Objective: To conduct a study to establish MIC quality control (QC) ranges for GSK214094, a topoisomerase inhibitor, using the reference CLSI broth microdilution (BMD) method. This antimicrobial is being developed for the treatment of conventional and biothreat pathogens including methicillin-resistant Staphylococcus aureus (MRSA) and biofilm-forming pathogens, including against resistant variants to currently available agents.

Methods: An eight-laboratory study design was completed with CLSI-2A3 guidelines. Four QC strains were tested (E. coli ATCC 25923, S. aureus ATCC 29213, and two HI bacterium). Both HI and SP to also include all reported results. No results were considered unacceptable, regardless of the QC strain tested. Only one MIC value among 1,120 total results per laboratory was considered acceptable. The CLSI antimicrobial susceptibility committee determined an outlier site and could be supported by the Defense Threat Reduction Agency under Contract H98230-07-M-0001.

The zone diameters reported by eight laboratories for GSK214094 using three media lots (three manufacturers) of Mueller-Hinton broth (MHB) are reported in Figure 1. The CLSI study was performed to establish disk dilution and broth microdilution susceptibility tests. Figure 2 presents the results of the MIC QC study.

Materials and Methods: Eight laboratories were used in these two separate studies to establish a QC range. These laboratories were experienced microbiology facilities and each followed the CLSI procedures for disk dilution and broth microdilution methods.

For MIC tests, reference frozen-fresh broth microdilution panels were prepared by ThermoFisher Scientific (Cleveland, Ohio, USA) according to Good Manufacturing Practice (GMP) guidelines and shipped to all sites. Panels contained four lots of citrate-adjusted Mueller-Hinton broth (DH, HemMedia, USA; BD, Sparks, Maryland, USA; and Difco, Detroit, Michigan, USA) as well as four lots of Mueller-Hinton Test Medium (HIM) and four lots of Mueller-Hinton broth supplemented with 2.5% lysed horse blood (lot #304123, #H12361, #3024248). Three comparator disks from BD were used: azithromycin 15-µg (lot #2304320), levofloxacin 5-µg (lot #2272187) and linezolid 30-µg (lot #2291112). Three manufacturers (Remel, Lenexa, Kansas; Hardy Diagnostics, Santa Maria, California; and Becton Dickinson, Sparks, Maryland) were compared.

For disk diffusion tests, two lots of 10-µg GSK214094 disks were manufactured by two companies (MaST Group, New York, NY (lot #39072 and #39073)). Three comparator disks from BD were used: azithromycin 15-µg (lot #2304320), levofloxacin 5-µg (lot #2272187) and linezolid 30-µg (lot #2291112). Three manufacturers (Remel, Lenexa, Kansas; Hardy Diagnostics, Santa Maria, California; and Becton Dickinson, Sparks, Maryland) were compared.

Table 1. CLSI approved quality control (QC) range for GSK214094 in Mueller-Hinton broth

<table>
<thead>
<tr>
<th>Fabricant</th>
<th>Lot</th>
<th>MIC (µg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GSK214094</td>
<td>0.12</td>
<td>1 – 2</td>
</tr>
<tr>
<td>GSK214094</td>
<td>0.25</td>
<td>1 – 2</td>
</tr>
<tr>
<td>GSK214094</td>
<td>0.5</td>
<td>1 – 2</td>
</tr>
</tbody>
</table>

Four laboratories tested 10 replicates of E. coli ATCC 29223, E. coli ATCC 29223, and S. aureus ATCC 49247. Each laboratory tested 10 replicates of S. aureus ATCC 25923, ATCC 29213, S. pneumoniae ATCC 49619, and S. pneumoniae ATCC 49619. All laboratories tested five replicates of S. pneumoniae ATCC 49619 (2.7 x 10⁵ CFU/ml), S. pneumoniae ATCC 29213 (3.2 x 10⁵ CFU/ml), and S. aureus ATCC 25923 NAa NAa 23 – 29 (24 – 29)b 96.4 (96.4)b

Conclusions: Proposed MIC QC ranges for GSK214094 are considered acceptable and within CLSI approved range. However, the zone diameters for this study with 96.4% of reported patient zones found within the CLSI published QC ranges.

References: