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ARTICLE

Longitudinal evaluation of azithromycin and cytokine concentrations in amniotic fluid following one-time oral dosing in pregnancy

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Abstract

To utilize noninvasive collection of amniotic fluid in the setting of preterm premature rupture of membranes (PPROMs) to report the time concentration profile of azithromycin in amniotic fluid over 7 days from a single dose, and evaluate the correlation between azithromycin concentration and inflammatory markers in amniotic fluid. Prospective cohort study of five pregnant patients admitted with PPROMs and treated with a single 1 g oral azithromycin dose. Amniotic fluid was collected from pads and used to quantify azithromycin concentration as well as TNFa, IL-1a, IL-1b, IL-6, IL-8, and IL-10 concentrations. Primary outcome was time/concentration profile of azithromycin in amniotic fluid. Secondary outcome included correlation between azithromycin concentration and cytokine concentrations. Five patients were enrolled. Mean gestational age on admission with PPROM was 27.5 ± 2.3 weeks with a median latency of 7 days (interquartile range [IQR] = 4–13). A median of two samples/day (IQR = 1–3) were collected per participant. Azithromycin was quantified in duplicate; intra-assay coefficient of variation was 17%. Azithromycin concentration was less than 60 ng/ml after day 3. Azithromycin concentration was positively correlated with IL-8 ($r = 0.38$, $p = 0.03$), IL1a ($r = 0.39$, $p = 0.03$), and IL-1b (r = 0.36, $p = 0.04$) in amniotic fluid. Azithromycin is detectable in amniotic fluid over 7 days from a single 1 g maternal dose, however, it is not sustained over the range of minimum inhibitory concentration for common genitourinary flora. Based on correlation with specific cytokines, azithromycin penetration in amniotic fluid may relate to maternal monocyte concentration in amniotic fluid in the setting of PPROM.

Study Highlights

WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

Azithromycin is used to prevent infection in setting of preterm premature rupture of membranes (PPROMs), but there is no established dose. One cross-sectional study with one patient found amniotic fluid azithromycin concentration was below

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minimum inhibitory concentration (MIC)50 of common genitourinary (GU) flora 7 days after a single 1 g maternal dose. There is no reported correlation between maternal azithromycin treatment and the concentration of drug and inflammatory cytokines in amniotic fluid.

WHAT QUESTIONS DID THIS STUDY ADDRESS?

Our objective was to utilize noninvasive collection of amniotic fluid in pregnant patients admitted with PPROM to address whether one-time maternal dosing of azithromycin can produce a sustained amniotic fluid concentration greater than the MIC of common GU flora over 7 days, and secondarily, whether amniotic fluid azithromycin concentration was correlated with inflammatory cytokine concentrations.

WHAT DOES THIS STUDY ADD TO OUR KNOWLEDGE?

Azithromycin is detectable in amniotic fluid over 7 days from a single 1 g maternal dose, however, it is not sustained over the range of MIC for common GU flora. Azithromycin concentration in amniotic fluid is correlated with specific cytokines that suggest that azithromycin penetration in amniotic fluid may relate to maternal monocyte concentration in amniotic fluid in the setting of PPROM.

HOW MIGHT THIS CHANGE CLINICAL PHARMACOLOGY OR TRANSLATIONAL SCIENCE?

Amniotic fluid may be collected noninvasively from patients with PPROM and used to evaluate the fetal environment, including medication penetrance and inflammatory markers. Given the benefit of maternal and fetal exposure to azithromycin for prevention of adverse perinatal outcomes, it is important to study how maternal dosing leads to azithromycin concentration within the maternal fetal unit in order to optimize therapy.

INTRODUCTION

Preterm birth complicates ~ 10% of pregnancies, is the leading cause of neonatal morbidity and mortality, and has rising incidence in the United States over the past 3 years.[1] Preterm premature rupture of membranes (PPROMs) is responsible for ~ 25% of preterm births. Prior to 34 weeks' gestation, PPROM is managed expectantly with the goal of prolonging pregnancy, which is associated with improved neonatal outcomes. Broad spectrum antibiotics increase latency to delivery, reduce the rate of intra-amniotic infection, and improve neonatal morbidity.[2,3] The historically recommended regimen is i.v. erythromycin and ampicillin for 48 h followed by p.o. erythromycin and amoxicillin for 5 days (total 7 day course).[2] There is no consensus of optimal azithromycin dosing in PPROM and a variety of dosing regimens are used.[4] Pharmacokinetic studies comparing azithromycin dosing in pregnancy or the neonatal impact of maternal dosing in PPROM are lacking, resulting in a critical gap in knowledge.

Azithromycin accumulates in phagocytes and is delivered in high concentration to sites of infection, which is why it may be dosed infrequently and serum levels alone do not reflect efficacy.[5] The exact mechanism of action by which macrolides in increased latency and reduced chorioamnionitis in PPROM is unclear. PPROM is not always associated with specific bacterial infection, although even in the absence of identified infection it is generally an inflammatory condition and is associated with both clinical (~ 15% incidence) and histologic inflammation, also known as chorioamnionitis (~ 60%).[4] Most puerperal infections are polymicrobial and azithromycin is active against common genitourinary bacteria. Immunomodulatory properties may also increase latency to delivery by inhibiting inflammatory pathways, which precipitate preterm birth.

Physiologic changes in pregnancy can impact a drug's pharmacokinetics (PKs), these changes include changes in hepatic metabolism, significantly increased renal clearance, significantly increased total body water, plasma volume, and adipose content, all of which impact drug bioavailability.[6] Azithromycin PKs, however, are not dependent on those systems. It is rapidly distributed from serum into intracellular compartments, and ultimately to tissues. It has low serum binding, and most of the absorbed drug remains unmetabolized within the body. Due to its PKs, serum concentrations of azithromycin are not clinically relevant, rather its concentration at tissue of action is a better predictor of efficacy.[7] Fetal exposure can be measured through cord blood, amniotic fluid, and placental concentration. Concentration in umbilical cord blood alone may not be indicative of fetal exposure for a medication like azithromycin where tissue distribution

rapidly depletes serum concentrations.[8,9] Given the important role of intra-amniotic infection and inflammation in perinatal outcomes in preterm birth and PPROM, it is critical to evaluate azithromycin concentration in the amniotic fluid compartment.

Pregnancy-specific PKs of azithromycin are poorly defined.[10] One study in pregnancy[9] evaluated a single 1 g p.o. azithromycin dose, but only had one participant with data >3 days postdose. Azithromycin concentration was above minimum inhibitory concentration (MIC)50 at day 3 but fell below MIC50 by day 7 ($N = 1$) in uterine myometrium, placenta, and amniotic fluid for genital *C. trachomatis* (MIC50 = 60 ng/ml),[11] and *Ureaplasma spp* (MIC50 = 500 ng/ml).[12] Another study examining azithromycin PKs 8 h after a 500 g i.v. maternal dose found median concentration in amniotic fluid was 33 ng/ml around 1 h postdose, well below MIC50 for ureaplasma.[13] A primate model found that compared to a one-time dose, daily dosing resulted in sustained elevated azithromycin concentration in amniotic fluid.[14] Although these studies are limited in number, size, and scope, they suggest that the one-time 1 g dosing of azithromycin commonly used may be suboptimal for a 1 week therapy course in PPROM.

There are no established pharmacodynamic (PD) markers of azithromycin therapy. In non-pregnant patients, azithromycin attenuates the expression of inflammatory cytokines IL-6, IL-8, and TNFa, and enhances expression of IL-10, an anti-inflammatory cytokine.[5] In PPROM, earlier delivery,[15] choriamnionitis,[16,17] funisitis, and fetal inflammatory response[18,19] are associated with elevated inflammatory cytokines, especially IL-6 and IL-8, in maternal serum and amniotic fluid. Azithromycin may improve latency to delivery through both antibacterial and immunomodulatory effects and these cytokines are relevant biomarkers to study in evaluating azithromycin dosing.

Previous studies have demonstrated the ability to noninvasively sample amniotic fluid,[18] and the correlation between noninvasive sampling of amniotic fluid and invasive sampling with regard to inflammatory markers[18,20] and microbial invasion.[20] Our objective was to utilize noninvasive collection of amniotic fluid in the setting of PPROM to report the time concentration profile of azithromycin in amniotic fluid over 7 days from a single dose, and evaluate the correlation among azithromycin concentration, inflammatory markers in amniotic fluid, and placental pathological markers, including histologic chorioamnionitis. We chose to focus on amniotic fluid azithromycin concentration because, unlike serum, it reflects target site concentration, and unlike myometrium or cord blood, it can be sampled throughout pregnancy, not just at delivery. This allows for a longitudinal evaluation of azithromycin concentration at the site of action for a clinically relevant PK model that can be used to guide dosing for the purpose of improving perinatal outcomes.

METHODS

Study design

This is a prospective, observational cohort study of pregnant patients admitted with PPROM at Thomas Jefferson University Hospital from November 2019 to March 2020 who received azithromycin for latency. Azithromycin was administered as part of standard clinical care. The only exclusion was non-English speaking, as translated consents were not available. Recruitment for this effort stopped in March 2020 due to the coronavirus disease 2019 pandemic restrictions and then subsequent initiation of a randomized trial relating to PPROM (NCT04294069). All patients received a standard of care dose of 1 g oral azithromycin once as part of latency antibiotic regimen (in addition, patients received i.v. ampicillin/oral amoxicillin). Participants were given Always flex-foam pads (Proctor & Gamble) to line their underwear. As amniotic fluid leaked, they changed their pads as per their comfort and pads were collected. Because pads were collected per participant comfort rather than at timed intervals, sample time points were grouped in 8-h windows to allow comparison among participants. Amniotic fluid was extracted from pads using a press, samples were centrifuged at 1500 *g* for 10 min and stored at −80 degrees similar to previous study.[18] In addition, baseline characteristics, gestational age, latency to delivery, and delivery outcomes were collected. All participants provided written consent and this study was approved by Thomas Jefferson University institutional review board.

Azithromycin quantification

Liquid chromatography mass spectrometry (LCMS) analysis for standards and samples were performed using Thermo Exactive Orbitrap coupled with Dionex 3000 HPLC system. Mass spectrometry method was established in the scanning mass range of 240 to 420 m/z, using Thermo Exactive Plus Orbitrap mass spectrometer (Thermo Scientific). All the scans were performed under positive ion mode and electron spray ionization. Sheath gas flow rate was maintained at 20 psi; auxiliary gas flow was 5 psi; electron spray voltage used was 4 kV; and capillary temperature was 375°C. Capillary, tube lens, and skimmer voltage were kept at 27.50, 65, and 16 V, respectively. These parameters were optimized using Xcaliber software to get target mass intensity of 106 to 107 units, for azithromycin. The method was integrated with the high-performance liquid chromatography HPLC method to get method for LCMS runs.

Analysis was performed using Dionex Ultimate 3000 HPLC system (Thermo Fischer) attached to Thermo orbitrap mass spectrometer. LCMS runs of calibration standard and

isocratic elution with 50:50 (water with 0.1% formic acid [FA]: acetonitrile [can] with 0.1% FA). All the samples and standards injection volume were set to 5 µl and chromatographic separations were performed using HSS XSelect C18 reverse phase column (4.6 × 100 mm), Waters (Milford, MA) at flow rate of 0.25 ml/min with run time of 5 min. Column compartment and sampler temperature was set to 30 and 20°, respectively. LCMS data (for samples and standards) was acquired and saved for further quantitative analysis.

Calibration curve was established using blank amniotic fluid spiked with azithromycin. Primary stocks of azithromycin were made in the concentration range of 10 to 2000 ng/ml in ACN. These primary stocks were spiked in the amniotic fluid at a concentration range of 1 to 200 ng/ml with 100 µl as final volume and azithromycin-D3 as an internal standard at final concentration of 100 ng/ml. Extraction of azithromycin (from calibration spike standards and samples) was performed using protein precipitation method, with amniotic and ACN ratio as 1:2.

Azithromycin concentration in amniotic fluid was ascertained with LCMS. Mass spectrometry method was established in the scanning mass range of 240 to 420 m/z, using Thermo Exactive Plus Orbitrap mass spectrometer (Thermo Scientific). Analysis was performed using Dionex Ultimate 3000 HPLC system (Thermo Fischer) attached to Thermo orbitrap mass spectrometer. The lower limit of detection and quantitation was found to be 1 and 10 ng/ml, respectively.

Cytokine quantification

Samples were analyzed using the human Cytokine/Chemokine Magnetic Bead Panel Millipore plates specific for the following analytes: TNF-α, IL-1α, IL-1β, IL-6, IL-8, and IL-10. Samples were analyzed in duplicate by a FlexMAP 3D (Luminex) and average value used. Standard curves were generated for each cytokine, and median fluorescent intensities were transformed into concentrations by six-point, nonlinear regression.

Statistical analysis

Because participants leaked fluid and changed the pads ad lib, any sample collected 0–8 h was marked as 8 h postdose, any sample collected 8–16 was marked as 16 h postdose and so on, this allowed data to be grouped and analyzed across participants. The primary outcome was to describe the concentration/time profile of azithromycin in amniotic fluid over the course of 7 days from a single 1 g oral azithromycin maternal dose. Secondary outcomes include the correlation between azithromycin concentration and cytokines IL-1a, IL-1b, IL-6, IL-8, IL-10, and TNF-α, as well as to examine the correlation between these and histologic chorioamnionitis. Correlation among azithromycin concentration, inflammatory markers, and histologic chorioamnionitis was evaluated with Pearson correlation coefficient. The R package was used for statistical analysis. Any p value less than 0.05 was considered significant.

RESULTS

Five patients were enrolled, one set of twins (ID #1) and four singleton gestations. Mean gestational age on admission with PPROM was 27.5 ± 2.3 weeks and body mass index 32.8 ± 6.2 kg/m^2 (Table 1). Mean gestational age at delivery was 28.7 ± 2.6 weeks with a median latency of 7 days (interquartile range (IQR) = 4–13). Four (80%) patients had histologic chorioamnionitis and one (20%) had clinical chorioamnionitis. Azithromycin was well-tolerated and there was no treatment-related adverse event.

Sampling

A median of two samples/day (IQR = 1–3) were collected per participant. Up to 10 cc of amniotic fluid per pad was collected and stored; median sample volume was 6 cc (IQR = 3–8). Azithromycin was successfully quantified in duplicate. Coefficient of variation between duplicates was 17%. In examining variation within each day, overall, the coefficient of variation (SD/mean) appeared to trend down after 48 h (Table 2).

TABLE 1 Baseline characteristics and delivery outcomes of included participants with PPROM receiving 1 g azithromycin one time on admission

Baseline characteristics	
Race *N* (%)`	
African American	3 (60)
White	1 (20)
Hispanic	1 (2)
Age	30.8 ± 3.3
BMI (kg/M^2)	32.8 ± 6.2
Gestational age on admission (weeks)	27.5 ± 2.3
Twin gestation	1 (20%)
Delivery outcomes	
Gestational age at delivery (weeks)	28.7 ± 2.6
Latency (days)	7 [4–13]
Histologic chorioamnionitis	4 (80)
Clinical chorioamnionitis	1 (20)

Note: Data presented as mean ± SD or median [IQR] or *N* (%).

Abbreviations: BMI, body mass index; IQR, interquartile range.

TABLE 2 Azithromycin concentration in amniotic fluid 1–7 days after a single 1 g oral azithromycin maternal dose

Amniotic fluid AZ (ng/ml)	Day 0 (0–24 h)	Day 1 (24–48 h) $N = 4, n = 9$	Day 2 (48–72 h) $N = 4, n = 9$	Day 3 (72–96 h) $N = 4, n = 11$	Day 4 (96–120 h) $N = 2, n = 6$	Day 5 (120–144 h) $N = 3, n = 8$	Day 6 (144–168 h) $N = 2, n = 6$	Day 7 (168–172 h) $N, n = 1$
Mean ± SD	N/A	63 ± 59	53 ± 41	33 ± 23	27 ± 16	53 ± 22	27 ± 18	18
Coefficient of variation (SD/mean)	N/A	0.94	0.77	0.70	0.59	0.42	0.67	N/A
Median [IQR]	N/A	46 [18–102]	42 [19–97]	25 [21–41]	21 [7–42]	54 [45–71]	27 [10–45]	18

Note: There were no samples collected 0–24 h for any participants so that data is not available (N/A). N = number of participants and n = total number of samples.

Abbreviations: AZ, azithromycin; IQR, interquartile range.

Azithromycin pharmacokinetics and pharmacodynamics

Azithromycin concentration trended down and was less than 60 ng/ml after day 3 (Figure 1a,b). Azithromycin concentration was positively correlated with IL-8 ($r = 0.38$, $p = 0.03$), IL1a ($r = 0.39$, $p = 0.03$), and IL-1b ($r = 0.36$, $p = 0.04$) in amniotic fluid (Figure 2). Azithromycin concentration was not correlated with TNF-α ($r = 0.30$, $p = 0.07$), IL-6 ($r = -0.06$, $p = 0.75$), or IL-10 ($r = 0.23$, $p = 0.2$) concentration in amniotic fluid. Only IL1b ($r = 0.49$, $p = 0.004$), TNF-α ($r = 0.45$, $p = 0.009$), and IL-8 ($r = 0.78$, $p < 0.001$) were correlated with histologic chorioamnionitis (Figure 3).

DISCUSSION

Main findings

We have demonstrated a persistent, although declining, level of azithromycin in amniotic fluid through 7 days from a single 1 g maternal dose. One gram of maternal azithromycin dose did not maintain amniotic fluid concentrations above MIC for common genitourinary (GU) bacteria through the 7-day sampling period. Azithromycin concentration was correlated with monocyte specific cytokine concentration, suggesting azithromycin penetration in amniotic fluid may be related to maternal monocyte concentration in amniotic fluid in the setting of PPROM.

Results in the context of what is known

Using a noninvasive technique, we have demonstrated reliable azithromycin quantification in amniotic fluid with a coefficient of variation less than 20%, which is deemed acceptable by the US Food and Drug Administration's (FDA's) standards.[21] Our coefficient of variation each day postdose was similar to that found by Ramsey et al. who used amniocentesis for sampling suggesting our technique for noninvasive sampling is reliable and comparable to amniocentesis. Previous studies have described this noninvasive collection technique for quantification of inflammatory markers and demonstrated results were concordant with samples collected by invasive amniocentesis.[20] Our study builds on the work of Ramsey et al. with increased sampling, longitudinal rather than cross-sectional data, and more participants and more samples greater than 3 days after dosing.[9] This is the first study to our knowledge that evaluates noninvasive collection of amniotic fluid for both longitudinal azithromycin concentration, and correlation between drug concentration and inflammatory markers.

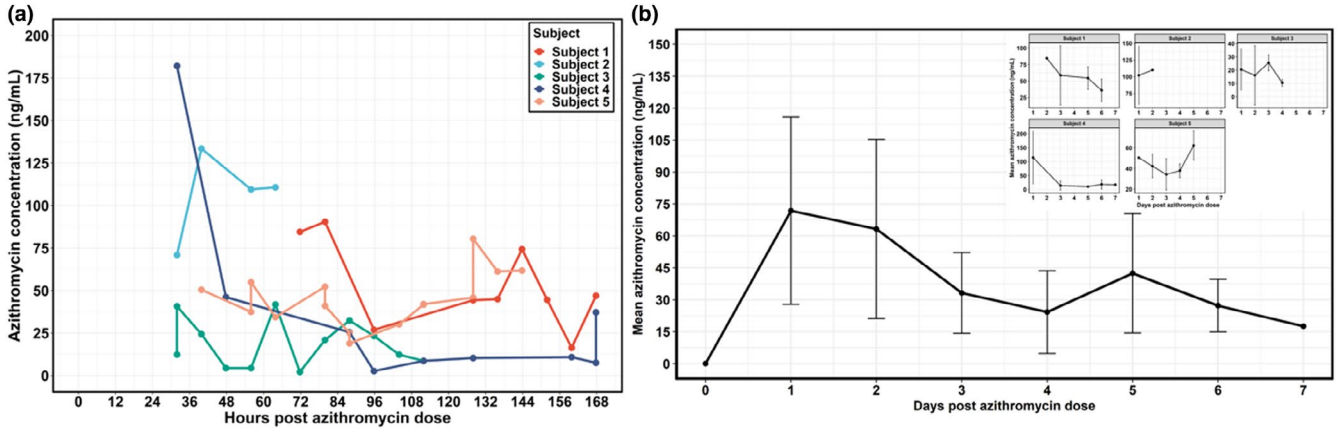

FIGURE 1 Azithromycin concentration in amniotic fluid following 1 g oral maternal dose in patients with preterm premature rupture of membranes. (a) Individual azithromycin concentration/time plots (b) Azithromycin concentration/time plots for individual and aggregate with azithromycin concentration averaged over a 24-h period

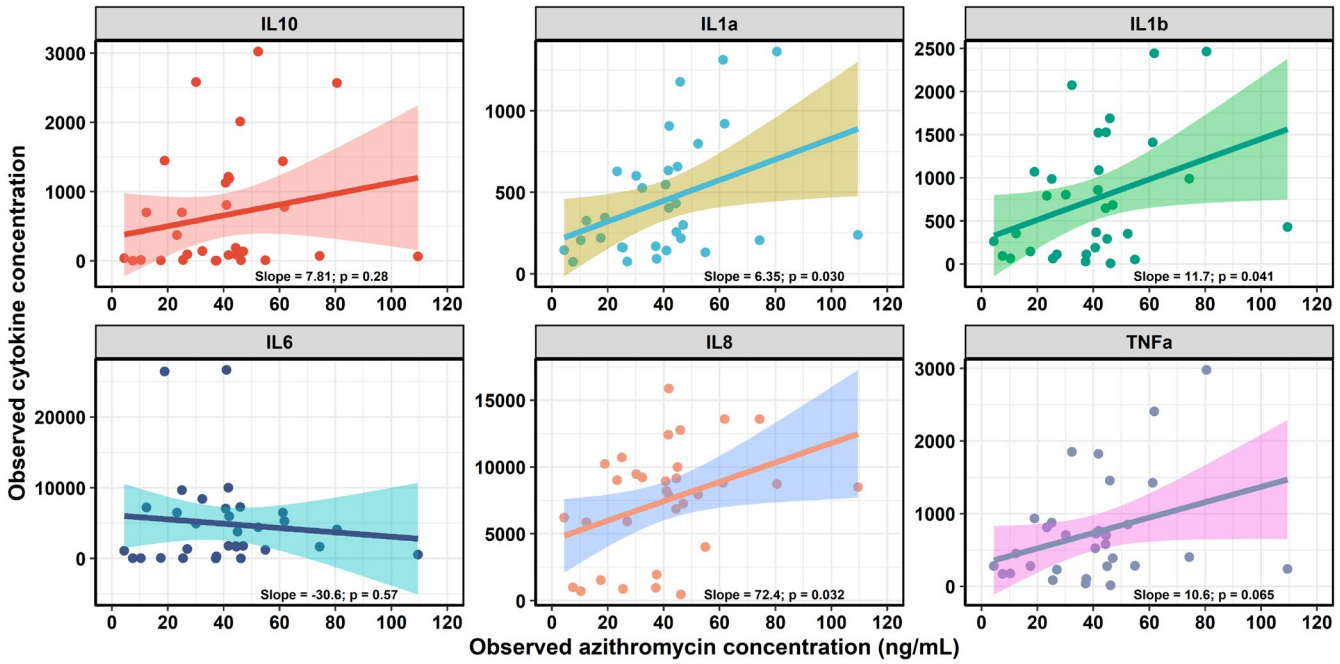

FIGURE 2 Correlation between mean azithromycin concentration (ng/ml) and cytokine concentration (pg/ml) in amniotic. Linear regression performed for azithromycin versus individual cytokine concentrations, $p < 0.05$ considered significant

Azithromycin efficacy is generally reported by its MIC. Efficacy of azithromycin in PPROM is difficult to estimate because there is no single bacteria targeted. Common GU bacteria vary in reported MIC—*C. trachomatis* (30–125 ng/ml), *N. gonorrhoeae* (16–60 ng/ml), *Staphylococcus* and *Streptococcus* spp (64–500 ng/ml), and *Ureaplasma spp* (500 ng/ml). A single azithromycin dose was not sufficient to maintain amniotic fluid concentrations above this entire polymicrobial range of MIC.

Fetal exposure to any medication depends on placental transfer. Substrates that are small, lipophilic, or uncharged, are more likely to be able to passively diffuse from maternal blood, across the placental interface, and be taken into fetal circulation. In addition, there are active placental transporters that may either concentrate or diminish the fetal exposure of any maternally administered medication.[6] By the third trimester, amniotic fluid is produced primarily from fetal urinary excretion and respiratory excretion; transudation from fetal skin and placenta are limited after ~ 22–24 weeks. Fetal swallowing and reabsorption of any drug in amniotic fluid can lead to increased fetal accumulation of drug.

It is notable, that while azithromycin is undetectable in maternal serum after 4–7 days after a single dose,[9] it

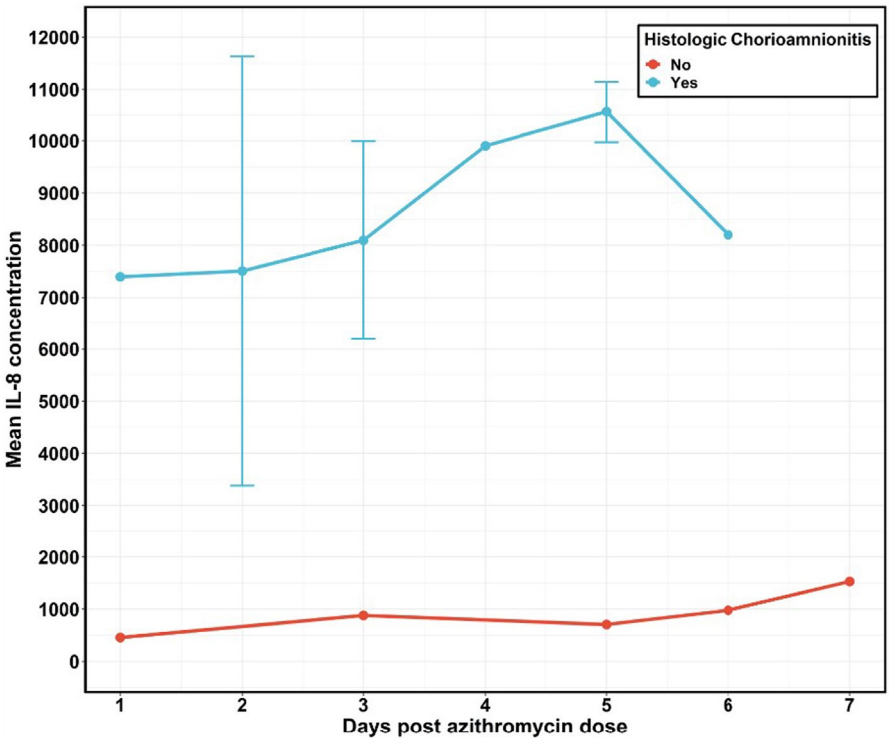

FIGURE 3 Correlation between amniotic fluid IL-8 concentration and histologic chorioamnionitis

persists in amniotic fluid. Amniotic fluid is an active tissue and plays an important role in innate and cellular immunity. Although monocytes and neutrophils are almost undetectable in the amniotic fluid of a normal pregnancy, they are increased in the setting of PPROM and intra-amniotic infection.[22,23] Azithromycin accumulates in phagocytes, therefore, intra-amniotic infection or inflammation may be associated with a higher amniotic fluid azithromycin concentration due to increased monocyte localization. One study of pregnant women found that amniotic fluid monocytes were increasingly of maternal origin as intra-amniotic infection progresses.[24] Notably, the same investigators found that monocytes in amniotic fluid primarily produced IL1-a, IL-1b, and IL-8,[23] which we also found were the cytokines correlated with higher azithromycin concentration in amniotic fluid. Thus, in this setting, fetal exposure to azithromycin may relate primarily to maternal monocyte transfer into amniotic fluid rather than more traditional mechanisms of placental drug transfer.

Our data are not sufficient to establish PD markers of azithromycin therapy. We are unable to differentiate cause/effect—do higher inflammatory markers imply increased inflammation, increased monocytes, and therefore increased azithromycin concentration? Will this eventually result in reduced inflammatory markers? The reported anti-inflammatory effect of azithromycin is a secondary effect and not as a first-line effect. Further study needs to be done on how azithromycin concentration in the fetal compartment impacts inflammatory pathways and pregnancy outcome.

Research implications

A better understanding of the disposition over time of azithromycin in the maternal/fetal compartment is critical to better understanding its use, especially in the situations of cervical insufficiency and PPROM, where the goal is to reduce inflammation, reduce the rate of chorioamnionitis, and increase latency to delivery. Further study should be done to evaluate the interaction between azithromycin concentration and amniotic fluid inflammatory markers, which have been associated with both maternal and neonatal outcomes; these markers are potential PD indicators for azithromycin therapy and/or prognostic indicators for outcome. Additionally, it is critical to evaluate how maternal azithromycin dosing impacts fetal exposure and potentially neonatal outcomes, including risk of respiratory illness,[25–27] in order to determine appropriate maternal azithromycin dosing.

Clinical implications

Azithromycin is used routinely in PPROM, but is also being studied in other high-risk pregnancy conditions, including cervical insufficiency,[28] prevention of maternal/neonatal sepsis,[29] and perioperative prophylaxis.[30] A better understanding of azithromycin disposition in the maternal fetal unit would allow for rational dosing strategies for each of these conditions. The commonly used single 1 g maternal azithromycin dose may not be optimal to maintain MIC antibiotic concentration for the expected 7-day course in the setting of

PPROM. Pharmacometric modeling approaches quantitate drug exposure to PK response, and the data presented here could inform generation of optimized dosing regimens using PK/PD simulations.

Strengths and limitations

This study has a number of strengths. We used noninvasive sampling to study azithromycin PKs in the population of interest, those with PPROM, for whom serial amniocentesis may not be feasible. Furthermore, given the inflammatory state of PPROM and the unique nature of azithromycin to concentrate locally, PK studies on pregnant healthly women at term may not be applicable to a patients with preterm PPROM. This is the only study we could identify with longitudinal sampling of multiple participants over 7 days following maternal azithromycin dosing. We focused on azithromycin concentration in the amniotic fluid as this may be most clinically relevant for evaluating its efficacy.

There are also a number of limitations, the sample size is limited, which does not allow for any conclusions regarding azithromycin concentration, inflammatory milieu, and clinical outcomes. Given the sparse opportunistic sampling, there were no samples available for the first 24 h postdose when azithromycin concentration would be expected to peak[9] thus we could not report maximum plasma concentration (C_{max}), time to Cmax (T_{max}), area under the curve (AUC), or clearance. Traditional PK parameters, such as C_{max} and T_{max} for azithromycin in pregnancy have previously been reported.[9,13] We were interested in the trough levels, as maintaining levels above MIC for 7 days is what is clinically important for azithromycin dosing in PPROM. Only one dosing regimen was used, thus doses cannot be compared. Due to additional azithromycin dosing prior to cesarean delivery in the setting of PPROM,[31] it was not possible to universally evaluate placental or cord blood azithromycin concentrations from initial azithromycin dosing on admission. It is possible that leakage of urine was captured, however, this is unlikely given our values are similar to prior reported concentration of azithromycin in amniotic fluid and over 200-fold less than what is observed in maternal urine.[9]

CONCLUSION

Amniotic fluid may be collected noninvasively from patients with PPROM and used to evaluate the fetal environment, including medication penetrance and inflammatory markers. This is particularly important when evaluating therapies aimed at the maternal/fetal unit (uterus/placenta/membranes/amniotic fluid/fetus), as many interventions for prevention of preterm birth and puerperal infection are. A single dose of azithromycin does not maintain amniotic fluid concentration above MIC for many common GU pathogens over 7 days. Azithromycin concentration in amniotic fluid along with inflammatory markers may be used to compare dosing regimens and select one that optimizes concentration of drug at the site of action.

CONFLICT OF INTEREST

The authors declared no competing interests for this work.

AUTHOR CONTRIBUTIONS

All authors wrote the manuscript. R.C.B., E.L., A.R., and W.K.K. designed the research. R.C.B., A.R., and G.K. performed the research. R.C.B. and E.L. analyzed the data.

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