOH | 300 | Isocratic conditions (60:40) | |
| LNZ    | Poroshell 120EC, C18, 50 x 4.6 mm, 2.7 um | 0.1% FA in H2O | 0.1% FA in MeOH | 300 | Isocratic conditions (60:40) | |
| LVX    | Gemini-NX, 5µm, C18, 50 x 2.0 mm | 0.1% FA in H2O | 0.1% FA in ACN | 400 | | |
| CLZ    | Gemini-NX, 5µm, C18, 50 x 2.0 mm | 0.1% FA in H2O | 0.1% FA in ACN | | | |