Preliminary Antimicrobial Susceptibility Interpretive Criteria for Cefetamet (Ro 15-8074) and Cefeteram (Ro 19-5247) Disk Tests

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Received 20 February 1987/Accepted 4 June 1987

Preliminary interpretive zone criteria were calculated for cefetamet (Ro 15-8074) and cefeteram (Ro 19-5247) by using 10- and 30-μg disks and three possible MIC susceptibility breakpoints. Absolute interpretive agreement between MICs and zone size criteria ranged from 91.8 to 97.2%. Very major errors (false susceptibility) were ≤1.2% for both cephalosporin disk tests. *Morganella morganii* strains appeared to produce the highest rates of very major interpretive errors with cefetamet disks.

Cefetamet (formerly Ro 15-8075) and cefeteram (formerly Ro 19-5248 and T-2588) are orally administered cephalosporin pivoxyl esters which, as free acids, possess an antimicrobial activity most similar to that of cefixime (1, 4, 5, 9-11, 13). Each free acid (Ro 15-8074 and Ro 19-5247 or T-2525) has been linked to form the pivoxyl oxygenester that allows gastrointestinal tract absorption but produces a somewhat lower concentration in serum than do currently available cephalosporins (12; data on file at Hoffmann-La Roche, Inc., Nutley, N.J.). Even with these lower drug levels in blood, cefetamet and cefeteram have sufficient potency to cover members of the family Enterobacteriaceae, *Haemophilus influenzae*, pathogenic *neisseriae*, *Branhamella catarrhalis*, and most streptococci (1, 4, 9-11, 13). In this brief report, we compare the antimicrobial activities of the free acids of these two new orally administered cephalosporins. We additionally present the preliminary determinations of disk diffusion test interpretive criteria with 10- and 30-μg cefetamet and cefeteram disks.

Cefetamet and cefeteram were provided by Hoffmann-La Roche, Inc. (R. Cleeland). Both drugs were tested in cation-supplemented Mueller-Hinton broth by dilution methods described by the National Committee for Clinical Laboratory Standards (8). Additional medium supplementation for fastidious organisms was prepared according to the National Committee for Clinical Laboratory Standards M7-A recommendations (8). The disk diffusion tests were also performed by National Committee for Clinical Laboratory Standards procedures (7) by using 10- and 30-μg disks prepared by the investigators to 110% of the stated potency. Regression analyses were calculated by the least-squares method as adapted to a computer by using the zone diameter as the independent variable. The error rates were calculated for three possible susceptibility MIC breakpoints to try to minimize the false-susceptibility error rate to ≤1.0% (6).

Recent typical clinical isolates were collected from the microbiology laboratories at The Cleveland Clinic Foundation, Cleveland, Ohio; St. Francis Hospital, Wichita, Kans.; St. Vincent Hospital and Medical Center, Portland, Oreg.; Northwestern Memorial Hospital, Chicago, Ill.; and the Kaiser Permanente Health Care Program Regional Laboratory, Clackamas, Oreg. The 671 isolates are listed in Table 1. These included 238 strains of *Enterobacteriaceae*, 84 of staphylococci (18 meticillin resistant), 80 of *Streptococcus* spp., 34 of enterococci, 10 of *Listeria monocytogenes*, 10 of *Brachyhalia catarrhalis*, 15 of *Acinetobacter anitratus*, 49 of *Haemophilus influenzae* (25 beta-lactamase producers), 19 of *Neisseria meningitidis*, 45 of *Neisseria gonorrhoeae* (23 beta-lactamase producers), and 87 of seven pseudomonad species.

The antimicrobial activities of the two cephalosporins are compared in Table 1. As measured by MIC results for 90% of strains tested, cefeteram was generally equivalent in activity to cefetamet against the Enterobacteriaceae and *H. influenzae* but superior against the *B. catarrhalis* and gram-positive coccus strains. Cefetamet was only marginally effective against *A. anitratus* (MIC for 90% of the strains tested, 8.0 μg/ml) and had no activity against most staphylococci. Neither drug significantly inhibited pseudomonad, enterococcus, or *L. monocytogenes* isolates. Among the enteric bacilli, two significant activity differences were observed. (i) The activity of cefetamet was very poor against some strains of *Morganella morganii*, whereas cefeteram remained effective (9), and (ii) some K1 beta-lactamase-producing *Klebsiella oxytoca* isolates were highly resistant to cefetamet but were inhibited by cefetamet (9) (MIC, 32 μg/ml [Table 1]).

Figure 1 shows a scattergram comparing the cefetamet and cefeteram MICs with their respective 30-μg disk zones of inhibition. Table 2 contains the regression analyses for the 600 to 604 strains tested against each drug and disk content. Both drugs had acceptable correlation coefficients (>0.80), and 251 to 296 organisms contributed to the statistical analyses. To date, little information has been published on the pharmacokinetics of the final clinical formulations of the cephalosporin esters. However, early reports assure levels in serum ranging from 0.9 to 4.7 μg/ml after oral administration of 100- to 500-mg tablets or suspensions (12; K. Stoeckel, E. Weidekamm, and P. Probst, Program Abstr. Vth Mediterranean Congr. Chemother. 1986, abstr. no. 0-50, p. 189). Therefore, these regression statistics were interpreted for three possible MIC susceptibility breakpoints (≤2, ≤4, and ≤8 μg/ml), pending more definitive pharmacokinetic information or improved levels in serum as a result of formulation changes. The cefetamet disk diffusion test produced slightly more false-susceptibility results (very major errors) than did the cefeteram disk test. This was primarily due to the results obtained with *M. morganii* strains. If the *M. morganii* results were omitted from the error rates, the very major error rates were in the acceptable range of 0.8 to 1.2%. The absolute agreement between cefetamet MICs and disk results was 92.9 to 97.2% with the *M. morganii* strains.

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TABLE 1. Cefetamet and ceftetam MICs for 671 bacterial strains

<table>
<thead>
<tr>
<th>Organism (no. of strains tested)</th>
<th>Cefetamet</th>
<th>Cefetam</th>
<th>90%</th>
<th>Range</th>
<th>90%</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acinetobacter anitrus (15)</td>
<td>8.0</td>
<td>2.0-32</td>
<td>32</td>
<td>8.0-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Branhamella catarrhalis (10)</td>
<td>0.5</td>
<td>0.25-1.0</td>
<td>≤0.06</td>
<td>≤0.06-0.12</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Haemophilus influenzae (49)</td>
<td>≤0.06</td>
<td>≤0.06-0.12</td>
<td>≤0.06</td>
<td>≤0.06-0.12</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neisseria meningitidis (19)</td>
<td>≤0.008</td>
<td>0.002-0.12</td>
<td>≤0.004</td>
<td>&lt;0.001-0.12</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neisseria gonorrhoeae (45)</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>16-&gt;32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pseudomonas aeruginosa (55)</td>
<td>&gt;32</td>
<td>0.12-&gt;32</td>
<td>&gt;32</td>
<td>0.5-&gt;32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pseudomonas spp. (32)¹</td>
<td>&gt;32</td>
<td>0.12-0.5</td>
<td>0.25</td>
<td>0.25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Citrobacter diversus (10)</td>
<td>0.25</td>
<td>0.12-0.5</td>
<td>0.25</td>
<td>0.25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Citrobacter freundii (10)</td>
<td>&gt;32</td>
<td>1.0-32</td>
<td>&gt;32</td>
<td>0.5-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enterobacter aerogenes (20)</td>
<td>0.5</td>
<td>0.12-32</td>
<td>0.25</td>
<td>0.12-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enterobacter agglomerans (10)</td>
<td>1.0</td>
<td>≤0.06-32</td>
<td>0.5</td>
<td>≤0.06-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enterobacter cloacae (21)</td>
<td>1.0</td>
<td>0.25-32</td>
<td>1.0</td>
<td>0.12-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Escherichia coli (34)</td>
<td>0.5</td>
<td>0.12-32</td>
<td>0.25</td>
<td>0.12-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Klebsiella spp. (30)</td>
<td>0.12</td>
<td>≤0.06-1.0</td>
<td>0.25</td>
<td>≤0.06-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Morganella morganii (10)</td>
<td>32</td>
<td>2.0-32</td>
<td>4.0</td>
<td>≤0.06-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proteus mirabilis (25)</td>
<td>≤0.06</td>
<td>≤0.06-0.12</td>
<td>≤0.06</td>
<td>≤0.06-0.25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proteus vulgaris (10)</td>
<td>0.12</td>
<td>≤0.06-32</td>
<td>0.25</td>
<td>≤0.06-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Providencia rettgeri (10)</td>
<td>≤0.06</td>
<td>≤0.06-0.5</td>
<td>≤0.06</td>
<td>≤0.06-16</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Providencia stuartii (24)</td>
<td>≤0.06</td>
<td>≤0.06-2.0</td>
<td>0.25</td>
<td>≤0.06-4.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serratia marcescens (24)</td>
<td>2.0</td>
<td>0.5-32</td>
<td>2.0</td>
<td>0.5-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>32</td>
<td>32-&gt;32</td>
<td>2.0</td>
<td>1.0-4.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Methicillin susceptible (49)</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Methicillin resistant (10)</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Coagulase-negative Staphylococcus spp.</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Methicillin susceptible (17)</td>
<td>8.0</td>
<td>4.0-32</td>
<td>1.0</td>
<td>1.0-4.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Methicillin resistant (8)</td>
<td>&gt;32</td>
<td>32-&gt;32</td>
<td>32</td>
<td>4.0-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Streptococcus agalactiae (20)</td>
<td>1.0</td>
<td>0.5-8.0</td>
<td>≤0.06</td>
<td>≤0.06-2.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Streptococcus pneumoniae (20)</td>
<td>1.0</td>
<td>0.25-32</td>
<td>0.12</td>
<td>≤0.06-2.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Streptococcus pyogenes (20)</td>
<td>≤0.06</td>
<td>≤0.06-1.0</td>
<td>≤0.06</td>
<td>≤0.06-1.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Streptococcus groups C and G (20)</td>
<td>≤0.06</td>
<td>≤0.06</td>
<td>≤0.06</td>
<td>≤0.06-0.25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enterococcus faecalis (24)</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>8.0-32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enterococcus faecium (10)</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td>&gt;32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Listeria monocytogenes (10)</td>
<td>&gt;32</td>
<td>4.0-32</td>
<td>&gt;32</td>
<td>4.0-32</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

¹ Includes P. acidovorans (three strains), P. cepacia (four strains), P. fluorescens (five strains), P. maltophilia (six strains), P. putida (five strains), and P. stutzeri (nine strains).

Note: MIC values are in micrograms per milliliter (µg/ml).

TABLE 2. Regression analyses of 10- and 30-µg cefetamet and cefetam disk comparisons with MICs determined by the broth microdilution method (8)

<table>
<thead>
<tr>
<th>Cephalosporin and disk content (µg)</th>
<th>No. of organisms included in statistics¹</th>
<th>y intercept</th>
<th>Slope</th>
<th>r</th>
<th>Mean zone diam (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cefetamet 10</td>
<td>251</td>
<td>16.9</td>
<td>-0.33</td>
<td>0.83</td>
<td>24.7</td>
</tr>
<tr>
<td>Cefetamet 30</td>
<td>285</td>
<td>17.6</td>
<td>-0.31</td>
<td>0.86</td>
<td>26.5</td>
</tr>
<tr>
<td>Cefetam 10</td>
<td>281</td>
<td>16.7</td>
<td>-0.34</td>
<td>0.81</td>
<td>23.0</td>
</tr>
<tr>
<td>Cefetam 30</td>
<td>296</td>
<td>18.2</td>
<td>-0.35</td>
<td>0.85</td>
<td>26.5</td>
</tr>
</tbody>
</table>

¹ With each disk, 600 to 604 strains were tested. Only strains having on-scale MICs (0.12 to 32 µg/ml) and zones of inhibition ranging from 7 to 40 mm were used for regression calculations.

These studies confirm the antimicrobial activities of these new cephalosporins (1, 4, 9-11, 13). However, their clinical usefulness will ultimately be determined by the pharmacokinetics of orally administered formulations. Many strains of Enterobacteriaceae and gram-positive species may not be inhibited if sustained levels in serum remain below 8 µg/ml. Our recommendations for MIC and zone diameter breakpoints for the two agents are shown in Table 3. All proposed interpretive criteria conform to the calculated regression statistics except those cited for the cefetamet disks. After excluding M. morganii strains, unacceptable false-susceptibility rates (>1.0%) required a 2-mm-larger zone breakpoint than that calculated from the cefetam disk regression formulae. Therefore, for the 30-µg cefetamet disk, we recommend an interpretive zone diameter of ≥20 mm for susceptibility to correspond to an MIC of ≥8 µg/ml. Our results for cefetamet were identical to those reported by an investigator from Hoffmann-La Roche (P. Hohl, Abstr. 86th Annu. Meet. Am. Soc. Microbiol. 1986, A-70, p. 12). This investigator also reported interpretive error problems with M. morganii, although he used an agar dilution MIC method and the agar overlay modification of the National Committee for Clinical Laboratory Standards disk diffusion test (7, 8).
FIG. 1. Scattergrams comparing cefetamet and ceferam MICs with their zones of inhibition around a 30-μg disk on the basis of tests of 600 bacterial isolates. The preliminary interpretive zone recommendations for a ≤4-μg/ml susceptibility MIC breakpoint are represented by vertical lines. The regression line was drawn for the MIC interval of 0.12 to 32 μg/ml.
TABLE 3. Recommended interpretive criteria for 10- and 30-μg cefetamet and cefetram disks, including interpretive error rates

<table>
<thead>
<tr>
<th>Cephalosporin and disk content (μg)</th>
<th>MIC breakpoint criterion (μg/ml)</th>
<th>Zone size interpretation (mm)</th>
<th>Error ratea</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Susceptible</td>
<td>Intermediate</td>
<td>Resistant</td>
</tr>
<tr>
<td>Cefetamet</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>≤2</td>
<td>4</td>
<td>≥8</td>
</tr>
<tr>
<td></td>
<td>≤4</td>
<td>8</td>
<td>≥16</td>
</tr>
<tr>
<td></td>
<td>≤8</td>
<td>16</td>
<td>≥32</td>
</tr>
<tr>
<td>30</td>
<td>≤2</td>
<td>4</td>
<td>≥8</td>
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<tr>
<td></td>
<td>≤4</td>
<td>8</td>
<td>≥16</td>
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<tr>
<td></td>
<td>≤8</td>
<td>16</td>
<td>≥32</td>
</tr>
<tr>
<td>Cefetram</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>≤2</td>
<td>4</td>
<td>≥8</td>
</tr>
<tr>
<td></td>
<td>≤4</td>
<td>8</td>
<td>≥16</td>
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<td></td>
<td>≤8</td>
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<td>30</td>
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<td>4</td>
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<td>≤4</td>
<td>8</td>
<td>≥16</td>
</tr>
<tr>
<td></td>
<td>≤8</td>
<td>16</td>
<td>≥32</td>
</tr>
</tbody>
</table>

a Very major. Falsely susceptible by disk diffusion test result; Major, falsely resistant by disk diffusion test result; Minor, intermediate result by one of the two methods compared.

b Corrected by adding approximately 2 mm to minimize false-susceptibility errors.

Our preliminary cefetram disk test recommendations are similar to those reported by Beskid et al. (G. Beskid, J. Durkin, J. Siebelist, V. Fallat, E. Lipschitz, R. Cleeland, D. McGarry, and E. Squires, Abstr. 86th Annu. Meet. Am. Soc. Microbiol. 1986, A-71, p. 12). However, the 30-μg disk susceptibility interpretive zone for ≤4 μg of Ro 19-5247 per ml reported by these investigators was ≥20 mm compared with the ≥21 mm cited here.

The newer orally administered cephalosporins may have compromised concentrations in serum that require a lower MIC breakpoint than the traditional ≤8 μg/ml (2, 3). If this proves to be the case for cefetamet and cefetram, it may be necessary to use larger interpretive zones with the 30-μg disk (Table 3) or, alternatively, to use a lower drug content disk, as has been suggested for cefixime (2). Final in vitro testing criteria await the results of more extensive clinical and pharmacokinetic studies.

We thank the following for technical support and manuscript contributions: P. Hohl, R. Cleeland, P. Fuchs, R. R. Packer, J. McClung, and C. Thornsberry.

LITERATURE CITED