Tolerability and Outcomes of Kinetically Guided Therapy With Gentamicin in Critically Ill Neonates During the First Week of Life: An Open-Label, Prospective Study

Jirina Martínková, MD, PhD[1]; Pavla Pokorná, MD[2]; Jiri Záhora, PhD[3]; Jaroslav Chládek, PhD[1]; Václav Vobruba, MD[2]; Iva Selke-Krulichová, PhD[3]; and Jirina Chládková, MD[4]

[1]Department of Pharmacology, Charles University in Prague, Faculty of Medicine in Hradec Králové, Hradec Králové, Czech Republic; [2]Intensive Care Unit, Department of Pediatrics and Adolescent Medicine, General Teaching Hospital, Prague, Czech Republic; [3]Department of Biophysics, Charles University in Prague, Faculty of Medicine in Hradec Králové, Hradec Králové, Czech Republic; and [4]Department of Pediatrics, Charles University in Prague, Faculty of Medicine in Hradec Králové, Hradec Králové, Czech Republic

ABSTRACT

Background: Aminoglycosides are bactericidal antibiotics used worldwide for the treatment of serious infections in critically ill patients, including neonates. Critically ill neonates constitute a unique challenge in dosing owing to the pathologic alterations that accompany severe illness and the rapidly changing conditions of these patients.

Objectives: The main objective of this study was to analyze the kinetically guided dosage adjustment of gentamicin in neonates critically ill during the first week of life based on plasma concentrations after the first dose and to identify the impact of covariates (eg, fluid intake, body fluid retention) with respect to gestational age (GA). Tolerability of therapy was also assessed.

Methods: This 10-day, open-label, prospective study included neonates critically ill during the first week of life admitted to the neonatal intensive care unit of a children's hospital between January 2006 and July 2009. Hearing and renal assessments were conducted over a 24-month follow-up period. The patients were treated with gentamicin for suspected sepsis, proven sepsis, or pneumonia as an early sign of sepsis. The first and second doses of gentamicin 4 mg/kg were adjusted according to birth weight and GA: group 1 (GA $<$ 34 weeks), 48-hour interdose intervals; group 2 (GA 34–38 weeks), 36 hours; and group 3 (GA $>$ 38 weeks), 24 or 48 hours. Individual pharmacokinetic parameters were estimated after the first dose (given in 30-minute intravenous infusions) using 4 concentrations. Individual pharmacokinetic parameters were estimated by fitting the parameters of a 2-compartment model into 4 concentrations. The last 2 blood samples were taken 30 minutes before the fourth infusion ($C_{trough,3}$) and 1 hour after its start ($C_{max,4}$). Dosing was individualized to reach target ranges for the $C_{trough,3}$ (0.5–2.0 mg/L) and $C_{max,4}$ (6–10 mg/L) values. If needed, initial dosing was changed after the second dose by adjusting (reducing or increasing) the third and subsequent doses, or by adjusting (prolonging or shortening) the interdose intervals. $C_{trough,3}$ and $C_{max,4}$ were assessed to determine differences between predicted and assayed values. Fluid retention was registered as the difference between fluid intake and urine output at different intervals related to the first dose per kilogram of birth weight, and from the start of the first infusion (0 hour) to the day of the fourth infusion. The C_{max}/minimum inhibitory concentration (MIC) ratio was determined for assessment of optimal response. Tolerability was evaluated during the 24-month follow-up period using renal sonography to screen for nephrocalcinosis and transient evoked otoacoustic emission recordings to evaluate hearing abnormalities.

Results: A total of 84 neonates (all white; 53 males, 31 females; birth weight range, 0.8–4.56 kg; GA range, 24–42 weeks) were enrolled in 3 groups: group 1, GA $<$ 34 weeks, n = 27; group 2, GA 34–38 weeks,

Accepted for publication October 4, 2010.
doi:10.1016/j.clinthera.2011.01.013
0149-2918/$ - see front matter

© 2010 Elsevier HS Journals, Inc. All rights reserved.

n = 22; and group 3, GA > 38 weeks, n = 35. The C_{max} value detected 1 hour after the start of the first infusion ($C_{max,1}$) reached the target range of 6–10 mg/L in 66 of the 84 neonates (79%). After the initial dose, $C_{max,1}$ was variable (%CV, 29%); the failure rate to reach 6 mg/L was 13%. V_d decreased with GA (r = −0.30, $P < 0.01$) and achieved mean (SD) rates of 0.51 (0.10), 0.48 (0.13), and 0.40 (0.15) L/kg in groups 1, 2, and 3, respectively. Neither C_{max} nor V_d was correlated with fluid intake relative to the first infusion. Mean gentamicin clearance measured after dose 1 (0.47 [0.23], 0.66 [0.26], and 0.76 [0.32] mL/min/kg) increased with GA (r = 0.45, $P < 0.001$). The interdose interval was prolonged after the second and subsequent infusions in 8 of 84 neonates (10%) or by decreasing the third dose and subsequent doses in 51 neonates (61%). The target $C_{max,4}$ and $C_{trough,3}$ values occurred in 63% (22 of 35) and 83% (29 of 35) of full-term patients (GA >38 weeks), respectively. In preterm neonates, the target range for $C_{max,4}$ was reached in 11 of 27 patients (41%) in group 1 and 11 of 22 patients (50%) in group 2; for $C_{trough,3}$, the target range was reached in 25 patients (93%) in group 1 and in 16 (73%) in group 2. $C_{trough,3}$ >2 mg/L was detected in 1 full-term neonate, and gentamicin was withdrawn. Suspected fluid retention within the time period of 0 hour to the day of the fourth infusion was well correlated with actual body weight (r = 0.58, $P < 0.001$), but it was negatively correlated with $C_{max,4}$ (r = −0.25, P = 0.02). Thirteen of the 84 neonates (15%) had confirmed sepsis. C_{max}/MIC was >12 except for 2 resistant staphylococcal infections (C_{max}/MIC = 0.4); amikacin and vancomycin were substituted for gentamicin in these cases. Clinical signs and laboratory data indicative of suspected sepsis disappeared in 5 to 10 days in 68 of 71 neonates. In 1 neonate, gentamicin was withdrawn after dose 4 because of a high $C_{trough,3}$ value. In the 3 remaining neonates, C-reactive protein was decreased >10 days without changing therapy. Two neonates died, 1 of severe hypoxic-ischemic encephalopathy as a consequence of perinatal asphyxia and another of stage IV intraventricular hemorrhage. Transient renal dysfunction attributable to gentamicin was detected in 1 case. No signs of late toxicity (nephrocalcinosis) were found during the second year of follow-up. Two neonates were diagnosed with unilateral hearing loss, a secondary phenomenon of hypoxic-ischemic encephalopathy thought to be related to the severe perinatal asphyxia.

Conclusions: The initial dose of gentamicin 4 mg/kg for these critically ill premature and mature neonates with sepsis during the first week of life was high enough to reach bactericidal $C_{max,1}$ within 6–10 mg/L. $C_{max,1}$ <6 mg/L occurred in 13% of neonates. The interdose interval modified according to the recommendation resulted in C_{trough} values within the target range of 0.5–2.0 mg/L in all but 2 neonates. The kinetically guided maintenance dosing of gentamicin based on plasma concentrations after the first dose should be optimized, taking into account actual body weight. (EudraCT number: 2005-002723-13). (*Clin Ther.* 2010;32:2400–2414) © 2010 Elsevier HS Journals, Inc.

Key words: aminoglycosides, gentamicin, neonates, sepsis, tolerability.

INTRODUCTION

Aminoglycosides are bactericidal antibiotics used worldwide for the treatment of serious infections in critically ill patients, including neonates.[1] Their therapeutic efficacy is correlated more with C_{max} values than with dose.[2] The effective value of C_{max} (and its range) is derived from the minimum inhibitory concentration (MIC) determined in vitro for the particular microorganism. To reach a therapeutic drug level, the C_{max}/MIC ratio for a widely prescribed aminoglycoside (eg, gentamicin) should be within 8 to 12.[3,4] However, the clinical use of aminoglycosides is limited by toxicity. Nephrotoxicity and ototoxicity are reportedly more likely to occur after repeated exposure and prolonged courses of therapy with aminoglycosides. Both toxicities reportedly were reduced if the plasma concentration detected 30 minutes before the infusion (C_{trough}) is <2 mg/L,[2,5–7] whereas transiently elevated C_{max} concentrations (>12 mg/L) do not appear to influence the risk of toxicity.[8]

Optimal dosing with aminoglycosides such as gentamicin requires a thorough knowledge of their pharmacokinetics and pharmacodynamics. In published studies,[9–15] a wide interindividual and intraindividual variability in efficacy has been found repeatedly, mainly in critically ill patients and ascribed by the authors to factors (covariates) strongly influencing aminoglycoside pharmacokinetics. The extended V_d of aminoglycosides during sepsis and septic shock, and the ensuing subtherapeutic plasma concentrations achieved after the first infusion, have been reported in earlier studies focused on

adult patients postsurgery or those suffering from sepsis.[9–11] In critically ill patients, a rapidly changing and extended V_d of aminoglycosides[11,12] is attributed to a large volume of fluid administered as prevention against hypotension[13] or to fluid retention as a part of a homeostasis response to the failing heart during sepsis.[12] Fluid retention also may be a consequence of renal failure.[12,13] In contrast, a decrease in V_d and a higher aminoglycoside clearance (CL) are associated with improvement of the patient's condition owing to successful therapy.[14] Such continuously changing conditions, which are difficult to identify, have led to the view that the therapeutic benefit of gentamicin is uncertain or even unpredictable.[15,16]

In light of these findings, fluid intake (including volume expansion) and body fluid retention might be important pathophysiologic covariates that should be taken into consideration when treating neonates critically ill with sepsis. To the best of our knowledge, the importance of these covariates on the pharmacokinetics of aminoglycosides in neonates has not yet been reported. The main goal of the present study was to analyze the kinetically guided dosage adjustment of gentamicin in neonates critically ill during the first week of life based on plasma concentrations after the first dose and to identify the impact of covariates (eg, fluid intake, body fluid retention) with respect to gestational age (GA). Tolerability of therapy was also assessed.

PATIENTS AND METHODS

This prospective study (January 2006–July 2009) enrolled neonates admitted to the Neonatal Intensive Care Unit (NICU), Department of Pediatrics and Adolescent Medicine, General Teaching Hospital (Prague, Czech Republic), for suspected sepsis, proven sepsis, or pneumonia as an early sign of sepsis.

The study protocol was approved by the ethics committees of University Hospital in Hradec Králové and Charles University in Prague. Written informed consent was provided before treatment by 1 parent and 1 witness (a nurse). Inclusion criteria were confirmed as proven sepsis, suspected sepsis, and pneumonia. Exclusion criteria included enterococcal endocarditis, congenital defects, and acute renal failure defined as a serum creatinine level >1.5 mg/dL despite normal maternal function. This value is predominantly oliguric (urine output <1 mL/kg/h).[17,18]

Proven sepsis was defined as clinical signs of a systemic response to the infection and a positive blood culture confirmed by the MIC obtained before the start of antibiotic therapy (or later, if expected efficient outcome of therapy was not reached). The C_{max}/MIC ratio was determined to verify if the ratio was sufficient for optimal response. For gentamicin, C_{max}/MIC ratios of 8–12 are considered optimal for most infections caused by gram-negative microorganisms.[19]

Suspected sepsis was defined as clinical signs indicative of the systemic response with a negative blood culture prompting ≥5 days of antibiotic therapy.[20] C_{max} values ranging from 6–10 mg/L are considered adequate for bactericidal activity.[2] Clinical signs were evaluated according to clinical examination and supportive therapy (**Table I**) and were considered as follows: severe apnea requiring nasal continuous positive airway pressure or mechanical ventilation using biphasic positive airway pressure, ventilatory support, high-frequency oscillation ventilation or inhaled nitric oxide administration, cardiac failure, capillary refill (>4–5 seconds), or temperature instability (>38°C or <36.2°C); lethargy or hypotonia; feeding intolerance; and jaundice. *Laboratory sepsis screen* was defined as an immature/total neutrophil ratio >0.2 (evaluated according to GA and patent ductus arteriosus [PDA]); white blood cell count >25,000 or <5000/mm^3; and C-reactive protein (CRP) >10 mg/L. Pneumonia has also been considered one of the signs of the systemic response to infection.[21] CRP values in serum >10 mg/L were considered markers of sepsis.[22] Further signs of infection were early jaundice, thermoinstability, systemic hypoperfusion, arterial hypertension or hypotension, and a variable response to the initiation of therapeutic moderate whole-body hypothermia.

Early-onset sepsis was manifested in 72 hours after delivery, whereas late-onset sepsis appeared later (because 72 hours after delivery included the cases treated more than 10 days). The duration of sepsis treatment with antibiotics was dependent on clinical examination, laboratory parameters of infection, and culture-proven sepsis recovery. This therapy usually lasted for 5–10 days.

The therapeutic regimen in the NICU consisted of a combination of gentamicin with ampicillin 50 mg/kg IV every 12 hours or ampicillin/sulbactam 75 mg/kg/d divided every 12 hours for preterm neonates and 150 mg/kg/d divided every 12 hours for full-term neonates suffering from suspected sepsis, proven sepsis, or pneumonia (caused by gram-negative strains). Two initial (standard) doses of gentamicin and the first interdose interval were based on birth weight and GA as previously

Table I. Demographic and clinical characteristics of 84 neonates stratified according to gestational age (GA).

Characteristics	Group 1: GA <34 Wk (n = 27)	Group 2: GA 34–38 Wk (n = 22)	Group 3: GA >38 Wk (n = 35)
Sex, no.			
Male	18	13	22
Female	9	9	13
Mean (SD) GA, wk	29.9 (0.43)	35.8 (1.14)	40.0 (0.98)
Mean (SD) body weight at birth, kg	1.53 (0.43)*†	2.75 (0.50)‡	3.41 (0.58)
Perinatal asphyxia§ + HIE III, no. (%)	0	0	3 (9)
Perinatal asphyxia + HIE III + TH, no. (%)	0	0	14 (40)
Early asphyxia, no. (%)	6 (22)	5 (23)	10 (29)
Assisted ventilation using BIPAP, no. (%)	17 (63)	15 (68)	32 (91)
Assisted ventilation using HFOV, no. (%)	9 (33)	1 (5)	0
PH treated with inhaled NO, no. (%)	5 (19)	4 (18)	3 (9)
Cardiovascular failure, no. (%)	25 (93)	16 (73)	24 (69)
PDA treated with ibuprofen, no. (%)	12 (44)	4 (18)	0
IUGR, no. (%)	1 (4)	0	9 (26)

BIPAP = biphasic positive airway pressure; HFOV = high-frequency oscillation ventilation; HIE III = grade III hypoxic-ischemic encephalopathy; IUGR = intrauterine growth restriction; NO = nitric oxide; PDA = patent ductus arteriosus; PH = pulmonary hypertension; TH = therapeutic modest whole-body hypothermia.

* $P < 0.001$, group 1 versus 2.

† $P < 0.001$, group 1 versus 3.

‡ $P < 0.001$, group 2 versus 3.

§ Apgar score ≤4 was one of the parameters identifying perinatal asphyxia.

recommended.[23,24] The drug was given in 30-minute intravenous infusions via the peripheral vein (Alaris GS/GH/CC programmable pumps; Cardinal Health, Rolle, Switzerland). All infusions lasted for 30 minutes.

To determine gentamicin plasma concentrations after the first dose, 4 blood samples were taken from the central venous catheter or the peripheral arterial catheter different from that used to infuse the drug. The first postinfusion sample ($C_{max,1}$) was drawn at 1 hour after the start of the first infusion. Additional samples were taken at 6 and 12 hours (group 1) and at 12 and 24 hours (groups 2 and 3). The fourth sample was taken 30 minutes before the second dose ($C_{trough,1}$). Blood samples were collected into standard EDTA tubes, centrifuged (10 minutes at 4°C) within 1 hour after sampling, and stored for no longer than 2 days at –20°C. Gentamicin concentrations were assayed by a fluorescence polarization immunoassay (TDx Analyzer; Abbott Laboratories, Abbott Park, Illinois). The limit of quantitation was 0.3 mg/L, and the interday %CVs were 4.5% at 1 mg/L, 2.7% at 4 mg/L, and 2.4% at 8 mg/L. The laboratory was certified to perform gentamicin analyses by the Division for Drug Analyses (Referenzinstitut fur Bioanalytik, Bonn, Germany).

Individual pharmacokinetic parameters were estimated by fitting the parameters of a 2-compartment model[25] into 4 concentrations assayed after the first infusion using MW\Pharm program version 3.15A (Medi\Ware, Groningen, the Netherlands). These individual parameters were used to simulate the time profile of gentamicin concentrations and, starting from the interval after the second dose, to individualize the dosing regimen to achieve the target ranges related to the fourth dose: $C_{trough,3}$, detected 30 minutes before the fourth infusion (within the range of 0.5–2.0 mg/L), and $C_{max,4}$, detected 1 hour after the start of the fourth infusion (within the range 6–10 mg/L). The range of plasma concentration after the fourth infusion was chosen as the target for dosing prediction, as in many cases (group 1) the fourth dose of gentamicin was the last one given to the patients. After dose 4, the steady-state of plasma gentamicin

concentrations predicted was achieved in all but 3 preterm patients characterized by very low CL_1 values (gentamicin clearance estimated after dose 1). In these cases, 90%–95% of steady-state predicted values were determined. The last 2 blood sample concentrations ($C_{trough,3}$ and $C_{max,4}$) assayed could thus be compared with those predicted and validated by determining the predictive performance[26] (as described in the Statistical Analysis section).

Body fluid retention was estimated as the difference between fluid intake and urine loss measured every hour per day and kilogram of birth weight. Urine was collected by a catheter. Volumes of fluid lost by perspiration insensible, sweating (perspiration sensible), and feces were not considered; metabolic water was also not considered. The input-output difference in the fluid volume was presented as suspected body fluid retention and evaluated within the following time intervals: (1) from 2 hours to the start of the first infusion; (2) from −6 to 1 hour relative to the start of the first infusion; (3) from the start of the first infusion (0 hour) to 24 hours relative to the start of the first infusion; and (4) fluid retention from the start of the first infusion (0 hour) to the start of the fourth infusion.

Neonates were kept in warm humidified incubators (Draeger Medical GmbH, Lübeck, Germany [preterm neonates, groups 1 and 2]), warm beds (Alfamedic, Lišov, Czech Republic [full-term nonasphyxiated neonates]), and hypothermic Dartin beds equipped with the hypothermic Gaymar Medi-Therm III system (Gaymar Industries, Inc, made in Prague, Czech Republic [full-term asphyxiated neonates]). The temperature of the incubators was set up to maintain rectal body temperature within 36°C–37°C. Asphyxiated neonates were treated with whole-body hypothermia. The goal was to maintain rectal body temperature within 33°C–34°C for 72 hours. Rectal body temperature was monitored continuously. Systolic blood pressure (SBP) and diastolic blood pressure (DBP) were obtained from intra-arterial lines and registered simultaneously with cardiac examination every hour per day using an oscillometric device. Mean blood pressure was calculated as the sum of the DBP and one third of the pulse pressure, defined as the difference between the DBP and the SBP.[27] Heart rate was registered continuously using electrocardiography.

Laboratory examinations were conducted at the departments of Microbiology, Clinical Chemistry, and Hematology of the General Teaching Hospital (Prague, Czech Republic). Microbiologic cultures were analyzed using an automated continuous-monitoring blood culture system (Bactec 9240 Blood Culture Analyzer, Fluorescent Series; Becton Dickinson Microbiology Systems, Cockeysville, Maryland).

During the 24-month follow-up period after therapy termination, tolerability of kinetically guided therapy with gentamicin was monitored, with a focus on ototoxicity and nephrotoxicity. Hearing screening was conducted by means of 2 successive transient evoked otoacoustic emission recordings (using pure-tone testing between 0.25 and 8 kHz with a portable DANAX AS 72 Audiometer Echo-screen TDA [Widex Line, Prague, Czech Republic]), followed in cases of suspected hearing loss by brainstem auditory evoked potential recordings and otorhinolaryngology consultation.

The incidence of renal disorder was studied using criteria for evaluation of renal dysfunction[28] and nephrocalcinosis.[29] *Renal dysfunction* was defined as progressive oliguria <1 mL/kg/h and serum creatinine concentrations twice as high as the initial value; both were obtained within the interval determined by day of gentamicin dose 1 and dose 4 administration.[28,30] Signs of nephrocalcinosis were regularly examined by renal sonography using the Acuson Aspen Electric Medical Service (Acuson Aspen, Cyprus, Czech Republic).

Statistical Analysis

Standard statistical methods were used for the structural analysis of the data. Means, SDs, and minimal and maximal values were used to describe the data sets. As the first step before starting the inductive statistics, the D'Agostino normality test of the data was provided. When the resulting P value was >0.05, the 2-sample t test was then used for statistical decision with the Pearson correlation coefficient, and the corresponding P value was used to evaluate a linear relationship. When the resulting P value was <0.05, the Mann-Whitney test for statistical decision, the Spearman correlation coefficient, and the corresponding P value were used to evaluate a linear relationship. In all cases, $P < 0.05$ indicated statistical significance.

The accuracy and precision of predicting $C_{trough,3}$ and $C_{max,4}$ concentrations were evaluated using the mean prediction error (MPE) and the mean absolute prediction error (MAPE), calculated as follows: $MPE = \Sigma(C_{predicted} - C_{assayed})/N$ and $MAPE = SUM |(C_{predicted} - C_{assayed})|/N$.[26] The point estimates and 95% CIs were reported.

All calculations were conducted using the Kinetica 4.0 package (Inna Phase, Thermo Fisher Scientific Inc, Waltham, Massachusetts), MS Excel 2003 (Microsoft Corpora-

tion, Redmond, Washington; 1985–2003), and Statistica version 8.0 (StatSoft, Inc, Tulsa, Oklahoma; 1984–2007).

RESULTS

A total of 129 neonates were originally screened for inclusion in the study. Blood samples were missing in 23 neonates or gentamicin therapy was stopped very early (within 47 hours) in 18 neonates. Four patients died of organ failure not attributable to the drug. The final cohort thus included 84 neonates.

The demographic and clinical characteristics of the 84 neonates stratified into 3 groups according to GA are summarized in **Table I**. Some comorbidities were frequent in a particular group. For example, intrauterine growth restriction occurred in 26% and severe perinatal asphyxia with hypoxic-ischemic encephalopathy (HIE) treated with therapeutic modest whole-body hypothermia occurred in 40% of the full-term neonates in group 3; 44% of patients in group 1 had PDA. Early asphyxia occurred at comparable rates throughout all groups (22%, 23%, and 29% in groups 1, 2, and 3, respectively).

Antibiotic Therapy

Ampicillin plus gentamicin was given to 67 of 84 neonates (80%), and ampicillin/sulbactam plus gentamicin was given to 17 (20%). Intravenous vancomycin was given for 7–10 days at a dosage of 10 mg/kg every 12 hours (coadministered with ampicillin) and 15 mg/kg every 8 hours (in monotherapy) in 2 cases resistant to *Staphylococcus saprophyticus* and *Staphylococcus epidermidis*, respectively.

The first and second doses of gentamicin and the first interdose interval followed guidelines for standardized dosing, which take into account birth weight and GA (**Table II**). Kinetically guided dose adjustments resulted in modification of the standard dosing in 58 of the 84 neonates (69%). Decrease of dosing rate was reached as follows: the interdose interval was prolonged after the second and subsequent infusions in 8 of 84 neonates (10%) and/or the third and subsequent doses were decreased in 51 neonates (61%) (**Table III**). **Figure 1** illustrates one case in which both the interval after the second infusion was prolonged and the third dose was reduced to reach the target $C_{max,4}$ and $C_{trough,3}$ levels. The interdose interval was shortened after the second and subsequent infusions in 7 of 84 neonates (8%) .

Pharmacokinetics of Gentamicin After the First Dose

Individual profiles of plasma gentamicin concentration observed after the first standard dose of 4 mg/kg and the mean concentration time profile for each of the

Table II. Dosing of gentamicin, plasma concentration, and pharmacokinetic parameters estimated after the first standard dose of 4 mg/kg given to critically ill neonates with sepsis during the first week of life.

Characteristics	Group 1: GA <34 Wk	Group 2: GA 34–38 Wk	Group 3: GA >38 Wk
Initial dose, mg/kg	4.0	4.0	4.0
The first interdose interval, h	48	36	24 or 48*
V_{d1}, L/kg†‡	0.51 (0.10) [0.24–0.77]	0.48 (0.13) [0.31–0.76]	0.40 (0.15) [0.12–0.72]
CL_1, mL/min/kg†§	0.47 (0.23) [0.094–1.04]	0.66 (0.26) [0.1–1.06]	0.76 (0.32) [0.20–1.6]
$C_{max,1}$, mg/L†‖	7.26 (1.90) [4.7–15.6]	7.47 (1.5) [4.4–10]	8.75 (2.7) [4.3–16]
$C_{trough,1}$, mg/L†¶	1.09 (0.63) [0.30–3.5]	0.70 (0.36) [0.20–1.7]	0.96 (0.54) [0.30–3.1]

CL_1 = CL measured after dose 1; $C_{max,1}$ = plasma concentration detected 1 hour after the start of the first infusion; $C_{trough,1}$ = plasma concentration detected 30 minutes before the second infusion; GA = gestational age; Vd_1 = V_d measured after dose 1.

* In 6 neonates with asphyxia, the interval was prolonged from 24−48 hours.

† Mean (SD) [range].

‡ $P < 0.005$, group 1 versus 3.

§ $P < 0.001$, group 1 versus 2 and group 1 versus 3; $P = 0.07$ group 2 versus 3.

‖ $P = 0.02$, group 1 versus 3; $P = 0.04$, group 2 versus 3; $P = 0.67$, group 1 versus 2.

¶ $P < 0.01$, group 1 versus 2; $P = 0.046$, group 2 versus 3; $P = 0.19$, group 1 versus 3.

Table III. Kinetically guided adjustment of gentamicin dosing based on the individual pharmacokinetic characteristics estimated after the first infusion and simulation of the plasma concentrations detected 30 minutes before the fourth infusion ($C_{trough,3}$) and plasma concentrations detected 1 hour after the start of the fourth infusion ($C_{max,4}$).* Unless otherwise noted, values are given as mean (SD).

Characteristics of Dosing Regimen	Group 1: GA <34 Wk	Group 2: GA 34–38 Wk	Group 3: GA >38 Wk
No changes in the initial (standard) dosing regimen, n/N (%)	8/27 (30)	6/22 (27)	12/35 (34)
Decrease of dosing rate			
n/N (%)	16/27 (59)	16/22 (73)	19/35 (54)
By mg/kg/h	0.024 (0.010)	0.037 (0.025)	0.041 (0.020)
%	28 (13)	28 (14)	29 (12)
Decrease of the third and subsequent doses			
n/N (%)	16/27 (59)	16/22 (73)	19/35 (54)
By mg	1.6 (0.5)	2.7 (0.8)	3.1 (1.9)
%	29 (13)	24 (6)	23 (13)
Prolongation of the interdose interval after second and subsequent doses			
n/N (%)	0	5/22 (23)	3/35 (9)
By hour	—	14.4 (5)	12 (0)
%	—	50 (30)	50 (0)
Increase of dosing rate			
n/N (%)	3/27 (11)	0	4/35 (11)
By mg/kg/h	0.032 (0.022)	—	0.032 (0.025)
%	33 (17)	—	25 (17)
Increase of the third and subsequent doses			
n/N (%)	0	0	0
By mg	—	—	—
%	—	—	—
Shortening of the interdose interval after second and subsequent doses n/N (%)	3/27 (11)	0	4/35 (11)
By hour	12 (0)	—	12 (0)
%	25 (0)	—	25 (0)

GA = gestational age.

* Changes of initial (standard) dosing to obtain maintenance dosing as predicted according to individual kinetics of gentamicin. Changes are evaluated using dosing rate (milligram per kilogram per hour), doses (in milligrams), and interdose intervals (in hours). Every adjustment was also expressed as a percentage of the initial value.

3 groups are shown in **Figure 2.** Pharmacokinetic characteristics for V_d and CL measured after dose 1 (V_{d1} and CL_1, respectively), as well as the $C_{max,1}$ and $C_{trough,1}$ concentrations, are given in **Table II.** As expected, the mean value of V_{d1} normalized for body weight was the highest in group 1, followed by groups 2 and 3, respectively, whereas an opposite trend was observed for CL_1. There was a weak negative correlation between GA and the birth weight–normalized value of V_{d1} ($r = -0.30$, $P = 0.006$) with a modest correlation between GA and CL_1 ($r = 0.45$, $P < 0.001$). The $C_{max,1}$ values were numerically higher in full-term neonates

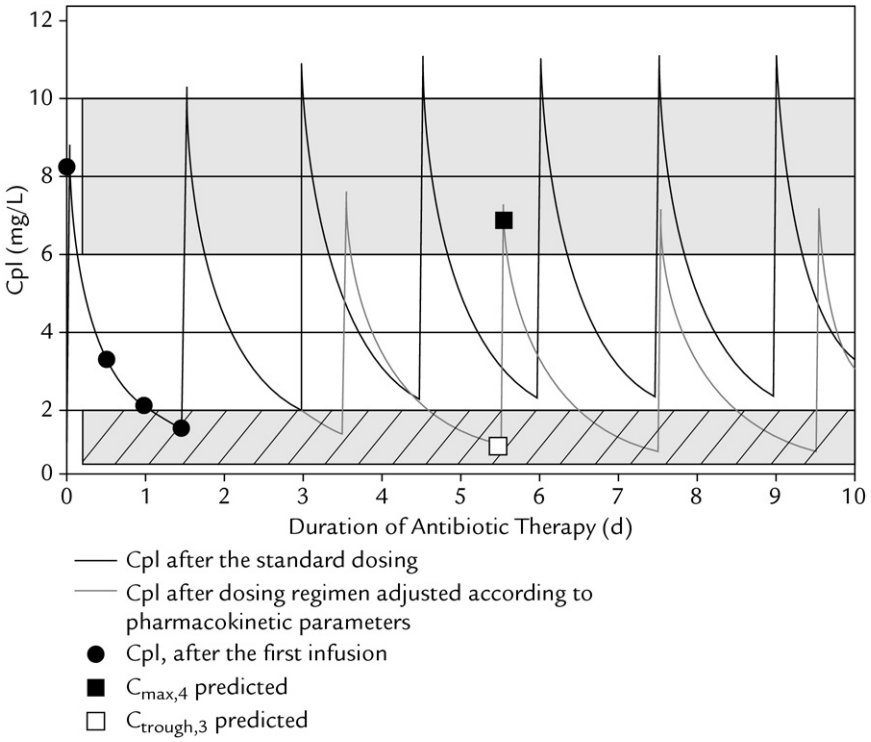

Figure 1. Fluctuation of plasma gentamicin concentration (Cpl). Simulation is based on individual gentamicin pharmacokinetic parameters estimated using gentamicin Cpl after the first initial (standard) dose. $C_{max,4} = C_{max}$ detected 1 hour after the start of the fourth infusion; $C_{trough,3}$ = Cpl detected 30 minutes before the fourth infusion.

(group 3) compared with both groups of premature neonates (Table II). In 66 of the 84 neonates (79%), the $C_{max,1}$ value reached the target range of 6–10 mg/L. In 11 neonates (13%), $C_{max,1}$ was <6 mg/L. The failure rate to reach this lower limit of the target window was low and comparable in all groups (11%, 14%, and 11%, respectively; P = NS). In 8 neonates (1 premature and 7 mature) (10%), $C_{max,1}$ was >10 mg/L. The $C_{trough,1}$ was <2 mg/L in all but 2 neonates.

There was no significant difference between groups in fluid intake over the 2-hour interval before the start of the first infusion (Table IV). It was, however, statistically higher in combined groups 1 and 2 (all preterm neonates) compared with the full-term neonates in group 3 (18.5 [10.5] vs 14.3 [7.44] mL/kg; $P = 0.04$). Body fluid retention in various intervals relative to the start of gentamicin dosing was numerically higher in preterm neonates from group 1 compared with the full-term neonates from group 3, whereas the volumes retained by preterm neonates from group 2 and the full-term neonates were comparable. Neither $C_{max,1}$ nor V_{d1} was significantly correlated with the intake of fluid in the interval from –2 hours to 0 hour or body fluid retention calculated from –6 hours to 1 hour relative to the start of the first infusion. Moreover, the intake of fluid and body fluid retention in neonates with $C_{max,1}$ <6 mg/L (n = 11) was not significantly different from those in neonates with higher $C_{max,1}$ (n = 73) (fluid intake, 12.5 [6.5] vs 17.4 [9.8] mL/kg; fluid retention, 3.7 [2.4] vs 4.8 [3.5] mL/kg). There were modest negative correlations between CL_1 and fluid retention in the intervals from –6 hours to 1 hour ($r = -0.31$, $P < 0.005$) and from 0 to 24 hours ($r = -0.37$, $P < 0.001$), respectively, but not between CL_1 and fluid intake from –2 hours to 0 hour.

Importance of Comorbidity Shown in PDA

Twelve of the 27 preterm neonates (group 1) and 4 of the 18 preterm neonates (group 2) experienced PDA (Table I); the remaining 15 neonates (group 1) and 18

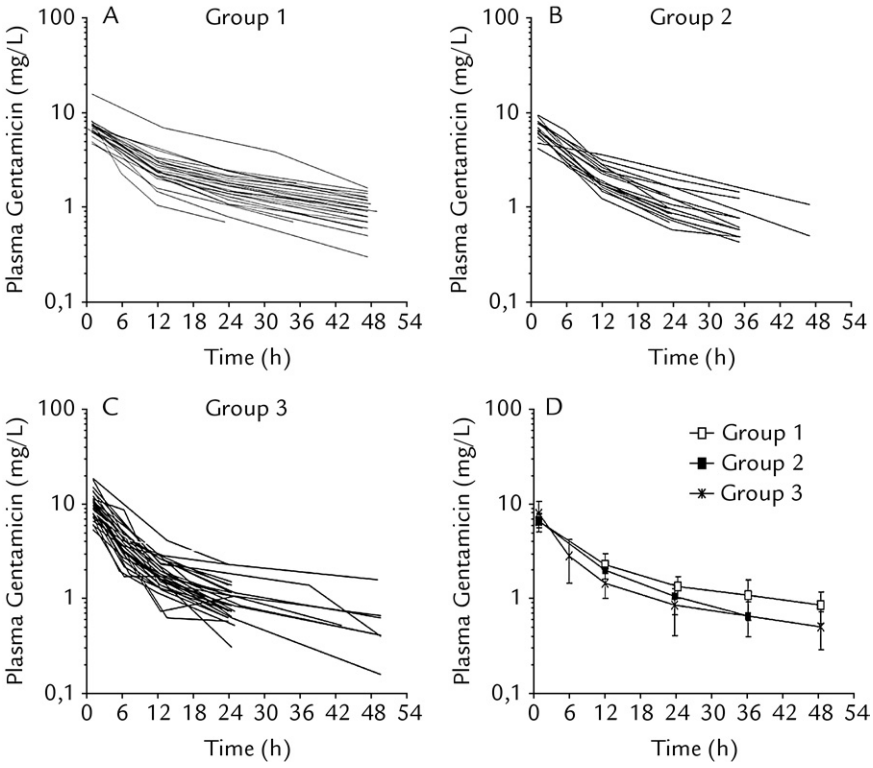

Figure 2. Individual and mean (SD) plasma profiles of gentamicin after a 30-minute IV infusion of 4 mg/kg to neonates with a gestational age of (A) <34 weeks (group 1), (B) 34–38 weeks (group 2), (C) >38 weeks (group 3), and (D) combined.

neonates (group 2) did not. Mean gentamicin CL reached 0.37 (0.13) mL/min/kg in the patients with PDA and 0.55 (0.26) mL/min/kg in those without PDA ($P = 0.004$). Body fluid retention from 0 hour to the day of the fourth dose was 356 (118) mL/kg in the former and 223 (98) mL/kg in the latter ($P = 0.048$).

Comparison of Assayed and Predicted Gentamicin $C_{max,4}$ and $C_{trough,3}$ Values

In all 3 groups, the assayed $C_{max,4}$ and $C_{trough,3}$ values were numerically lower compared with predictions. The MPE (95% CI) for $C_{max,4}$ was comparable between groups and was 0.83 (0.17–1.5), 1.1 (0.25–2.0), and 1.0 (0.2–1.8) mg/L for groups 1, 2, and 3, respectively. For $C_{trough,3}$, the corresponding values were 0.33 (0.15–0.52), 0.38 (0.10–0.67), and 0.32 (0.09–0.55) mg/L. No differences were statistically significant. Similarly, no significant differences between groups were found in the precision of prediction either for $C_{max,4}$ or for $C_{trough,3}$. The MAPE (95% CI) of $C_{max,4}$ was 1.5 (1.0–1.9), 1.8 (1.2–2.4), and 2.1 (1.6–2.6) mg/L for groups 1, 2, and 3, respectively. For $C_{trough,3}$, the corresponding MAPE values were 0.45 (0.31–0.59), 0.53 (0.31–0.76), and 0.49 (0.30–0.68) mg/L.

The assayed values of $C_{max,4}$ were within the target range of 6–10 mg/L in 44 of the 84 neonates (52%), and $C_{trough,3}$ was within the range of 0.5–2.0 mg/L in 70 neonates (83%). Neonates from the 3 groups who reached or failed to reach the target range for $C_{max,4}$ and $C_{trough,3}$ are summarized in **Table V**. Failure to achieve the $C_{max,4}$ within the target range was more frequent in premature neonates (combined groups 1 and 2) compared with the full-term neonates of group 3 (55% vs 31%; $P = 0.045$). Fluid retention in the interval from the first infusion (0 hour) to the fourth infusion was positively correlated with the postnatal change in body weight (**Figure 3**) and negatively correlated with $C_{max,4}$ (**Figure 4**). Moreover, in neonates with $C_{max,4} < 6$ mg/L compared with those with a higher $C_{max,4}$, fluid retention in the interval from the

Table IV. Intake of fluid (mean [SD]) in the 2-hour interval before the start of the first infusion with gentamicin and body fluid retention calculated as a difference between fluid intake and loss by urine at various time intervals.

Characteristics	Interval	Group 1: GA <34 Wk	Group 2: GA 34–38 Wk	Group 3: GA >38 Wk
Fluid intake, mL/kg*	From –2 to 0 h	21.1(12.2)	16.1 (8.3)	14.8 (7.8)
Fluid retention, mL/kg†	From –6 to 1 h	46.0 (28.3)	23.3 (17.4)	26.7 (18.5)
Fluid retention, mL/kg‡	From 0 to 24 h	84.0 (59.3)	33.8 (35.6)	49.6 (48.2)
Fluid retention, mL/kg§	From 0 h to 4th infusion	297 (127)	172 (75.8)	179 (85.2)

GA = gestational age.
* $P = 0.06$, group 1 versus 2; $P = 0.1$, group 1 versus 3; $P = 0.3$, group 2 versus 3.
† $P = 0.003$, group 1 versus 2; $P = 0.002$, group 1 versus 3; $P = 0.5$, group 2 versus 3.
‡ $P = 0.001$, group 1 versus 2; $P = 0.01$, group 1 versus 3; $P = 0.2$, group 2 versus 3.
§ $P = 0.002$, group 1 versus 2; $P < 0.005$, group 1 versus 3; $P = 0.8$, group 2 versus 3.

first infusion (0 hour) to the fourth infusion was higher (mean [SD], 243 [124] vs 193 [99.2] mL/kg; $P < 0.05$). The kinetically guided dose adjustment led to the absence of gentamicin accumulation as the $C_{trough,3}$ values were <2 mg/L in all but 1 neonate from group 3.

Clinical Outcome

The infecting microorganisms were identified in 13 of 84 patients (16%) as follows (corresponding MIC values are provided): *Acinetobacter baumannii*, <0.1 mg/L; *Klebsiella oxytoca*, 0.25 mg/L; *S epidermidis*, 0.125 mg/L; *S epidermidis*, 1 mg/L; *S saprophyticus* >16 mg/L; *Escherichia coli*, 0.25 mg/L; *Listeria monocytogenes*, <0.5 mg/L; *Pseudomonas aeruginosa*, 0.25 mg/L (in two cases); *P aeruginosa*, 0.5 mg/L; *Staphylococcus aureus*, 0.125 mg/L; and *Acinetobacter ursingii*, >0.1 mg/L.

Initial positive blood culture before the first gentamicin infusion was obtained in 7 of 13 neonates (54%); neonatal bloodstream infection was identified

Table V. Assayed concentrations of gentamicin in neonates who reached and failed to reach the target range for plasma concentrations detected 1 hour after the start of the fourth infusion ($C_{max,4}$; 6–10 mg/L) and plasma concentrations detected 30 minutes before the fourth infusion ($C_{trough,3}$; 0.5–2 mg/L).

Concentration	Group 1: GA <34 Wk	Group 2: GA 34–38 Wk	Group 3: GA >38 Wk
$C_{max,4}$, mg/L*	6.06 (1.80) [3.6–9.8]	5.97 (1.88) [2.7–8.9]	6.77 (1.83) [3.4–11.1]
$C_{max,4}$ 6–10 mg/L, n/N (%)	11/27 (41)	11/22 (50)	22/35 (63)
$C_{max,4}$ <6 mg/L, n/N (%)	16/27 (59)	11/22 (50)	11/35 (31)
$C_{max,4}$ >10 mg/L, n/N (%)†	0/27 (0)	0/22 (0)	2/35 (6)
$C_{trough,3}$, mg/L*‡	1.09 (0.63) [0.30–3.5]	0.69 (0.36) [0.20–1.7]	0.96 (0.54) [0.30–3.1]
$C_{trough,3}$ 0.5–2 mg/L, n/N (%)	26/27 (96)	16/22 (73)	29/35 (83)
$C_{trough,3}$ <0.5 mg/L, n/N (%)	1/27 (4)	6/22 (27)	5/35 (14)
$C_{trough,3}$ >2 mg/L, n/N (%)	0/27 (0)	0/22 (0)	1/35 (2)

* Mean (SD) [range].
† Conditions of the test have not been met.
‡ $P < 0.005$, group 1 versus 2.

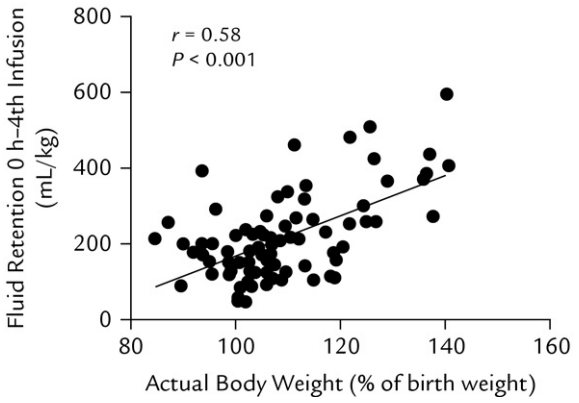

Figure 3. The relationship between the postnatal change in body weight and suspected fluid retention over the interval from the start of the first infusion to the day of the fourth infusion.

in 6 neonates on days 5–13 after the start of antibiotic therapy as a sign of nosocomial superinfection.[31] In the former, gentamicin $C_{max,1}$ did not reach 6 mg/L in 2 of the 7 neonates; in the latter, gentamicin $C_{max,4}$ did not reach 6 mg/L in 5 of 6 neonates. Nevertheless, the C_{max}/MIC ratio >12 was reached in every examined case except for 2 (owing to resistant *S epidermidis* and *S saprophyticus* characterized by a C_{max}/MIC ratio of 0.4). In the first case, gentamicin was substituted with amikacin. The second infection caused by *S saprophyticus* resistant to gentamicin was considered artificial contamination and treated solely with vancomycin. Neonatal-proven sepsis was cured in all 13 patients.

Clinical signs and laboratory data indicative of sepsis disappeared in 5–10 days as shown in 68 of 71 neonates (96%). Consequently, empirical antimicrobial therapy was withdrawn. In 1 case, gentamicin was withdrawn because of the high $C_{trough,3}$ value. In the remaining 3 neonates, CRP decreased to a normal value (after >10 days) without any change in antimicrobial therapy.

No patient required cessation of treatment owing to acute toxicity of gentamicin. Two neonates died. The first died of HIE as a consequence of perinatal asphyxia, documented by EEG findings. Gentamicin $C_{max,4}$ and $C_{trough,3}$ were assayed 13 days before death ($C_{max,4}$, 4.5 mg/L; $C_{trough,3}$, 1.9 mg/L). The second neonate died of stage IV intraventricular hemorrhage. In this case, gentamicin plasma concentrations were detected 9 days before death ($C_{max,4}$, 4.5 mg/L; $C_{trough,3}$, 0.7 mg/L). Neither case revealed signs of renal disorder attributable to gentamicin.

Renal dysfunction was detected in 23 neonates as follows: in 8 preterm neonates in group 1, 2 preterm neonates in group 2, and 13 full-term neonates in group 3. Eight of the 13 neonates from group 3 were diagnosed with intrauterine growth restriction. In 13 of the 23 neonates with renal dysfunction, serum creatinine reached a normal value before gentamicin withdrawal. In 7 of the 23 neonates, serum creatinine reached a normal value after gentamicin had been withdrawn (maximum of 1 week later). There was a relationship to gentamicin plasma concentration in 1 case only ($C_{max,1}$, 7.5 mg/L; $C_{trough,1}$, 2.6 mg/L; $C_{max,4}$, 11.1 mg/L; and $C_{trough,3}$, 3.1 mg/L). This neonate should have been treated with a reduced maintenance dose, as recommended by the clinical pharmacologist. This advice was not accepted by the physician in charge although the $C_{trough,1}$ value previously detected had been too high to be considered normal. $C_{trough,3}$ increased greatly (the upper limit of C_{trough} is <2 mg/L). Gentamicin was then immediately withdrawn, and serum creatinine concentrations returned to the normal range within 2 days. The additive contribution of multiorgan dysfunction syndrome cannot be excluded. In this case, no abnormality of hearing was found.

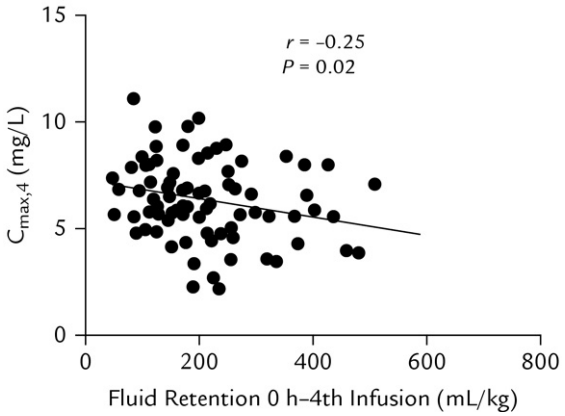

Figure 4. The relationship between the plasma concentration detected 1 hour after the start of the fourth infusion ($C_{max,4}$) and suspected body fluid retention calculated from the start of the first infusion (0 hour) to the day of the fourth infusion.

Results of sonography examination found that 68 of 82 neonates (83%) were examined during the first year, and 46 of these neonates (56%) were reexamined during the second year of life (24-month follow-up). Data were missing because some families failed to return for follow-up visits.

There were some signs of nephrocalcinosis on ultrasonography. Nonspecific abnormalities were described in 3 neonates (4%) during the first year. No signs of nephrocalcinosis were found by reexamination during the second year.

Transient evoked otoacoustic emission recordings were used to evaluate hearing abnormalities in 68 of 82 neonates (83%) during the first year (recording 1) and 46 of 82 patients (56%) during the second year (recording 2). Again, there was incomplete data for recordings 1 and 2 because some families failed to return for follow-up visits. Based on these recordings, 2 neonates were diagnosed with HIE grade III–IV (a consequence of severe perinatal asphyxia[32]). Moreover, these 2 patients were found to have unilateral hearing loss attributable to high severity of HIE.

DISCUSSION

It is generally accepted that in neonates critically ill during the first week of life, the interindividual variability of gentamicin pharmacokinetics is large. In the present study, after the first standard dose of 4 mg/kg, the $C_{max,1}$ concentration observed was variable, ranging from 4.4–16.0 mg/L (%CV, 29%). The estimates of V_{d1} and CL_1 obtained were in good agreement with data from the literature.[33] GA as a maturational covariate strongly— and in the expected manner—influenced both Vd_1 and CL_1,[34–36] whereas fluid intake did not. Fluid retention showed modest negative correlations with CL_1. Despite a higher mean value of the body weight–normalized Vd_1 and correspondingly lower mean $C_{max,1}$ in premature neonates, the failure rate to reach the threshold level of 6 mg/L was relatively low (13%) and comparable in all 3 groups of neonates.

In contrast, a large body fluid retention in the interval from before the first infusion to the fourth infusion, which was associated with a marked gain in body weight, resulted in $C_{max,4}$ values below the target range. It can be ascribed to lower intravascular retention,[37] but the interstitial volume was much larger.[38] Fluid easily leaves the plasma, enters the interstitium, and may accumulate to reach the higher volume detectable by a weight gain. In critically ill neonates suffering from edema, anasarca, or ascites, this process has been well described using input-output fluid volume balance. Analysis of $C_{max,4}$ assayed revealed levels <6 mg/L in most neonates from group 1 with the lowest GA, who at the same time had the highest fluid retention per kilogram of body weight. Considerable systemic capillary leak (sequelae of bacteriemia and sepsis[39]) that increases extracellular fluid volume might enhance escape of gentamicin from intravascular fluid extravascularly and contribute to this finding. Moreover, some amount of the drug is thought to be distributed, for instance, to peritoneal fluid, reaching even therapeutic concentrations.[40] Due to extended interdose intervals, C_{trough} concentrations after the first and third doses were within the target range and documented the absence of gentamicin accumulation. This outcome might also be attributed to successful therapy resulting in an increase in glomerular filtration rate and gentamicin clearance within a few days.[14]

The individual range of bactericidal plasma concentrations should be taken into consideration when determining MIC and consequently the C_{max}/MIC ratio. With higher sensitivity of microorganisms in vitro, the plasma concentration of gentamicin necessary for bactericidal activity diminishes. In this study, MIC was generally very low and the C_{max}/MIC ratio was estimated to be >12 for both $C_{max,1}$ and $C_{max,4}$ values, except for 2 resistant staphylococcal infections, which necessitated substituting gentamicin for amikacin and vancomycin.

In the present study, creatinine CL was neither calculated nor measured because several studies, including a recent population analysis, reported that this value is an unreliable indicator of the glomerular filtration rate in neonates during the first week of life and the correlation between calculated creatinine CL and gentamicin CL is weak or absent.[41–44] It is understood that when renal function is changing rapidly, estimation of the CL of aminoglycoside antibiotics may be a more accurate indicator of glomerular filtration rate than serum creatinine.[45]

The infection in all 13 neonates with proven sepsis was cured, and signs of sepsis abated within several days. In accordance with any study dealing with neonatal sepsis, it was difficult to assess antibiotic efficacy and prevent treatment failure because of the relatively rare incidence of culture-proven cases.[46,47]

This study also focused on renal disorders and abnormalities of hearing monitored during the 24-month

follow-up period. Nephrotoxicity is a well-known transient consequence of therapy with all aminoglycosides.[48,49] Neonates are thought to be at higher risk for aminoglycoside toxicity, but some published data are conflicting.[50,51] Toxicity appears to be related to both C_{trough} values and duration of therapy. In the present study, $C_{trough,3}$ values exceeded the target value of 2 mg/L only in 1 full-term neonate. In contrast, a retrospective analysis[52] performed at the same institution 4 years earlier evaluated therapy with gentamicin without a kinetically guided approach over the years 2001 to 2004. That analysis found a $C_{trough,4}$ value >2 mg/L in 37% of neonates.

Renal dysfunction in 23 neonates was associated with multiorgan dysfunction syndrome after perinatal asphyxia or therapeutic modest whole-body hypothermia, sepsis, prematurity, and intrauterine growth restriction in accordance with the literature.[53] Only 1 neonate experienced renal dysfunction that appeared related to gentamicin administration; serum creatinine concentrations decreased rapidly after gentamicin withdrawal, and gentamicin plasma concentrations were considered within recommended limits.

Hearing disorders related to therapy with aminoglycosides have been attributed to their distribution to the statoacoustic apparatus with subsequent apoptotic death of hair cells and vestibular destruction.[54–56] In neonates born at a GA <34 weeks or birth weight <1000 g, the prevalence of hearing loss in a representative NICU population was 3.2%.[57] Independent risk factors for hearing loss were severe birth asphyxia and assisted ventilation lasting >5 days. Recently, a second mutation involving a thymidine deletion in the ribosomal RNA gene has been identified that can predispose patients to aminoglycoside auditory toxicity.[58] The present study found 2 cases of unilateral hearing loss that appeared to be related to documented severe HIE.

Limitations

The present investigation involved only a small number of patients at a single center and used only data routinely registered and available in the NICU in charge. Therefore, only the volume of urine was taken into account as the sole route of fluid loss, which caused some overestimation of the input-output difference. Additional investigations should focus on particular conditions, such as intrauterine growth restriction and perinatal asphyxia, that may particularly influence the pharmacokinetics of gentamicin in neonates.

CONCLUSIONS

The initial dose of gentamicin 4 mg/kg for these critically ill premature and mature neonates with sepsis during the first week was high enough to reach bactericidal $C_{max,1}$ within 6 to 10 mg/L. $C_{max,1}$ values <6 mg/L were found in 13% of neonates. The interdose interval modified according to the recommendation resulted in a C_{trough} value within the target range of 0.5–2.0 mg/L in all but 2 neonates. The kinetically guided maintenance dosing of gentamicin based on plasma concentrations after the first dose should be optimized, taking into account actual body weight.

ACKNOWLEDGMENTS

This study was supported by a grant from the Ministry of Health (IGA MZ 1A 8671-5/05) and the Research Project from the Czech Ministry of Education, Youth and Sports (MSM 0021620820).

The authors thank the staff of the Neonatal Intensive Care Unit, Department of Pediatrics and Adolescent Medicine, General Teaching Hospital, Prague, Czech Republic. They also thank the hospital's Laboratory of Clinical Chemistry for gentamicin detection, the Laboratory of Microbiology for MIC determination, and the Otoacoustic Laboratory for examination of hearing functions.

Dr. Martínková was responsible for the literature search, clinical pharmacologic service, and study design. Dr. Pokorná was responsible for data collection. Drs. Záhora and Chládek contributed to the statistical analysis and figures. Dr. Vobruba was responsible for the services of the NICU in charge. Drs. Selke-Krulichová and Chládková contributed to data interpretation and data analysis.

The authors have indicated that they have no conflicts of interest regarding the content of this article.

REFERENCES

1. Barriere SL. Bacterial resistance to beta-lactams, and its prevention with combination antimicrobial therapy. *Pharmacotherapy*. 1992;12:397–402.
2. García B, Barcia E, Pérez F, Molina IT. Population pharmacokinetics of gentamicin in premature newborns. *J Antimicrob Chemother*. 2006;58:372–379.
3. Moore RD, Smith CR, Lietman PS. The association of aminoglycoside plasma levels with mortality in patients with gram-negative bacteremia. *J Infect Dis*. 1984;149:443–448.

4. Moore RD, Smith CR, Lipsky JJ, et al. Risk factors for nephrotoxicity in patients treated with aminoglycosides. *Ann Intern Med*. 1984;100:352–357.
5. McCormack JP, Jewesson PJ. A critical reevaluation of the "therapeutic range" of aminoglycosides. *Clin Infect Dis*. 1992;14:320–339.
6. Hansen M, Christrup LL, Jarløv JO, et al. Gentamicin dosing in critically ill patients. *Acta Anaesthesiol Scand*. 2001;45:734–740.
7. Nicolau DP, Freeman CD, Belliveau PP, et al. Experience with a once-daily aminoglycoside program administered to 2,184 adult patients. *Antimicrob Agents Chemother*. 1995; 39:650–655.
8. Avent ML, Kinney JS, Istre GR, Whitfield JM. Gentamicin and tobramycin in neonates: Comparison of a new extended dosing interval regimen with a traditional multiple daily dosing regimen. *Am J Perinatol*. 2002; 19:413–420.
9. De Paepe P, Belpaire FM, Buylaert WA. Pharmacokinetic and pharmacodynamic considerations when treating patients with sepsis and septic shock. *Clin Pharmacokinet*. 2002;41:1135–1151.
10. Marik PE. Aminoglycoside volume of distribution and illness severity in critically ill septic patients. *Anaesth Intensive Care*. 1993;21:172–173.
11. Buijk SE, Mouton JW, Gyssens IC, et al. Experience with a once-daily dosing program of aminoglycosides in critically ill patients. *Intensive Care Med*. 2002;28:936–942.
12. Power BM, Forbes AM, van Heerden PV, Ilett KF. Pharmacokinetics of drugs used in critically ill adults. *Clin Pharmacokinet*. 1998;34:25–56.
13. van Dalen R, Vree TB. Pharmacokinetics of antibiotics in critically ill patients. *Intensive Care Med*. 1990; 16(Suppl 3):S235–S238.
14. Triginer C, Izquierdo I, Fernández R, et al. Gentamicin volume of distribution in critically ill septic patients. *Intensive Care Med*. 1990;16:303–306.
15. Reimann IR, Meier-Hellmann A, Reinhart K, Hoffmann A. Comments to consensus document. Once-daily dosing of aminoglycosides from N. Anaizi. A supplement to dosage and monitoring in critically ill patients. *Int J Clin Pharmacol Ther*. 1997;35: 397.
16. Niemiec PW Jr, Allo MD, Miller CF. Effect of altered volume of distribution on aminoglycoside levels in patients in surgical intensive care. *Arch Surg*. 1987;122:207–212.
17. Agras PI, Tarcan A, Baskin E, et al. Acute renal failure in the neonatal period. *Ren Fail*. 2004;26:305–309.
18. Mathur NB, Agarwal HS, Maria A. Acute renal failure in neonatal sepsis. *Indian J Pediatr*. 2006;73:499–502.
19. Craig WA. Once-daily versus multiple-daily dosing of aminoglycosides. *J Chemother*. 1995;7(Suppl 2): 47–52.
20. Stoll BJ, Hansen NI, Adams-Chapman I, et al; for National Institute of Child Health and Human Development Neonatal Research Network. Neurodevelopmental and growth impairment among extremely low-birth-weight infants with neonatal infection. *JAMA*. 2004;292:2357–2365.
21. Gross I, Vallard PL. Hormonal therapy for prevention of respiratory distress syndrome. In: Polin RA, Fox WW, Abman SH, eds. *Fetal and Neonatal Physiology*. 3rd ed. Philadelphia, Pa: Saunders; 2004:1069–1073.
22. Xanthou M, Fotopoulos S, Mouchtouri A, et al. Inflammatory mediators in perinatal asphyxia and infection. *Acta Paediatr Suppl*. 2002;91:92–97.
23. Isemann BT, Kotagal UR, Mashni SM, et al. Optimal gentamicin therapy in preterm neonates includes loading doses and early monitoring. *Ther Drug Monit*. 1996;18: 549–555.
24. Young TE, Mangum OB. *Neofax: A Manual of Drugs Used in Neonatal Care*. 15th ed. Raleigh, NC: Acorn Publishing; 2002:1–65.
25. Wagner JG. *Pharmacokinetics for the Pharmaceutical Scientist*. Lancaster, Pa: Technomic Publishing; 1993.
26. Sheiner LB, Beal SL. Some suggestions for measuring predictive performance. *J Pharmacokinet Biopharm*. 1981;9:503–512.
27. Ganong WF. Dynamics of blood and lymph flow. In: *Review of Medical Physiology*. 21st ed. New York, NY: McGraw-Hill Professional, Lange Medical Books; 1993:579–598.
28. Goldstein B, Giroir B, Randolph A; for International Consensus Conference on Pediatric Sepsis. International pediatric sepsis consensus conference: Definitions for sepsis and organ dysfunction in pediatrics. *Pediatr Crit Care Med*. 2005;6:2–8.
29. Narendra A, White MP, Rolton HA, et al. Nephrocalcinosis in preterm babies. *Arch Dis Child Fetal Neonatal Ed*. 2001;85:F207–F213.
30. Sweileh WM. A prospective comparative study of gentamicin- and amikacin-induced nephrotoxicity in patients with normal baseline renal function. *Fundam Clin Pharmacol*. 2009;23:515–520.
31. Modi N, Doré CJ, Saraswatula A, et al. A case definition for national and international neonatal bloodstream infection surveillance. *Arch Dis Child Fetal Neonatal Ed*. 2009;94:F8–F12.
32. Sarnat HB, Sarnat MS. Neonatal encephalopathy following fetal distress. A clinical and electroencephalographic study. *Arch Neurol*. 1976; 33:696–705.
33. Yaffe SJ. Antimicrobial therapy and the neonate. *Obstet Gynecol*. 1981; 58(Suppl 5):85S–94S.
34. Koren G, James A, Perlman M. A simple method for the estimation of glomerular filtration rate by gentamicin pharmacokinetics during routine drug monitoring in the newborn. *Clin Pharmacol Ther*. 1985;38: 680–685.
35. Alcorn J, McNamara PJ. Ontogeny of hepatic and renal systemic clear-

ance pathways in infants: Part I. *Clin Pharmacokinet*. 2002;41:959–998.
36. Semchuk W, Borgmann J, Bowman L. Determination of a gentamicin loading dose in neonates and infants. *Ther Drug Monit*. 1993;15:47–51.
37. Brace RA. Fluid distribution in the fetus and neonate. In: Polin RA, Fox WW, Abman SH, eds. *Fetal and Neonatal Physiology*. 3rd ed. Philadelphia, Pa: Saunders; 2004:1341–1350.
38. Matsuda N, Hattori Y. Vascular biology in sepsis: Pathophysiological and therapeutic significance of vascular dysfunction. *J Smooth Muscle Res*. 2007; 43:117–137.
39. Hotz HG, Foitzik T, Rohweder J, et al. Intestinal microcirculation and gut permeability in acute pancreatitis: Early changes and therapeutic implications. *J Gastrointest Surg*. 1998;2:518–525.
40. Serour F, Dan M, Gorea A, et al. Penetration of aminoglycosides into human peritoneal tissue. *Chemotherapy*. 1990;36:251–253.
41. Stonestreet BS, Bell EF, Oh W. Validity of endogenous creatinine clearance in low birthweight infants. *Pediatr Res*. 1979;13:1012–1014.
42. Saarela T, Lanning P, Koivisto M. Prematurity-associated nephrocalcinosis and kidney function in early childhood. *Pediatr Nephrol*. 1999;13: 886–890.
43. Gordjani N, Burghard R, Leititis JU, Brandis M. Serum creatinine and creatinine clearance in healthy neonates and prematures during the first 10 days of life. *Eur J Pediatr*. 1988;148:143–145.
44. Nielsen EI, Sandström M, Honoré PH, et al. Developmental pharmacokinetics of gentamicin in preterm and term neonates: Population modelling of a prospective study. *Clin Pharmacokinet*. 2009;48:253–263.
45. Holford NH. Pharmacokinetics and pharmacodynamics: Rational dosing and the time course of drug action. In: Katzung BG, Masters SB, Trevor AJ, eds: *Basic and Clinical Pharmacology*. 11th ed. New York, NY: McGraw-Hill Medical; 2009:37–
46. Spitzer AR, Kirkby S, Kornhauser M. Practice variation in suspected neonatal sepsis: A costly problem in neonatal intensive care. *J Perinatol*. 2005;25:265–269.
47. Kumar Y, Qunibi M, Neal TJ, Yoxall CW. Time to positivity of neonatal blood cultures. *Arch Dis Child Fetal Neonatal Ed*. 2001;85:F182–F186.
48. Dillon JJ. Nephrotoxicity from antibacterial, antifungal and antiviral drugs. In: Molitoris BA, Finn WF, eds. *Acute Renal Failure: A Companion to Brenner and Rector's the Kidney, 6th Edition*. Philadelpia, Pa: Saunders; 2001:349–364.
49. Martínez-Salgado C, López-Hernández FJ, López-Novoa JM. Glomerular nephrotoxicity of aminoglycosides. *Toxicol Appl Pharmacol*. 2007;223:86–98.
50. Patzer L. Nephrotoxicity as a cause of acute kidney injury in children. *Pediatr Nephrol*. 2008;23:2159–2173.
51. Chesney RW, Jones DP. Nephrotoxins. In: Avner ED, Harmon WE, Niaudet P, eds. *Pediatric Nephrology*. 5th ed. Philadelphia, Pa: Lippincott Williams & Wilkins; 2004:987–1004.
52. Pokorná P, Martinková J, Záhora J, et al. Therapeutic drug monitoring of gentamicin in neonates critically ill at the 1st week of life. Presented at the IXth World Conference on Clinical Pharmacology and Therapeutics, July 27–August 1, 2008. Quebec, Ontario, Canada. Poster T4W167.
53. Allegaert K, Anderson BJ, van den Anker JN, et al. Renal drug clearance in preterm neonates: Relation to prenatal growth. *Ther Drug Monit*. 2007;29:284–291.
54. Bareggi R, Narducci P, Grill V, et al. Localization of an aminoglycoside (streptomycin) in the inner ear after its systemic administration. A histochemical study using fluorescence microscopy. *Histochemistry*. 1986;84: 237–240.
55. Lii M, Ding D, Zheng XY, Salvi R. Vestibular destruction by slow infusion of gentamicin into semicircular canals. *Acta Otolaryngol Suppl*. 2004; 552:35–41.
56. Forge A, Li L. Apoptotic death of hair cells in mammalian vestibular sensory epithelia. *Hear Res*. 2000; 139:97–115.
57. Hille ET, van Straaten HI, Verkerk PH; for Dutch NICU National Hearing Screening Working Group. Prevalence and independent risk factors for hearing loss in NICU infants. *Acta Paediatr*. 2007;96:1155–1158.
58. Bates DE. Aminoglycoside ototoxicity. *Drugs Today (Barc)*. 2003;39:277–285.

Address correspondence to: Jirina Martínková, MD, PhD, Department of Pharmacology, Charles University in Prague, Faculty of Medicine in Hradec Králové, Simkova 870, 500 38 Hradec Králové, Czech Republic. E-mail: martinkova@lfhk.cuni.cz