Meropenem Pharmacokinetics in the Newborn

John N. van den Anker,1,2,3,4 Pavla Pokorna,5 Martina Kinzig-Schippers,6 Jirina Martinkova,7 Ronald de Groot,8 G. L. Drusano,9* and Fritz Sorgel6,10

Departments of Pediatrics1 and Pharmacology & Physiology,2 The George Washington University School of Medicine and Health Sciences, Washington, DC; Division of Pediatric Clinical Pharmacology, Children’s National Medical Center, Washington, DC3; Department of Pediatrics, Erasmus MC-Sophia Children’s Hospital, Rotterdam, The Netherlands4; Department of Pediatrics, Charles University, Prague, Czech Republic5; Institute of Biomedical and Pharmaceutical Research, Heroldsgen, Germany6; Department of Pharmacology, University of Hradec Kralove, Hradec Kralove, Czech Republic7; Department of Pediatrics, University Children’s Hospital Nijmegen, Nijmegen, The Netherlands8; Ordway Research Institute, Albany, New York9; and Department of Pharmacology, University of Duisburg—Essen, Essen, Germany10

Received 14 March 2009/Returned for modification 12 May 2009/Accepted 24 June 2009

We studied meropenem in 23 pre-term (gestational age, 29 to 36 weeks) and 15 full-term (gestational age, 37 to 42 weeks) neonates. Meropenem doses of 10, 20, and 40 mg/kg were administered as single doses (30-min intravenous infusion) on a random basis. Blood was obtained for determining the meropenem concentration nine times. Each child required other antimicrobials for proven/suspected bacterial infections. Samples were assayed by high-performance liquid chromatography analysis. Population pharmacokinetic parameter values were obtained by employing the BigNPAG program. Model building was performed by the likelihood ratio test. The final model included estimated creatinine clearance (CLcr) (Schwartz formula) and weight (Wt) in the calculation of clearance (meropenem clearance = 0.00112 × CLcr + 0.0925 × Wt + 0.156 liter/hr). The overall fit of the model to the data was good (observed = 1.037 × predicted − 0.096; r2 = 0.977). Given the distributions of estimated creatinine clearance and weight between pre-term and full-term neonates, meropenem clearance was substantially higher in the full-term group. A Monte Carlo simulation was performed using the creatinine clearance and weight distributions for pre-term and full-term populations separately, examining 20- and 40-mg/kg doses, 8- and 12-h dosing intervals, and 0.5-h and 4-h infusion times. The 8-h interval produced robust target attainments (both populations). If more resistant organisms were to be treated (MIC of 4 to 8 mg/liter), the 40-mg/kg dose and a prolonged infusion was favored. Treating clinicians need to balance dose choices for optimizing target attainment against potential toxicity. These findings require validation in clinical circumstances.

In many countries, pre-term infants are frequently admitted to the neonatal intensive care unit. Because of this, they frequently suffer infections with nosocomially acquired organisms (15, 24, 25). Meropenem, because of its broad-spectrum activity, which includes the majority of nosocomially acquired pathogens, would be an agent of great utility in these circumstances.

Unfortunately, little information is available regarding the disposition of antimicrobial agents in the neonatal patient population (22). This information is critical if the clinician is to be able to prescribe the correct dose and schedule of an antimicrobial agent that will minimize toxicity and maximize the probability of a good outcome. For β-lactam antibiotics, the work of a number of laboratories has indicated that the time that plasma concentrations exceed the MIC of the pathogen is the pharmacodynamic variable most closely linked to clinical outcome (10). Therefore, the dosing interval which is selected for antimicrobial administration will have a major impact on the adequacy of the dose chosen.

Newborns are well known to have clearances of drugs which differ from those seen in children and adults (2). Much of this difference is attributable to the maturation of both renal and nonrenal clearance pathways. Furthermore, volumes of distribution (V) tend to be larger, when adjusted for weight, in newborns than those seen in older children and adults (2). Consequently, any evaluation of a new agent which does not take into account physiological differences of newborn infants relative to older children or adults will not be able to provide optimal information to the clinician that will allow the choice of the proper dosing interval to maximize the time above the MIC (Time>MIC).

Because meropenem is such a potentially valuable agent for the neonatally infected group, we decided to examine both full-term and pre-term infants for their dispositions towards meropenem. We report here the single-dose pharmacokinetic parameters seen after doses of 10, 20, and 40 mg/kg of body weight of meropenem in both pre- and full-term newborns.

MATERIALS AND METHODS

Patients and study design. Thirty-eight newborn infants were studied at two centers: the Neonatal Intensive Care Units of the Erasmus MC-Sophia Children’s Hospital, Rotterdam, The Netherlands, and Charles University, Prague, Czech Republic. For inclusion in this clinical investigation, potential participants were required to meet the following criteria. (i) Participants had to be hospitalized pre-term and full-term neonates in their first 4 weeks of life who had received a minimum of 48 h of antibiotic therapy for a known or presumed bacterial infection. (ii) The subject's overall condition needed to be good or fair. (iii) Parents or legal guardians needed to have given written informed consent.
IV. The participants were not to meet any of the following exclusion criteria: (a) so severely ill that they were not likely to survive the duration of the trial; (b) born to mothers who are known or suspected to be positive for human immunodeficiency virus or have hepatitis; (c) born to mothers who are addicted to drugs or alcohol; (d) having a major congenital abnormality; (e) having a known history of immediate hypersensitivity to any β-lactam antibiotic; (f) having seizure within the previous 3 days; (g) having uncorrected bilirubin concentrations sufficiently high enough to warrant exchange transfusion; (h) having received ceftriaxone or cefotaxin (known to interfere with the meropenem assay) within the previous 3 days; (i) having plasma creatinine values of >140 μmol/liter; (j) having received a systemic investigational drug; or (k) having any condition which in the opinion of the investigator made the subject unsuitable for enrollment. All studied infants continued to receive their standard antibiotic regimens. In addition, a single dose of meropenem was administered as a 30-min intravenous infusion. The neonates were divided into a pre-term group (29 to 36 weeks of gestational age; n = 23) and a group of full-term infants (37 to 42 weeks of gestational age; n = 15). Pre-term and full-term infants were further randomly subdivided into three subgroups each. Each of the three groups was administered one of the following doses: 10 mg/kg, 20 mg/kg, or 40 mg/kg of meropenem.

Informed consent. Written informed consent was obtained from the parents of the infants according to institutional guidelines.

Sampling scheme and sample handling. Blood samples (200 μl) were collected immediately before and at the following time points after the start of the infusion: 0.25, 0.5, 0.75, 1, 2, 4, 8, 12, and 24 h. Blood samples were collected into NH4-heparinized tubes and shaken slightly. Blood samples were kept cold (4°C) for at least 3 but no more than 15 min under centrifugation (4°C for 10 min at 3,500 rpm). The plasma was split and transferred into two plastic tubes. Samples were quickly frozen on dry ice and maintained on dry ice until stored at −70°C. The creatinine clearance was calculated as follows: $\text{CL}_{\text{cr}} = 0.45 \cdot \text{crown}-\text{heel length}/\text{plasma creatinine}$ (2).

Pharmacokinetic methods. Population pharmacokinetic parameter estimation for meropenem was performed using the NPAG (non-parametric adaptive grid with adaptive γ) program package developed by Leary et al. (20). This program provides maximum-likelihood estimates of the population mean pharmacokinetic parameter values and their distributions without making assumptions as to the shape of the underlying distributions. One- and two-compartment open models with zero-order input and first-order elimination from the central compartment were evaluated. For observation weighting, the inverse of the assay variances was used as weights.

The retention times of meropenem and cefpodoxime were 9.0 and 12.8 min, respectively. The creatinine clearance was calculated as follows: $\text{CL}_{\text{cr}} = 0.45 \cdot \text{crown}-\text{heel length}/\text{plasma creatinine}$ (2).
from 29 to 36 weeks with weights ranging from 952 to 2,830 g and the crown-heel length ranging from 36 to 47.5 cm. Postnatal ages ranged from 2 to 28 days of life. Creatinine clearance, calculated according to the method of Schwartz et al. (30), ranged from 13.3 to 34.2 ml/min/1.73 m². Nine pre-term infants received 10 mg/kg of meropenem, while 8 received 20 mg/kg and 6 received 40 mg/kg of meropenem. The full-term infants consisted of 10 males and 5 females. Their gestational ages ranged from 2 to 14 days of life. The creatinine clearances of interfering with meropenem pharmacokinetics are as follows: amoxicillin, ampicillin, azlocillin, cefuroxime, cefotaxime, ceftazidime, oxacillin, ticarcillin, and furosemide.

**TABLE 2. Demographic data and sequence of treatments of hospitalized full-term neonates**

| Center/  |
| --- | --- | --- | --- | --- | --- |
| Gestational  |
| Wt (kg) | Crown-heel | CLcr | Dosing |  |
| patient no | age (wk) | length (cm) | (ml/min/1.73 m²) | (mg/kg) |  |
| 0001/0110 | M | 40 | 3.26 | 52.0 | 51.7 | 40 |
| 0001/0102 | F | 39 | 3.02 | 47.0 | 60.3 | 20 |
| 0001/0103 | M | 38 | 3.42 | 53.0 | 39.8 | 10 |
| 0001/0104 | M | 39 | 3.22 | 53.0 | 39.8 | 20 |
| 0001/0105 | M | 38 | 3.02 | 47.0 | 41.6 | 40 |
| 0001/0106 | M | 38 | 2.82 | 49.0 | 40.6 | 10 |
| 0001/0107 | F | 41 | 3.56 | 51.0 | 56.4 | 20 |
| 0001/0108 | M | 38 | 3.19 | 49.0 | 41.5 | 10 |
| 0001/0109 | F | 39 | 3.15 | 49.0 | 48.7 | 40 |
| 0001/0110 | F | 40 | 4.00 | 52.0 | 48.1 | 10 |
| 0001/0111 | M | 39 | 2.95 | 48.0 | 49.0 | 40 |
| 0001/0112 | M | 39 | 4.05 | 50.0 | 34.3 | 20 |
| 0002/0101 | M | 42 | 2.47 | 49.0 | 55.7 | 40 |
| 0002/0102 | F | 37 | 2.34 | 43.0 | 20.1 | 20 |
| 0002/0103 | M | 38 | 3.12 | 50.0 | 31.6 | 10 |

**Mean**

| 39 | 3.17 | 49.5 | 43.9 |
| 1 | 0.47 | 3 | 10.5 |
| 37 | 2.34 | 43 | 20.1 |
| 42 | 4.05 | 53 | 60.3 |

**SD**

**Min**

**Max**

\[
\text{CLcr} = 0.45 \cdot \text{crown-heel length/plasma creatinine (2)}
\]

For meropenem plasma clearance, the demographic variables chosen as descriptors were as follows:

\[
\text{Clearance} = 0.0133 \times \text{CLcr} + 0.1088 \times \text{Wt} - 0.261
\]

In the equation, volume is in liters and CLcr is in units of ml/min/1.73 m² (estimated by the Schwartz formula [23]), and weight (Wt) is in kg. It should be noted that weight along with height are included in the Schwartz formula. Nevertheless, the estimator was able to find weight as affecting clearance in addition to the Schwartz formula-estimated creatinine clearance. Overall, the regression was highly statistically significant. The \( P \) value for CLcr was 0.000017 and for Wt was 0.036. The regression relationship explained 67.9% of the variance (i.e., \( r^2 = 0.679 \)). The relationship was the same when performed stepwise forward or stepwise backward.

For \( V \), the physiological/demographic variable chosen was CLcr.

\[
\text{Volume} = 0.0278 \times \text{CLcr} + 0.147
\]

In the equation, volume is in liters and CLcr is in units of ml/min/1.73 m². Again, the relationship was highly statistically significant, with a \( P \) of <0.0001. The relationship explained 35.2% of the variance (\( r^2 = 0.352 \)). The general linear model procedure identified a larger model (including weight) stepwise backward but only CLcr stepwise forward. To be conservative, we employed the stepwise forward procedure.

**Population modeling.** The population model for all infants \((n = 38)\), performed without covariates, had a maximum-likelihood score of \(-655.7\). A general linear model was developed, relating both volume of the central compartment and clearance to covariates (see above). Volume was related to estimated creatinine clearance. Clearance was related to weight and estimated creatinine clearance.

Population analyses were performed with the volume and clearance related to the covariates as indicated below:

\[
\text{Clearance} = \text{Clearance}_{\text{int}} + A \times \text{Wt} + B \times \text{CLcr}
\]

\[
\text{Volume} = \text{Volume}_{\text{int}} + C \times \text{CLcr}
\]

The likelihood scores for the base model and the expanded models are shown in Table 3. The fit of the full model to the data was good. After the Bayesian step, the \( r^2 \) was 0.977; the
The fit of the full model prior to the Bayesian step was also quite acceptable. The $r^2$ was 0.742. Observed meropenem concentrations were calculated as follows: $1.037 \times$ predicted meropenem concentrations $-0.096$. Bias was $-0.154$ mg/liter; precision was 1.003 (mg/liter)$^2$. This is displayed in Fig. 2.

The final parameter values for the integrated analysis of all 38 neonates are presented in Table 4.

More information regarding the $V$ and plasma clearance can be obtained by examining the ranges observed for these parameters after the Bayesian step for the two populations. For the $V$, the 20% to 80% bounds on volume for the pre-term and full-term groups are 0.44 to 0.98 and 0.82 to 1.54 liters, showing considerable overlap. However, when one examines the distributions for clearance, the full-term children have a 20% to 80% range of 0.414 to 0.753 liters/h, whereas for pre-term

---

**TABLE 3.** Likelihood scores of competing models$^a$

<table>
<thead>
<tr>
<th>Model</th>
<th>Likelihood score</th>
<th>$2\times$ likelihood difference</th>
<th>df</th>
<th>$P$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Base model</td>
<td>$-655.7$</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wt and CL$_{cr}$ (in clearance)</td>
<td>$-648.8$</td>
<td>13.8</td>
<td>2</td>
<td>$&lt;0.001$</td>
</tr>
<tr>
<td>CL$_{cr}$ (in vol)</td>
<td>$-648.4$</td>
<td>0.8</td>
<td>1</td>
<td>NS</td>
</tr>
</tbody>
</table>

$^a$ df, degrees of freedom; NS, not significant.
40% of the dosing interval (11). Thus, 40% Time kill occurs when the Time dispersion. For carbapenem antibiotics, organism maximal cell estimates of the mean parameter values and the estimates of MIC.

Meropenem is distributed in extracellular water and is excreted mainly by glomerular filtration. Therefore, changes in body water and development of renal function influence the disposition of meropenem. Meropenem has a larger V and lower clearance in premature neonates, and even more so compared to adults, and dosing regimens thus have to be adjusted accordingly. Improvement of clearance follows the gestational age- and postnatal age-dependent increases in the glomerular filtration rate (9). Other factors in the neonatal intensive care unit that directly influence V or renal function, such as extracorporeal membrane oxygenation or exposure to indomethacin, were shown to significantly alter the neonatal pharmacokinetics of other primarily renally excreted drugs (7, 8, 9, 27).

Meropenem pharmacokinetics have been reported in seven pre-term neonates, showing a V of 0.74 (range, 0.24 to 1.2) liters/kg (30). The half-life was circa 3.4 h, which is substantially longer than the 1-h rate in adults (21). From the previous study, those authors concluded that, because of the increased half-life, a two-times-daily meropenem dose of 15 mg/kg would suffice. Most recently, Bradley and colleagues (4) studied 37 neonates and administered single doses of 10 and 20 mg/kg of meropenem as a 30-min infusion. These authors demonstrated that an 8-h interval may be more appropriate for organisms with higher MICs. They found that a 20-mg/kg, 8-h dose would provide robust coverage for their patients but that 40 mg/kg may be necessary for some infections with more resistant pathogens, like Pseudomonas aeruginosa. It should be noted, however, that these data were based on very sparse sampling, where any one patient had three blood samples obtained on one of two schedules. In contrast, the patients reported here had seven to nine samples obtained for analysis.

Here we report the results of a single-dose pharmacokinetic study of 38 newborn infants (23 pre-term and 15 full-term) using three different doses (10, 20, and 40 mg/kg) of meropenem intravenously. Because of the limits on blood withdrawal and because of ethical considerations, it was only possible to perform this study on newborns that were already infected and required antimicrobial therapy. Because of this, it is clear that there may have been some competition, particularly at the renal tubular level between drugs being adminis-

**DISCUSSION**

Neonatal sepsis remains one of the main causes of mortality and morbidity of newborn infants admitted to a neonatal intensive care unit (1, 14, 16, 18). Furthermore, invasive infections such as pneumonia, meningitis, and necrotizing enterocolitis threaten the newborn infant.

Meropenem is distributed in extracellular water and is excreted mainly by glomerular filtration. Therefore, changes in body water and development of renal function influence the disposition of meropenem. Meropenem has a larger V and lower clearance in premature neonates, and even more so compared to adults, and dosing regimens thus have to be adjusted accordingly. Improvement of clearance follows the gestational age- and postnatal age-dependent increases in the glomerular filtration rate (9). Other factors in the neonatal intensive care unit that directly influence V or renal function, such as extracorporeal membrane oxygenation or exposure to indomethacin, were shown to significantly alter the neonatal pharmacokinetics of other primarily renally excreted drugs (7, 8, 9, 27).

**TABLE 4. Final model pharmacokinetic parameter values of meropenem in hospitalized pre-term and full-term neonates**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Vol (liter)</th>
<th>Kmi (h⁻¹)</th>
<th>Kni (h⁻¹)</th>
<th>CLSLPPr (liter/h/m/1.73 m²)</th>
<th>CLSLPwni (liter/kg)</th>
<th>CL-INT (liter/h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>0.969</td>
<td>7.71</td>
<td>30.0</td>
<td>0.00112</td>
<td>0.0925</td>
<td>0.156</td>
</tr>
<tr>
<td>Median</td>
<td>0.883</td>
<td>5.862</td>
<td>41.6</td>
<td>0.00510</td>
<td>0.0918</td>
<td>0.172</td>
</tr>
<tr>
<td>Mode</td>
<td>0.896</td>
<td>0.100</td>
<td>44.8</td>
<td>0.00510</td>
<td>0.0460</td>
<td>0.0248</td>
</tr>
<tr>
<td>SD</td>
<td>0.527</td>
<td>6.67</td>
<td>17.9</td>
<td>0.00151</td>
<td>0.0606</td>
<td>0.122</td>
</tr>
</tbody>
</table>

*The parameter values are for the analysis of both pre- and full-term neonates. Simulations for the two subpopulations employ the separate weight and estimated creatinine clearance distributions which are shown in Tables 1 and 2. Vol, volume of the central compartment; Kmi and Kni, first-order intercompartmental transfer rate constants; CLSLPPr, the clearance slope term with estimated creatinine clearance; CLSLPwni, the clearance slope term with weight; CL-INT, the clearance intercept term. The estimate for meropenem clearance from the mean values is given by the following equation: CL = 0.156 + 0.00012 · CLcre + 0.0925 · Wt.*

FIG. 2. Predicted observed plot for meropenem concentrations after the Bayesian step for the total population, using the final model (Cfcr and weight in meropenem clearance—see Table 3).
tered at approximately the same period. We attempted to
minimize such an interaction by not administering the merop-
penem at a time when concentrations of any other competing
agent might be high. Nevertheless, it must be understood that
some interference may have taken place. However, it should
also be understood that the pharmacokinetics of meropenem
were determined in a clinically realistic setting.

The population modeling that we undertook demonstrated
clearly that meropenem $V$ values were in the range of those
reported previously for other agents of the $\beta$-lactam class.
Examination of the probability distribution of the volume for
the pre-term versus the full-term groups of newborns showed
that the means were similar and that the full-term infants,
somewhat surprisingly, were more variable in their $V$.

The clearances were significantly different in the two groups.
As can be seen by examining Tables 1 and 2, pre-term infants
had calculated creatinine clearances which were clearly less
than those seen in the full-term neonatal group. This is to be
expected on the basis of their gestational age (19, 27, 28, 29).
Indeed, when one compares the probability distribution of
clearance for the full population versus each of the two sub-
populations, it is clear that there is at most a 15 to 20% overlap
between the pre-term and full-term groups. We hypothesized
that this nonoverlap of meropenem plasma clearance was due,
mainly, to the differences in estimated creatinine clearances in the two patient populations.

These between-group differences need to be taken into account when designing dosing regimens that would have a high likelihood of being efficacious in the empirical therapy setting. Multiple investigators (5, 13) have demonstrated that Time > MIC is the pharmacodynamic variable most closely linked to outcome for β-lactam antibiotics. Consequently, it is important to be able to make dosing recommendations which the clinician can employ at the bedside to generate concentrations of meropenem in the plasma which remain above the MIC of clinically important pathogens for a relatively high percentage of the dosing interval.

Monte Carlo simulation combined with target attainment rate analysis has been introduced as a way of rationally evaluating dose and schedule as well as setting MIC breakpoints (3), and it has been reviewed for its applicability to children (17). This approach has been prospectively validated in a number of circumstances, both for bacteria and viruses (3, 5, 12).

We evaluated meropenem at a prospective dose and schedule of 20 and 40 mg/kg every 8 and 12 h using the pre-term and full-term population pharmacokinetics (Fig. 3). Maintaining free-drug concentrations (meropenem is approximately 2% bound) of >MIC for 40% of a dosing interval achieves a maximal cell kill (11). Examination of Fig. 3 demonstrates that a dose of 40 mg/kg with an 8-h dosing interval produces target attainment rates of >90% for achieving maximal cell kill at 8 mg/liter for both pre-term and full-term infants. Use of a
prolonged (4-h) infusion markedly improves target attainment rates for the 20-mg/kg dose. Obviously, antimicrobial chemotherapy is a balance between efficacy and toxicity. Meropenem has little in the way of concentration-dependent toxicity, but the balance should be explicitly judged when the decision for a 20-mg/kg versus a 40-mg/kg dose is to be made.

As indicated above, previous work in a smaller number of neonates (30) resulted in a recommendation for a 12-h dosing interval. It is likely that such a recommendation would be successful for infections where non-pseudomonal or non-<em>Acinetobacter</em> isolates were being treated, where MICs would be highly likely to be \( \leq 2 \) mg/liter. The circumstance of the infection becomes clear after pathogen identification. It may be prudent, then, to choose an 8-h interval until the pathogen is identified and an MIC is obtained.

These data indicate that 20 to 40 mg/kg with an 8-h dosing interval should provide robust coverage for the vast majority of nosocomially acquired pathogens seen in this population. If the 20-mg/kg dose is chosen, consideration should be given to the prolonged (4-h) infusion. Care needs to be exercised for clinicians in settings where there is a high likelihood of methicillin-resistant <em>Staphylococcus aureus</em>, as meropenem would not be adequate empirical therapy for this pathogen. In addition, meropenem has been given to children at doses of 40 mg/kg every 8 h and has been well tolerated (empirical treatment of meningitis). The clinician’s choice of dose, schedule, and interval should be weighted upon the probability of target attainment versus the probability of toxicity, modulated by the MIC distribution of likely pathogens present in their specific institution.

Finally, given the rate of loss of antibiotics from the physician’s armamentarium due to resistance, regimens that help suppress this resistance should be considered, if the toxicity price is not great. Figure 4 shows the target attainment for achieving a trough value of free drug 1.7 times a baseline MIC (combination therapy with an aminoglycoside) and 6.2 times a baseline MIC (monotherapy with meropenem), a target proposed by Tam et al. (26) for resistance suppression for <em>Pseudomonas aeruginosa</em>. For an 8-h interval and a 4-h infusion with a 40-mg/kg dose, this target is met \( >80\% \) of the time for both pre-term and full-term neonates out to an MIC of 2 mg/liter when combined with an aminoglycoside, which encompasses the vast majority of the wild-type population sensitivity of <em>Pseudomonas aeruginosa</em> to meropenem. For monotherapy, the resistance suppression goal falls below 79\% after an MIC of 1.0 mg/liter. This provides another reason to favor the large dose every 8 h with a 4-h infusion, at least empirically.

These findings need to be prospectively validated in the clinic. Furthermore, we would like to stress that the aforementioned recommendations are primarily derived from pre-term infants with gestational ages of more than 30 weeks, because only one infant was studied with a gestational age of less than 30 weeks.

Data on the pharmacokinetics of meropenem in pre-term infants with gestational ages of less than 30 weeks are still needed before doses and schedules of meropenem can be calculated at the bedside for this very young population.

**ACKNOWLEDGMENTS**

We have no conflicts to declare. The work was supported by AstraZeneca Pharmaceuticals.

**REFERENCES**