PHARMACOKINETIC STUDY AFTER SINGLE ORAL DOSE OF BETA-LACTAM AND FLUOROQUINOLONE ANTIMICROBIALS IN BANGLADESHI HEALTHY MALE VOLUNTEERS

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ABSTRACT

The variability of response to antimicrobial has been inadequately explained because of lack of pharmacokinetic data. The present pharmacokinetic study was designed to provide information beneficial to formulate a population pharmacokinetic model appropriate for Bangladeshi population. Among the beta-lactams, amoxicillin (500 mg), flucloxacillin (250 mg), cefuroxime (500 mg) and among the fluoroquinolones, ciprofloxacin (500 mg), levofloxacin (500 mg), gatifloxacin (400 mg) were studied in 15 healthy Bangladeshi male volunteers. The C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ *and* C_{trough} *were studied with all antimicrobials. Later, the* C_{max} *and* $AUC_{0-infinity}$ *were adjusted for bodyweight and dose. Among the studied antimicrobials, the* C_{max} *varied to great extent even after adjustment for bodyweight and dose. The* C_{max} *after adjustment was highest in case of flucloxacillin and lowest in case of ciprofloxacin, indicating excellent absorption of flucloxacillin in Bangladeshi population. The information obtained through this study generates necessity of new cut-off value for the antimicrobials.*

Key words*: Pharmacokinetics, Beta-lactam, Fluoroquinolones, Bangladeshi volunteer*

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INTRODUCTION

Least developed countries like Bangladesh differs greatly from the developed countries especially in disease pattern. Bacterial infections are the principal reason of morbidity and mortality in these countries. Consequently, antimicrobials became the group of medicine that contributing mostly in medicine consumption of country like Bangladesh.[1-7] Though antimicrobial susceptibility surveillance data is required to support the selection of appropriate antimicrobial agents, there availability is generally poor.[8]

Plasma concentration of medicine is the most important factor in clinical response to pharmacotherapy and therefore, even when any medicine administered at one particular dose, some patients suffer from toxicity and some other fail to exhibit desired therapeutic effect.[9-12] There are several factors that influence the pharmacokinetic variability of medicines, which include ethnicity, gender, age, body weight, disease status, pregnancy, hepatic impairment, drug-drug interactions, binding to plasma proteins and host genetic factors.[11,13-14]

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The variability of response to antimicrobial is now only partially explained by the available sensitivity data, though detailed understanding about their pharmacokinetic parameters is required to explain such variability. Studies conducted in different countries provide information about the pharmacokinetic behavior of that studied population like Cambodia[15], China[16], Japan[17], Denmark[18], Switzerland[19], Uganda[20] and Jordan[21]. The great differences observed in findings of different studies indicate necessity of obtaining pharmacokinetic data from the population for whom the dose of a medicine needs to be optimized. The extrapolation of data obtained from other sources is not possible because of high inter-individual variation as well as difference in ethnic, racial, social and cultural origin. Bangladeshi populations are ethnically, socially and genetically different from the studied population of different studies conducted so far.

In this backdrop, the pharmacokinetic behavior of Bangladeshi population warrants immediate detail evaluation in order to get the understanding adequate to generate new ideas. There have been efforts to correlate plasma concentration with clinical outcome, though very little progress has been made so far.

Though some initiative and effort was made towards understanding the pharmacokinetics of antimicrobials in Bangladeshi children[22] and adult population[23-24], very little development has been achieved. There were different challenges like selection of volunteers, laboratory facilities, establishment of laboratory methods and hospitalization of volunteers, for which this type of pharmacokinetic study could not be conducted before in this region. Inadequate information about the pharmacokinetic status of Bangladeshi people led to the necessity of research to enable scientists to understand these issues. The present pharmacokinetic study was designed to provide information beneficial to formulate a population pharmacokinetic model appropriate for Bangladeshi population.

Study period

The study was conducted from January 2010 to December 2010.

Place of study

The study was conducted in the Department of Clinical Pharmacy and Pharmacology, University of Dhaka, Department of Biochemistry, Bangabandhu Sheikh Mujib Medical University (BSMMU) and ZH Sikder Women's Medical College & Hospital, Dhaka.

Subject of the study

This study included 15 (fifteen) Bangladeshi Bangalee male healthy volunteers, aged 20–30 years (24.80 ± 2.43 years), weighing between 51–60 kg (55.93 ± 3.33 kg), height between 160.00 - 172.00 cm (166.00 ± 4.00 cm), Body Mass Index (BMI) between 19.10 - 22.31 (20.39 ± 0.94), serum creatinine between 0.60 - 0.80 mg/dL (0.73 ± 0.07 mg/dL), serum ALT between 30.00 - 45.00 U/L (35.87 ± 4.73 U/L) and prothrombin time between 9.00 - 12.00 sec (10.07 ± 0.88 second). Volunteers were screened to ensure that they have no cardiac, renal, hepatic, hematological, neurological, gastrointestinal and pulmonary disorders and allergy to ciprofloxacin. The volunteers were requested to stay away from any medication for two weeks prior to the study and up to its completion. Moreover, they were requested not to take any beverages like alcohol, coffee and tea in 48 h prior to first dose and until the collection of last blood sample.[25]

Medicines (beta-lactam)

Among the commonly used beta-lactams of Bangladesh, following were included in the study:

1. Commercially available pioneer brand of amoxicillin 500 mg capsules
2. Commercially available pioneer brand of flucloxacillin 250 mg capsules
3. Commercially available pioneer brand of cefuroxime 500 mg tablets

Medicines (fluoroquinolones)

Among the commonly used fluoroquinolones of Bangladesh, following were included in the study:

1. Commercially available pioneer brand of ciprofloxacin 500 mg tablets
2. Commercially available pioneer brand of levofloxacin 500 mg tablets
3. Commercially available pioneer brand of gatifloxacin 400 mg tablets

Medicine administration schedule

This study was conducted in an open label design with one-week washout period between oral doses of different medicines. At the time of study, the healthy volunteers were hospitalized at Z H Sikder Women's Medical College & Hospital, Dhaka, Bangladesh for collection of blood samples. The volunteers were observed by experienced physician to detect adverse effects (if any) during the study.

Amoxicillin: All volunteers received single oral dose of 500 mg amoxicillin capsule.

Flucloxacillin: All volunteers received single oral dose of 250 mg flucloxacillin capsule.

Cefuroxime: All volunteers received single oral dose of 500 mg cefuroxime tablet.

Ciprofloxacin: All volunteers received single oral dose of 500 mg ciprofloxacin tablet.

Levofloxacin: All volunteers received single oral dose of 500 mg levofloxacin tablet.

Gatifloxacin: All volunteers received single oral dose of 400 mg gatifloxacin tablet.

Blood sample collection

For collection of blood samples, a catheter *in situ* was placed in one arm by the researcher with all aseptic precaution. The catheter was removed immediately if phlebitis develops, otherwise normally after the study period. Blood samples (3 ml at each occasion) were obtained at appropriate intervals as follows:

Amoxicillin
Zero (immediately before the first dose), 30, 60, 120, 180, 360, 480, 960 and 1440 minutes after first dose of medicine

Flucloxacillin
Zero (immediately before the first dose), 30, 60, 120, 180, 360, 720, 1080 and 1440 minutes after first dose of medicine

Cefuroxime
Zero (immediately before the first dose), 30, 60, 120, 180, 360, 480, 720 and 1440 minutes after first dose of medicine

Ciprofloxacin
Zero (immediately before the first dose), 30, 60, 120, 180, 360, 480, 720 and 1440 minutes after first dose of medicine

Levofloxacin
Zero (immediately before the first dose), 30, 60, 120, 180, 360, 480, 720 and 1440 minutes after first dose of medicine

Gatifloxacin
Zero (immediately before the first dose), 30, 60, 120, 180, 360, 480, 720 and 1440 minutes after first dose of medicine

After collecting, blood samples were placed in sterile tubes with 100 µL of 10% EDTA solution. Immediately after each blood collection, the samples were centrifuged at 3000 × g for 15 min and plasma was then separated and stored at −70°C in the Department of Biochemistry, Bangabandhu Sheikh Mujib Medical University. The stored samples were then studied by HPLC in appropriate method for amoxicillin[23,26-27], flucloxacillin[28], cefuroxime[29], ciprofloxacin[24,30-32], levofloxacin[33] and gatifloxacin[34] in the Department of Clinical Pharmacy and Pharmacology, University of Dhaka and other recognized laboratory.

Sample analysis

Plasma concentrations were measured by high-performance liquid chromatography (HPLC). Specificity, linearity, lower limit of quantification (LLOQ), inter-day and intra-day precision and accuracy as well as absolute recovery and stability was evaluated.

Ethical clearance

The study was approved by the National Research Ethics Committee of Bangladesh Medical Research Council (BMRC/NREC/2007-2010/1709). Informed written consent was obtained from all participants after explaining the nature, risk and benefits of the study to them. Throughout the study, according to the obligations of the Helsinki declaration, rights and dignity of the subjects were protected.[35]

RESULTS

All volunteers completed the study without any event, which was ascertained by thorough medical examination by experienced physician after study completion.

In addition to the actual plasma concentration and the calculated different pharmacokinetic parameters based on those obtained values, an adjustment was done to minimize the variation in weight of the volunteers and dose of the administered medicines. For that purpose, the obtained actual plasma concentration was adjusted for bodyweight and expressed as per mg of antimicrobial/kg of body weight.

= Concentration of antimicrobial in plasma (µg/mL) X [(amount of antimicrobial administered (mg)/bodyweight of volunteer (kg)]

The values were then used for calculation of different pharmacokinetic parameters. Therefore, the $AUC_{0-infinity}$ and C_{max} were calculated with two values, one with the original plasma concentration of antimicrobials detected in HPLC and the other was with value obtained after adjustment for dose and bodyweight.

Table 1 shows that after single oral administration of amoxicillin (500 mg), the C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ and C_{trough} were 6.57 ± 0.53 µg/mL, 1265.20 ± 203.07 µg min/m, 72.00 ± 24.84 min, 89.95 ± 17.22 min and 0.29 ± 0.20 µg/mL respectively. When the values were adjusted for bodyweight and dose, the C_{max} and $AUC_{0-infinity}$, 0.74 ± 0.04 µg/mL per mg/kg and 141.31 ± 14.22 µg min/mL per mg/kg respectively.

In case of flucloxacillin (250 mg), the C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ and C_{trough} were 4.64 ± 0.62 µg/mL, 1005.88 ± 171.26 µg min/m, 76.00 ± 27.46 min, 81.59 ± 15.91 min and 0.46 ± 0.30 µg/mL respectively. When the values were adjusted for bodyweight and dose, the C_{max} and $AUC_{0-infinity}$, 1.03 ± 0.12 µg/mL per mg/kg and 223.05 ± 25.99 µg min/mL per mg/kg respectively.

In case of cefuroxime (500 mg), the C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ and C_{trough} were 3.70 ± 0.52 µg/mL, 806.68 ± 122.08 µg min/m, 104.00 ± 27.46 min, 88.34 ± 11.06 min and 0.64 ± 0.32 µg/mL respectively. When the values were adjusted for bodyweight and dose, the C_{max} and $AUC_{0-infinity}$, 0.83 ± 0.07 µg/mL per mg/kg and 180.26 ± 16.44 µg min/mL per mg/kg respectively.

Table 1

Pharmacokinetic parameters following single oral administration of beta-lactam (amoxicillin 500 mg, flucloxacillin 250 mg, cefuroxime 500 mg) antimicrobials in Bangladeshi healthy male volunteers

Pharmacokinetic parameters	Cap. Amoxicillin (500 mg) Mean ± SD	Cap. Flucloxacillin (250 mg) Mean ± SD	Tab. Cefuroxime (500 mg) Mean ± SD
C_{max} (µg/mL)	6.57 ± 0.53 (5.90 - 7.90)	4.64 ± 0.62 (3.21 - 5.35)	3.70 ± 0.52 (3.01 - 4.96)
$AUC_{0-infinity}$ (µg min/mL)	1265.20 ± 203.07 (1011.03 -1687.22)	1005.88 ± 171.26 (780.03 -1247.88)	806.68 ± 122.08 (656.27 - 1002.93)
T_{max} (min)	72.00 ± 24.84 (60.00 - 120.00)	76.00 ± 27.46 (60.00 - 120.00)	104.00 ± 27.46 (60.00 - 120.00)
$T_{1/2}$ (min)	89.95 ± 17.22 (46.31 - 113.17)	81.59 ± 15.91 (54.24 - 101.54)	88.34 ± 11.06 (74.97 - 110.33)
C_{trough} (µg/mL)	0.29 ± 0.20 (0.00 - 0.52)	0.46 ± 0.30 (0.00 - 0.93)	0.64 ± 0.32 (0.28 - 1.19)
C_{max} (µg/mL per mg/kg) *adjusted value*	0.74 ± 0.04 (0.67 - 0.81)	1.03 ± 0.12 (0.77 - 1.18)	0.41 ± 0.03 (0.36 - 0.51)
$AUC_{0-infinity}$ (µg min/mL per mg/kg) *adjusted value*	141.31 ± 14.22 (121.32 - 172.10)	223.05 ± 25.99 (187.21 - 254.57)	90.13 ± 8.22 (78.75 - 102.30)

Adjusted value means the value obtained after adjustment of the original values expressed as mg of antimicrobial per kg bodyweight

Table 2 shows that after single oral administration of ciprofloxacin (500 mg), the C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ and C_{trough} were 2.11 ± 0.48 µg/mL, 585.92 ± 93.64 µg min/m, 72.00 ± 24.84 min, 206.91 ± 16.91 min and 0.07 ± 0.07 µg/mL respectively. When the values were adjusted for bodyweight and dose, the C_{max} and $AUC_{0-infinity}$, 0.23 ± 0.04 µg/mL per mg/kg and 65.00 ± 6.95 µg min/mL per mg/kg respectively.

In case of levofloxacin (500 mg), the C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ and C_{trough} were 4.57 ± 0.65 µg/mL, 2326.69 ± 379.79 µg min/m, 76.00 ± 27.46 min, 377.04 ± 30.31 min and 0.53 ± 0.11 µg/mL respectively. When the values were adjusted for bodyweight and dose, the C_{max} and $AUC_{0-infinity}$, 0.53 ± 0.04 µg/mL per mg/kg and 269.42 ± 25.47 µg min/mL per mg/kg respectively.

In case of gatifloxacin (400 mg), the C_{max}, $AUC_{0-infinity}$, T_{max}, $T_{1/2}$ and C_{trough} were 3.45 ± 0.34 µg/mL, 1779.60 ± 247.00 µg min/m, 80.00 ± 29.28 min, 420.81 ± 32.79 min and 0.34 ± 0.11 µg/mL respectively. When the values were adjusted for bodyweight and dose, the C_{max} and $AUC_{0-infinity}$, 0.48 ± 0.02 µg/mL per mg/kg and 247.00 ± 19.91 µg min/mL per mg/kg respectively.

Table 2

Pharmacokinetic parameters following single oral administration of fluoroquinolones (ciprofloxacin 500mg, levofloxacin 500 mg and gatifloxacin 400 mg) in Bangladeshi healthy male volunteers

Pharmacokinetic parameters	Tab. Ciprofloxacin (500 mg) Mean ± SD	Tab. Levofloxacin (500 mg) Mean ± SD	Tab. Gatifloxacin (400 mg) Mean ± SD
C_{max} (µg/mL)	2.11 ± 0.48 (1.60 - 3.59)	4.57 ± 0.65 (3.13 - 5.60)	3.45 ± 0.34 (3.01 - 4.02)
$AUC_{0-infinity}$ (µg min/mL)	65.00 ± 6.95 (49.24 - 75.32)	2326.69 ± 379.79 (1610.58 - 2977.25)	1779.60 ± 247.00 (1490.40 - 2189.46)
T_{max} (min)	72.00 ± 24.84 (60.00 - 120.00)	76.00 ± 27.46 (60.00 - 120.00)	80.00 ± 29.28 (60.00 - 120.00)
$T_{1/2}$ (min)	206.91 ± 16.91 (165.39 - 233.23)	377.04 ± 30.31 (311.96 - 421.29)	420.81 ± 32.79 (357.84 - 458.10)
C_{trough} (µg/mL)	0.07 ± 0.07 (0.00 - 0.22)	0.53 ± 0.11 (0.36 - 0.74)	0.34 ± 0.11 (0.20 - 0.54)
C_{max} (µg/mL per mg/kg) *adjusted value*	0.23 ± 0.04 (0.19 - 0.37)	0.53 ± 0.04 (0.45 - 0.59)	0.48 ± 0.02 (0.43 - 0.51)
$AUC_{0-infinity}$ (µg min/mL per mg/kg) *adjusted value*	65.00 ± 6.95 (49.24 - 75.32)	269.42 ± 25.47 (231.19 - 313.18)	247.00 ± 19.91 (218.76 - 279.16)

Adjusted value means the value obtained after adjustment of the original values expressed as mg of antimicrobial per kg bodyweight

Fig 1. Amoxicillin

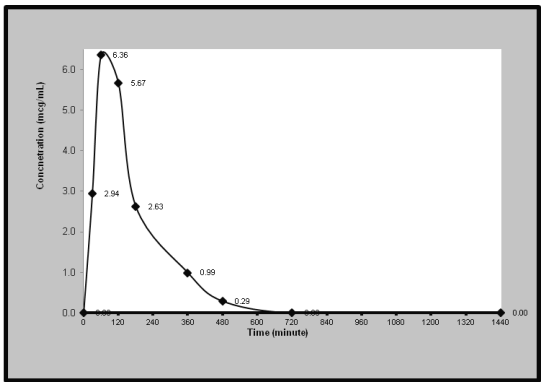

Figure 1: Time concentration curve obtained by plotting the mean plasma concentrations of amoxicillin estimated at different point of time (after single oral administration of 500 mg)

Fig 2. Flucloxacillin

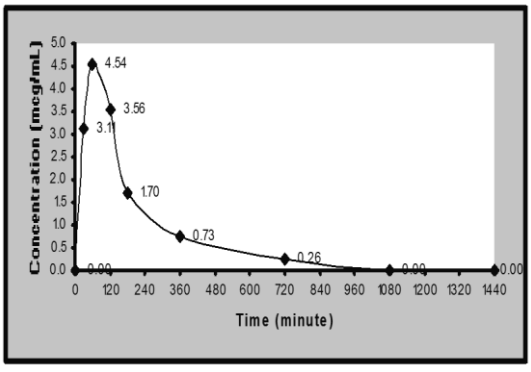

Figure 2: Time concentration curve obtained by plotting the mean plasma concentrations of flucloxacillin estimated at different point of time (after single oral administration of 250 mg)

Figure 3. Cefuroxime

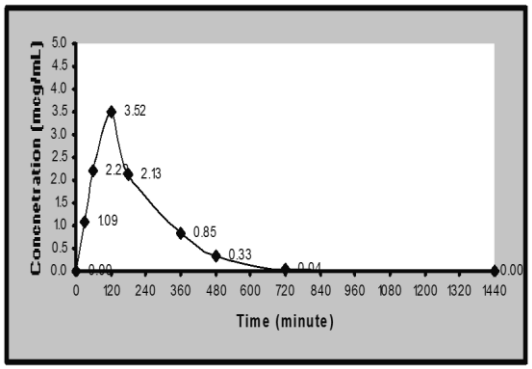

Figure 3: Time concentration curve obtained by plotting the mean plasma concentrations of cefuroxime estimated at different point of time (after single oral administration of 500 mg)

Figure 4. Ciprofloxacin

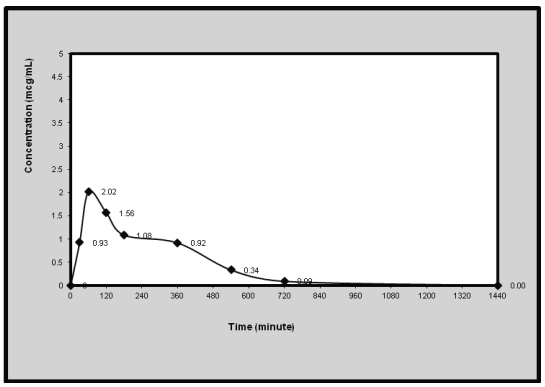

Figure 4: Time concentration curve obtained by plotting the mean plasma concentrations of ciprofloxacin estimated at different point of time (after single oral administration of 500 mg)

Figure 5. Levofloxacin

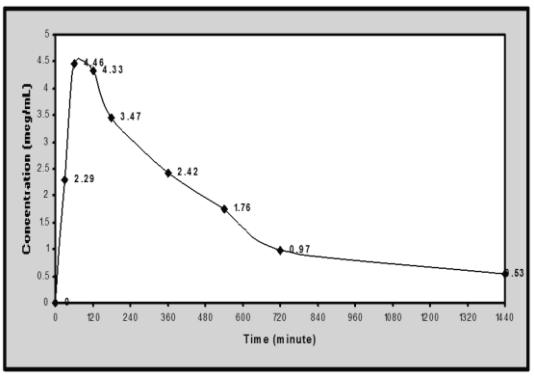

Figure 5: Time concentration curve obtained by plotting the mean plasma concentrations of levofloxacin estimated at different point of time (after single oral administration of 500 mg)

Figure 6 Gatifloxacin

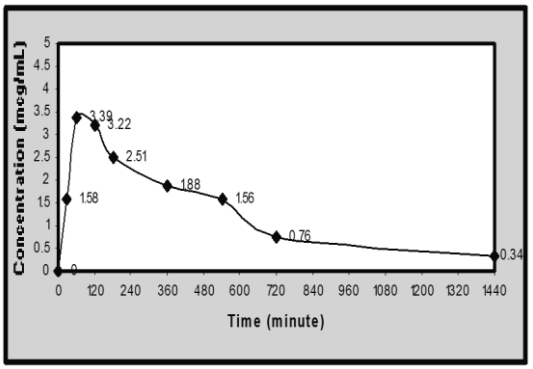

Figure 6: Time concentration curve obtained by plotting the mean plasma concentrations of gatifloxacin estimated at different point of time (after single oral administration of 400 mg)

DISCUSSION

Pharmacokinetic parameters of amoxicillin have been investigated previously in different studies at different doses of 500,[26] 875,[25] 1000[36] and 2000 mg[37]. The C_{max} and AUC_{0-8h} observed in the present study after single oral administration was lower than the findings of the previous study,[26] which might be due to the dissimilarity in the studied population. Nevertheless, the T_{max} observed in the present study was parallel to the values revealed by previous researchers using 500 mg[26] and 1000mg.[38] The similarity in this finding reiterates the fact that different doses of the same medicine have little influence on T_{max} values. The $T_{1/2}$ values, different studies revealed a range of 90 minutes to 180 minutes with a mean of 100 minutes[26,39] and the present study finding is similar to those observations.

Flucloxacillin have been studied at different doses of 1500, 2000 and 2500 mg in Chinese volunteers.[40] Regular dosing even while used intravenously, appears inadequate to treat the MSSA infections.[41] The pharmacokinetic feature was inconsistent in comparison to one study conducted in neonates, which can be explained by the dissimilarity of the studied population.[42-43] The plasma half-life ($t_{1/2}$) in the present study was higher than the previous study results,[44-45] which might be due to the slower metabolic rate of Bangladeshi population. However, the most important finding of the present study was that the C_{trough} was above than the minimum inhibitory concentration (MIC) for common strains of *Staphylococcus aureus*.[46]

In case of cefuroxime, the present study revealed that C_{max} in Bangladeshi healthy volunteer are lower than the findings of the previous study conducted with different population.[29,47] In spite of the lower C_{max}, the $AUC_{0-infinity}$ was almost equal to the previous result, which might be due to slower metabolic rate in subjects of the present study. The plasma half-life ($t_{1/2}$) higher

than the previous study[48] reiterates the possibility of slower metabolic rate. The peak serum concentration was attained little earlier (T_{max}) in the present study in comparison to previous study.[44]

Pharmacokinetic parameters of ciprofloxacin have been investigated at different doses of 250, 500, 750 and 1000 mg.[49-52] The C_{max} and AUC_{0-12h} observed in the present study was modestly lower than the previous studies.[53] The T_{max} observed in the present study was parallel to the values revealed by previous researcher, both using same dose[53] as well as different dose[54]. However, the similarity in these findings reiterating the fact that different doses of the same medicine have no influence on T_{max} values. The $T_{1/2}$ similar to previous findings, which ranged between 2.9 hours to 5.4 hours with a mean of 100 minutes.[49,50,54,55] The C_{trough} values, the present study findings are above than the minimum inhibitory concentration of the targeted microbes.

In case of levofloxacin, the C_{max} and $AUC_{0-infinity}$ was lower than the findings of the previous study,[56-57] which might be due to dissimilar study population and different dose used.[58] The plasma half-life ($t_{1/2}$) was similar to that of previous study,[47,48,56] though the peak serum concentration was attained little earlier (T_{max}) in the present study in comparison to previous reports.[45,59]

In case of gatifloxacin, the C_{max} and $AUC_{0-infinity}$ was similar to the findings of the previous studies.[54,59,60] The plasma half-life ($t_{1/2}$) was similar to one of previous study[60], however lower than other reports.[44,59] The conflicting plasma half-life indicates that the study subjects were not uniformly comparable to any of the study conducted beforehand. Though the peak serum concentration was attained little earlier (T_{max}) in the present study in comparison to previous report.[45]

Among the studied antimicrobials, the C_{max} varied to great extent even after adjustment for bodyweight and dose. The C_{max} after adjustment was highest in case of flucloxacillin and lowest in case of ciprofloxacin, indicating excellent absorption of flucloxacillin in Bangladeshi population.

CONCLUSION

The new understanding might restate the necessity of periodic pharmacokinetic study in some selected clinical cases. The information obtained through this study may provide idea about the necessity of new cut-off value for the antimicrobials.

REFERENCES

1. Rahman MS. New Global Situation in Drug Regulation: Redefined Responsibility of the Pharmacologists of Bangladesh. Bangladesh J Physiol Pharmacol 1999; 15(2): 41-42.
2. Rahman MS, Begum M, Khan IA, Chowdhury S, Islam AM, Sultana R, Hoque MZ, Akhter N. A Baseline Survey on the Use of Drugs at Private Practitioner Level in Bangladesh. Bangladesh J Physiol Pharmacol 1998; 14(1&2): 47-50.
3. Rahman MS. Changes Required in Pharmacotherapy Teaching to Ensure Rational Use of Drugs. Bangladesh J Physiol Pharmacol 1995; 11(1): 38-39.
4. Islam MS, Rahman MS, Misbahuddin M. Impact of 'Prescription Audit & Feedback' on Pattern of Prophylactic Antimicrobials in Caesarean Section: a Cost Reduction Perspective. Bangladesh J Physiol Pharmacol 2007; 23(1&2): 1-9.
5. Begum M, Rahman MS, Islam A, Khan IA, Akhter N. Eleven Years of the Undergraduate Medical Curriculum 1988: Review on the Changes in Pharmacology Written Questions. Bangladesh J Physiol Pharmacol 1999; 15(1): 27-30.
6. Das AK, Rahman MS. Prescribing vitamins at primary health care level: Exploration of facts, factors and solution. Bangladesh J Pharmacol 2011; 5(2): 92-97
7. Chowdhury AK, Rahman MS, Faroque AB, Hasan GA, Raihan SZ. Excessive use of avoidable therapeutic injections in the upazilla health complexes of Bangladesh. Mym Med J 2008; 17(2 Suppl): S59-64.
8. Jones RN, Pfaller MA, Doern GV, Erwin ME, Hollis RJ. Antimicrobial activity and spectrum investigation of eight broad-spectrum beta-lactam drugs: a 1997 surveillance trial in 102 medical centers in the United States. Cefepime Study Group. Diag Micro Infect Dis. 1998; 30(3): 215-28.
9. Maarzolini C, Telenti A, Decosterd LA, Greub G, Biollaz J, Buclin T. Efavirenz plasma levels can predict treatment failure and central nervous system side effects in HIV-1-infected patients. AIDS 2001; 15(1): 71-5.
10. Brundage RC, Yong FH, Fenton T, Spector SA, Starr SE, Fletcher CV. Intrapatient variability of efavirenz concentrations as a predictor of virologic response to antiretroviral therapy. Anti Agents Chemo. 2004; 48(3): 979-84.
11. Kappelhoff BS, van Leth F, Robinson PA, MacGregor TR, Baraldi E, Montella F, Uip DE, Thompson MA, Russell DB, Lange JM, Beinjen JH, Huitema AD, 2NN Study Group. Are adverse events of nevirapine and efavirenz related to plasma concentrations? Antiviral Therapy 2005; 10(4): 489-98.
12. Burger D, van der Heider I, la Porte C, van der Ende M, Groenveld P, Koopmans P, Kroon F, Sprenger H, Lindemans J, Schenk P, van Schaik R. Interpatient variability in the pharmacokinetics of the HIV non-nucleoside reverse transcriptase inhibitor efavirenz: the effect of gender, race, and CYP2B6 polymorphism. Br J Clin Pharmacol. 2006; 1(2): 148-54.
13. Rotger M, Csajka C, Telenti A. Genetic, ethnic, and gender differences in the pharmacokinetics of antiretroviral agents. Current HIV/AIDS Report 2006; 3(3): 118-25.
14. Stöhr W, Back D, Dunn D, Sabin C, Winston A, Gilson R, Pillay D, Hill T, Ainsworth J, Pozniak A, Leen C, Bansi L, Fisher M, Orkin C, Anderson J, Johnson M, Easterbrook P, Gibbons S, Khoo S, Liverpool TDM Database; UK CHIC Study. Factors influencing efavirenz and nevirapine plasma concentration: effect of ethnicity, weight and co-medication. Antiviral Therapy 2008; 13(5): 675-85.
15. Chou M, Bertrand J, Segeral O, Verstuyft C, Borand L, Comets E, Le Tiec C, Becquemont L, Ouk V, Mentre F, Taburet AM. Population pharmacokinetic-pharmacogenetic study of nevirapine in HIV-infected Cambodian patients. Anti Agents Chemo. 2010; 54(10): 4432-9.
16. Chen B, Zhang W, Gu Z, Li J, Zhang Y, Cai W. Population pharmacokinetic study of cyclosporine in Chinese renal transplant recipients. Euro J Clin Pharmacol. 2011; 67(6): 601-12.
17. Sugiyama E, Kaniwa N, Kim SR, Hasegawa R, Saito Y, Ueno H, Okusaka T, Ikeda M, Morizane C, Kondo S, Yamamoto N, Tamura

T, Furuse J, Ishii H, Yoshida T, Saijo N, Sawada J. Population pharmacokinetics of gemcitabine and its metabolite in Japanese cancer patients: impact of genetic polymorphisms. Clin Pharmacokinetics 2010; 49(8): 549-58.

18. Bergmann TK, Brasch-Andersen C, Gréen H, Mirza M, Pedersen RS, Nielsen F, Skougaard K, Wihl J, Keldsen N, Damkier P, Friberg LE, Peterson C, Vach W, Karlsson MO, Brosen K. Impact of CYP2C8*3 on paclitaxel clearance: a population pharmacokinetic and pharmacogenomic study in 93 patients with ovarian cancer. Pharmacogen J. 2011; 11(2): 113-20

19. Lubomirov R, di Iulio J, Fayet A, Colombo S, Martinez R, Marzolini C, Furrer H, Vernazza P, Calmy A, Cavassini M, Ledergerber B, Rentsch K, Descombes P, Buclin T, Decosterd LA, Csajka C, Telenti A; Swiss HIV Cohort Study. ADME pharmacogenetics: investigation of the pharmacokinetics of the antiretroviral agent lopinavir coformulated with ritonavir. Pharmacogenetics Genomics 2010; 20(4): 217-30.

20. Mukonzo JK, Röshammar D, Waako P, Andersson M, Fukasawa T, Milani L, Svensson JO, Ogwal-Okeng J, Gustafsson LL, Aklillu E. A novel polymorphism in ABCB1 gene, CYP2B6*6 and sex predict single-dose efavirenz population pharmacokinetics in Ugandans. Br J Clin Pharmacol 2009; 68(5): 690-9.

21. Shilbayeh S, Tutunji MF. Possible Interethnic Differences in Omeprazole Pharmacokinetics Comparison of Jordanian Arabs with Other Populations. Clin Pharmacokinetics 2006; 45(6): 593-610.

22. Chowdhury RZ, Dewan ZF. Free serum Ceftriaxone levels in children: Correlation with Protein Energy Malnutrition (PEM). J Med Sci Res 2004; 2(1): 12-18.

23. Chowdhury RZ, Islam MS, Rahman MS. Pharmacokinetic Parameters of Amoxicillin in Bangladeshi Volunteers: a Preliminary Evaluation. Bangladesh J Physiol Pharmacol 2010; 26(1&2): 1-9.

24. Chowdhury RZ, Islam MS, Rahman MS. Pharmacokinetic Parameters of Ciprofloxacin in Bangladeshi Volunteers: a Preliminary Evaluation. Bangladesh J Physiol Pharmacol 2011; 27(1&2): 1-8.

25. Kim YG, Kim HJ, Kwon JW, Kim WB, Lee MG. Bioequivalence of clarithromycin tablet formulations assessed in Korean males. Int J Clin Pharmacol Thera 2001; 39(8), pp. 356-61.

26. Pires de Abreu LR, Ortiz RM, de Castro SC, de Castro SC, Pedrazzoli J Jr. HPLC determination of amoxicillin comparative bioavailability in healthy volunteers after a single dose administration. J Pharm Pharmaceut Sci 2003; 6(2): 223-30.

27. Foroutan SM, Zarghi A, Shafaati A, Khoddam A, Movahed H. Simultaneous determination of amoxicillin and clavulanic acid in human plasma by isocratic reversed-phase HPLC using UV detection. J Pharma Biomed Analys 2007; 45(3): 531-34.

28. Landersdorfer CB, Kirkpatrick CM, Kinzig-Schippers M, Bulitta JB, Holzgrabe U, Drusano GL, Sörgel F. Population pharmacokinetics at two dose levels and pharmacodynamic profiling of flucloxacillin. Antimicrob Agents Chemo 2007; 51(9): 3290-97.

29. Al-Said MS, Al-Khamis KI, Niazy EM, El-Sayed YM, Al-Rashood KA, Al-Bella S, Al-Yamani MA, Al-Najjar TA, Alam SM, Dham R, Zaman Qumaruzaman Q. Bioequivalence evaluation of two brands of cefuroxime 500 mg tablets (Cefuzime and Zinnat) in healthy human volunteers. Bipharma Drug Disp. 2000; 21(6): 205-10.

30. Nix DE, De Vito JM, Schentag JJ. Liquid-chromatographic determination of ciprofloxacin in serum and urine. Clin Chem 1985; 31(5): 684–86.

31. Montgomery MJ, Beringer PM, Aminmanizani A, Louie SG, Shapiro BJ, Jelliffe R, Gill MA. Population Pharmacokinetics and Use of Monte Carlo Simulation To Evaluate Currently Recommended Dosing Regimens of Ciprofloxacin in Adult Patients with Cystic Fibrosis. Antimicrob Agents Chemo 2001; 45(12): 3468-73.

32. Olivera ME, Manzo RH, Junginger HE, Midha KK, Shah VP, Stavchansky S, Dressman JB, Barends DM. Biowaiver monographs for immediate release solid oral dosage forms: ciprofloxacin hydrochloride. J Pharmaceut Sci 2011; 100(1): 22-33.

33. Chien SC, Rogge MC, Gisclon LG, Curtin C, Wong F, Natarajan J, Williams RR, Fowler CL, Cheung WK, Chao AT. Pharmacokinetic Profile of Levofloxacin following once-daily 500 mg oral or intravenous doses. Anti Agents Chemo. 1997; 41(10): 2256-60.

34. Al-Dgither S, Alvi SN, Hammami MM. Development and validation of an HPLC method for the determination of gatifloxacin stability in human plasma. J Pharma Biomed Analys. 2006; 41(1): 251-55.

35. World Medical Association (WMA). WMA Declaration of Helsinki – Ethical Principles for Medical Research Involving Human Subjects. Adopted by the 18th WMA General Assembly, 1964; Helsinki, Finland, June 1964 with a series of amendments, the latest in October, 2008.

36. Baglie S, Rosalen PL, Franco LM, Ruenis AP, Baglie RC, Franco GC, Silva P, Groppo FC. Comparative bioavailability of 875 mg amoxicillin tablets in healthy human volunteers. Int J Clin Pharma Thera. 2005; 43(7): 350-54.

37. de Cássia Bergamaschi C, Motta RH, Franco GC, Cogo K, Montan MF, Ambrosano GM, Rosalen PL, Del Fiol Fde S, Groppo FC. Effect of sodium diclofenac on the bioavailability of amoxicillin. Int J Anti Agents 2006; 27(5): 417-22.

38. Molinaro M, Corona G, Fiorito V, Spreafico S, Bartoli AN, Zoia C. Bioavailability of two different oral formulations of amoxicillin in healthy subjects [abstract]. Arzneimittelforschung. 1997; 47(12): 1406-10.

39. Janknegt R, Boogaard-Van den Born J, Hameleers BA, Hooymans PM, Rang J, Smits CA, Willems-Thissen ME. Pharmacokinetics of amoxycillin in elderly in-patients [abstract]. Pharmaceutisch Weekblad Scientific edition 1992; 14(1): 27-29.

40. Huang C, Gao J, Miao L. Simultaneous determination of flucloxacillin and ampicillin in human plasma by ultra performance liquid chromatography–tandem mass spectrometry and subsequent application to a clinical study in healthy Chinese volunteers. J Pharmaceutical Biomed Analys 2012; 59(2012): 157– 161.

41. Ulldemolins M, Roberts JA, Wallis SC, Rello J, Lipman J. Flucloxacillin dosing in critically ill patients with hypoalbuminaemia: special emphasis on unbound pharmacokinetics. *J Antimicrob Chemo* 2010; 65(8): 1771-78.

42. Pullen J, de Rozario L, Stolk LM, Degraeuwe PL, van Tiel FH, Zimmermann LJ. Population pharmacokinetics and dosing of flucloxacillin in preterm and term neonates. *Thera Drug Monitor* 2006; 28(3): 351-58.

43. Pullen J, Stolk LM, Neef C, Zimmermann LJ. Microanalysis of amoxicillin, flucloxacillin, and rifampicin in neonatal plasma. *Biomed Chromato* 2007; 21(12): 1259-65.

44. Sweetman SC. [Editor]. In, Martindale The complete drug reference. Royal Pharmaceutical Society of Great Britain; Thirty third edition; 2002; Pharmaceutical Press, London, UK.

45. Anonymous. United States Pharmacopoeia Drug Information, 22nd Edition; 2002: Micromedex Thomson Healthcare, Massachusetts 02780, USA.

46. Steer JA, Hill GB, Robbins MJ, Newton JC, Wilson AP. In-vitro modelling of the bactericidal activity of teicoplanin versus flucloxacillin as used in surgical prophylaxis, against Staphylococcus aureus. *J Antimicrob Chemo 1*997; 39(4): 477-81.

47. McEvoy GK. [Editor]. Amoxicillin. In, AHFS Drug Information 1999. American Society of Health System Pharmacists; Bethesda, MD 20814, 1999(a); USA: 369-72.

48. McEvoy GK. [Editor]. Ciprofloxacin. In, AHFS Drug Information 1999. American Society of Health System Pharmacists; Bethesda, MD 20814, 1999(b); USA: 670-84.

49. Forrest A, Ballow CH, Nix DE, Birmingham MC, Schentag JJ. Development of a Population Pharmacokinetic Model and Optimal Sampling Strategies for Intravenous Ciprofloxacin. Anti Agents Chemo 1993; 37(5): 1065-72.

50. Shah A, Lettieri J, Blum R, Millikin S, Sica D, Heller AH. Pharmacokinetics of intravenous ciprofloxacin in normal and renally impaired subjects. J Antimicrob Chemo 1996; 38(1): 103-16.

51. Khan MK, Khan MF. Assessment of bioequivalence of ciprofloxacin in healthy male subjects using HPLC. Pak J Pharmaceut Sci 2008; 21(3): 299-306.

52. Khan MK, Khan MF, Khan H, Mustafa G. Bioavailability of ciprofloxacin tablets in humans and its correlation with the dissolution rates. Pak J Pharmaceut Sci 2009; 22(3): 329-34.

53. Shah A, Liu M, Vaughan D, Heller AH. Oral bioequivalence of three ciprofloxacin formulations following single-dose administration: 500 mg tablet compared with 500 mg/10 mL or 500 mg/5 mL suspension and the effect of food on the absorption of ciprofloxacin oral suspension. J Antimicrob Chemo 1999; 43(Suppl A): 49-54.

54. Lubasch A, Keller I, Borner K, Koeppe P, Lode H. Comparative Pharmacokinetics of Ciprofloxacin, Gatifloxacin, Grepafloxacin, Levofloxacin, Trovafloxacin, and Moxifloxacin after Single Oral Administration in Healthy. Antimicrob Agents Chemo 2000; 44(10): 2600-03.

55. Varela JE, Cohn SM, Brown M, Ward G, Namias N, Spalding PB. Pharmacokinetic and burn Escher penetration of intravenous ciprofloxacin in patients with major thermal injuries. *J Antimicrob Chemo* 2000; 45(3): 337.42.

56. Wagenlehner FM, Kinzig-Schippers M, Tischmeyer U, Wagenlehner C, Sörgel F, Dalhoff A, Naber KG. Pharmacokinetics of ciprofloxacin XR (1000 mg) versus levofloxacin (500 mg) in plasma and urine of male and female healthy volunteers receiving a single oral dose. Int J Antimicrob Agents 2006; 27(1): 7-14.

57. Zhang YY, Huang HH, Ren ZY, Zheng HG, Yu YS, Lü XJ, Xiao ZK, Yang HF, Xiu QY, Chen BY, Yue HM, Hao QL, Huang JA, Ma H, Xiao W, Guo DY, Si B, Sun SH, Zhang W, Li QH, Shen HH, Duan J, Li HY, Yao WZ, Gu JM, Xia QM, Ying KJ, Liu A, Yang HP, Shi MH, Sun TY, Ding GH, Wu GM. Clinical evaluation of oral levofloxacin 500 mg once-daily dosage for treatment of lower respiratory tract infections and urinary tract infections: a prospective multicenter study in China. J Inf Chemo 2009; 15(5): 301-11.

58. Nicolle L, Duckworth H, Sitar D, Bryski L, Harding G, Zhanel G. Pharmacokinetics/pharmacodynamics of levofloxacin 750 mg once daily in young women with acute uncomplicated pyelonephritis. Int J Antimicrob Agents 2008; 31(3): 287-9.

59. Kastrup EK. [Editor] Drug facts and Comparison. 60th Edition; Wolters Kluwer Health, Missouri 63146-3125, 2006: USA.

60. Boy D, Well M, Kinzig-Schippers M, Sörgel F, Ankel-Fuchs D, Naber KG. Urinary bactericidal activity, urinary excretion and plasma concentrations of gatifloxacin (400 mg) versus ciprofloxacin (500 mg) in healthy volunteers after a single oral dose. Int J Anti Agents 2004; 23(Suppl 1) S6-16.