The pharmacokinetics of isoniazid in low birth weight and premature infants

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Short running title: Isoniazid pharmacokinetics in infants

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Abstract

Background: Isoniazid (INH) is recommended as post-tuberculosis exposure preventive therapy in children. However, no pharmacokinetic data are available for INH in low birth weight (LBW)-infants, who are undergoing substantial developmental and physiological changes.

Objectives: To determine the pharmacokinetic parameters of INH at 10 mg/kg/day, and to define its pharmacokinetics relative to arylamine $N$-acetyltransferase-2 ($NAT2$)-genotype.

Methods: A prospective, intensive pharmacokinetic sampling study was conducted at Tygerberg Children’s Hospital, South Africa, measuring INH plasma concentrations at 2, 3, 4 and 5 hours post dose.

Results: Twenty LBW-infants (14 male; 16 HIV-exposed) were studied. Median birth weight was 1575 (interquartile range: 1190-2035) grams and median gestational age was 35 (interquartile range: 34-38) weeks. $NAT2$ acetylation status was as follows: 5 homozygous slow (SS); 11 heterozygous intermediate (FS) and 4 homozygous fast (FF).

Using a non-compartmental analysis approach, the median $C_{\text{max}}$ (maximum concentration) was 5.63 $\mu$g/ml, $T_{\text{max}}$ (time to $C_{\text{max}}$) was 2 hours, $AUC_{2-5}$ (area under the concentration-time curve) was 13.56 $\mu$g.h/ml, $t_{1/2}$ (half-life) was 4.69 hours and $k_e$ (elimination constant rate) was 0.15 h$^{-1}$. Alanine aminotransferase levels were normal, apart from 2 isolated values at twice and three times above normal. Only the three times elevated value was repeated at 6 months and normalized.
Conclusions: All LBW-infants achieved target INH plasma concentrations comparable to adult values. Reduced elimination was observed in smaller and younger infants, and in slow acetylators, cautioning against higher doses. Safety data, although limited, was reassuring. More data, however, are required in newborn infants.
Introduction

The HIV pandemic has seen a dramatic increase in tuberculosis (TB) rates amongst pregnant women(1). Maternal TB, regardless of HIV status, is associated with a high incidence of low birth weight (LBW; < 2500 grams) and prematurity (gestational age < 38 weeks) in infants(2-4). In South Africa, with its high burden of both TB and HIV, recent advances in neonatal care have improved infant survival, resulting in a considerable group of small and premature TB-exposed newborns requiring preventive therapy against TB. Isoniazid (INH) is the most widely recommended agent for preventing TB disease in children(5). Following Mycobacterium tuberculosis (M. tb) infection, up to 50% infants (< 12 months) will progress to TB disease in the absence of INH preventive therapy (IPT)(6), while post-exposure IPT reduces the risk of TB disease by 60-65%(7). There are limited pharmacokinetic data on INH in young children and none in newborns or LBW infants requiring IPT due to maternal TB. INH is primarily metabolized through acetylation in the liver and intestines, and the acetylation rate genetically determined(8). Developmental and physiological changes in the volume of distribution, maturity of liver enzymes, and the role of acetylation capacity may influence INH dosage requirements in this vulnerable population(9, 10). The aim of this study was to determine the pharmacokinetics of routinely administered INH at 10 mg/kg/day in TB-exposed LBW infants. The World Health Organization (WHO) recommends a dosage range of 10 to 15 mg/kg daily. We also describe the influence of N-acetyltransferase 2 (NAT2) genotype on INH pharmacokinetics.
Materials and Methods

Design and setting

An intensive sampling pharmacokinetic study was conducted at Tygerberg Children’s Hospital (TCH), Cape Town, South Africa, between May 2011 and May 2012. TCH is a tertiary referral hospital for 50 000 deliveries per annum. Here, the peak TB notification rate amongst young adults exceeded 1 400 per 100 000 population in 2009(11) and the provincial maternal HIV prevalence was 16.9% among public antenatal clinic attendees in 2009(12). TCH manages an average of 6 000 high-risk, complicated deliveries per year, 24% of which are LBW infants. The neonatal service has 136 neonatal beds and includes intensive- and high-care facilities.

Study procedures and drug administration

LBW infants born to HIV-infected and HIV-uninfected women were consecutively recruited if routinely receiving daily INH, either for 6 months IPT (10 mg/kg/day), or as in TB treatment, as per local guidelines(13). Treatment for TB disease consisted of a two-month intensive phase and comprised of daily INH, rifampicin (RMP) and, pyrazinamide (PZA), with or without ethionamide (ETH), followed by a four-month continuation phase of INH and RMP alone. MDR-TB preventive therapy (consisting of daily INH, ETH and ofloxacin) was administered in consultation with a paediatric infectious diseases specialist. Eligibility criteria included: weight ≥ 1.2 kg on the day of pharmacokinetic
sampling; being clinically stable in room air and tolerating oral preparations; and written
informed consent from the mother/legal guardian. Maternal HIV infection status was
routinely determined by an enzyme-linked immunosorbent assay, preceded by informed
consent and appropriate pre- and post-test counseling. All HIV-exposed newborns (born
to HIV-infected mothers) received nevirapine (NVP) for 6 weeks post-partum or until
cessation of breastfeeding, according to national prevention of maternal-to-child
transmission (PMTCT) guidelines(14). Exclusive breastfeeding was encouraged.

Data on the following characteristics were collected: gender, ethnicity, birth weight,
gestational age, weight and age at pharmacokinetic sampling, feeding type (breastfeeding
versus formula), HIV exposure and concomitant medications. The gestational age was
determined by date of last menstrual period, early ultrasound findings and/or a Ballard
score performed by a single experienced neonatologist. Data on alanine aminotransferase
(ALT), a proxy for drug-related hepatotoxicity, were collected at baseline (at
pharmacokinetic sampling) and at 3- and 6-month follow-up visits. The Division of
Microbiology and Infectious diseases (DMID) tables were used to grade potential
hepatotoxicity(15). Infants required regular feeding (2-3 hourly), as per routine care. INH
in powder form, obtained from Fluka Chemie AG (Buchs, Switzerland), was used on the
pharmacokinetic sampling days. The INH powder was accurately weighed to administer a
dose of 10 mg/kg according to the naked newborn’s weight (weighed by the study nurse
on the day prior to study drug administration). The INH powder was dissolved in 1-2 ml
of sterile water, administered through a nasogastric tube and flushed with 1 ml of water.
Pharmacokinetic sampling and laboratory analysis

On the day of pharmacokinetic assessment, the INH dose, time of administration and phlebotomy were documented precisely. Four arterial blood specimens of 0.5 ml each were taken at 2, 3, 4 and 5 hours post-dose and collected in ethylenediaminetetraacetic acid (EDTA) coated tubes. Specimens were kept on ice, and delivered to the laboratory within 30 minutes. After centrifugation, separation plasma was assayed using the high-performance liquid chromatographic (HPLC)-method as previously described (16). The remaining blood cells were used for the extraction of DNA for NAT2 genotyping.

Genomic DNA (gDNA) was prepared with a simple salting-out procedure for extracting DNA from human nucleated cells (17). The gDNA was analyzed for the NAT2*5, 2*6, 2*7, 2*12, 2*13 and 2*14 alleles, via a polymerase chain reaction (PCR)-based strategy (18). Separate PCR aliquots were restricted with the MspI, FokI, KpnI, TaqI, Ddel and BamHI restriction enzymes to delineate the polymorphisms at nucleotide positions 191, 282, 481, 590, 803 and 857, respectively. According to Vatsis nomenclature, the wild type fast allele (F) was assigned as NAT2*4, 2*12 or 2*13 (19). These alleles confer normal enzyme activity on the NAT2 protein, whilst the mutant slow alleles (S), classified as NAT2*5, 2*6, 2*7 and 2*14 in humans, confer a decreased enzyme activity on the NAT2 protein. Accordingly, study participants were classified as homozygous fast (FF), heterozygous intermediate (FS) or homozygous slow (SS) acetylators, depending on the allele combination observed.

Pharmacokinetic parameters and statistical analysis
The following pharmacokinetic parameters were generated using fixed sampling times for each patient through noncompartmental analysis (NCA): $C_{\text{max}}$ (the maximum drug concentration observed), $T_{\text{max}}$ (the time after drug administration to reach $C_{\text{max}}$), the $AUC_{2-5}$ (area under the time concentration curve from 2-5 hours), $t_{1/2}$ (the plasma half-life), and the $k_e$ (elimination constant). The $AUC$ was calculated according to the linear trapezoidal rule. Pharmacokinetic parameters were summarized using medians and interquartile ranges (IQR), except for $T_{\text{max}}$, which was summarized using means and standard deviations (SD). Pharmacokinetic parameters were compared by $NAT2$ genotype, birth weight, weight at the time of pharmacokinetic sampling, gestational age, age at sampling, gender, HIV exposure and feeding type. All covariates were analyzed dichotomously except for $NAT2$, which was analyzed categorically into three levels: FF (fast), FS (intermediate) and SS (slow) acetylator types. Since sample sizes were small, dichotomous covariates were analyzed using the Wilcoxon rank sum test. $NAT2$ was analyzed using the Kruskal Wallis test and, if statistically significant, the trend test was used to assess for trend across ordered $NAT2$ groups. Dichotomous confounders, weight at pharmacokinetic sampling and feeding type were assessed using the Fisher’s Exact test. Any $p$-value $<$0.05 was considered statistically significant. All data was analyzed using Stata™12.1 Special Edition software (StataCorp; 2011. College Station, TX USA).

Study approval was obtained from the Health Research Ethics Committee at Stellenbosch University (N10/07/232).
Results

Twenty infants, 16 (80%) HIV-exposed were enrolled; 17 received IPT and 3 treatment for TB. Ten infants (50%) had a birth weight below 1500 grams, and 17 (85%) were premature (table 1). On the day of pharmacokinetic sampling, the median weight was 1874 (interquartile range: 1361 - 2120) grams. Eight (40%) infants received theophylline for apnea of prematurity and 15 (75%) of the 16 HIV-exposed infants received NVP for PMTCT (One HIV-infected infant received abacavir, lopinavir/ritonavir, and lamivudine).

Pharmacokinetic measures are shown in table 2. $C_{\text{max}}$, $k_e$ and consequently the $t_{1/2}$, could not be accurately determined for one infant, in whom INH plasma concentrations remained high (a 2-hour value of 4.6 $\mu$g/ml, and a 5-hour value of 4.9 $\mu$g/ml, respectively). The $t_{1/2}$ ranged from 1.45 to 14.25 hours. Figure 1 illustrates the INH plasma concentrations at 2, 3, 4 and 5 hours post dose (n=20). Nineteen infants achieved plasma concentrations above the reference of 3 $\mu$g/ml at 2 hours(20); range 2.9 – 10.7 $\mu$g/ml, while all 20 infants achieved target drug plasma concentrations $> 1.5 \mu$g/ml at 3 hours post dose(21).

Pharmacokinetic parameters were compared by demographic and clinical covariates (table 3). Distribution of acetylation status was as follows: 5 were slow, 11 were intermediate and 4 were fast acetylators. Statistically significant differences in $C_{\text{max}}$, $AUC_{2-5}$, $t_{1/2}$ and $k_e$ were shown (p-values of 0.024; 0.020; 0.013 and 0.013, respectively)
between slow-, intermediate- and fast acetylators. Slow acetylators had a higher median 
$C_{\text{max}}$ (6.5 ug/ml), larger AUC$_{2-5}$ (16.93 $\mu$g.h/ml) and longer $t_{1/2}$ (6.56 hours) compared to 
intermediate and fast acetylators, in decreasing order. Figure 2 illustrates INH 
concentrations in relation to NAT2 genotype.

Infants below 1750 grams on the day of pharmacokinetic sampling had a higher $C_{\text{max}}$ and 
AUC$_{2-5}$ compared to heavier infants (7.66 $\mu$g/ml vs. 5.03 $\mu$g/ml; $p=0.030$ and 21.26 
$\mu$g.h/ml vs. 13.19 $\mu$g.h/ml; $p=0.044$, respectively). Figure 3 suggesting increased INH 
absorption and reduced clearance in smaller infants. Similar findings were observed in 
infants with lower gestational age; however, these differences were not statistically 
significant. Feeding also affected $C_{\text{max}}$ and AUC$_{2-5}$; exclusively breastfed infants had a 
higher $C_{\text{max}}$ and AUC$_{2-5}$ compared to formula fed infants (7.66 $\mu$g/ml vs. 4.94 $\mu$g/ml; 
$p=0.003$ and 21.26 $\mu$g.h/ml vs. 12.03 $\mu$g.h/ml; $p=0.014$, respectively). Since weight at 
pharmacokinetic sampling was associated with feeding status ($p=0.001$), feeding and 
weight were likely confounded; smaller babies being more likely to receive breast milk. 
Gender did not influence pharmacokinetic parameters.

ALT levels were determined in 19 (95%) infants at baseline, in 14 (70%) at 3 months, 
and in 11 (55%) at 6 months. Two 3-month values were abnormal; 1 was mildly elevated 
(<2.5 times elevated; DMID grade 1) and one moderately elevated (<5 times elevated; 
DMID grade 2). The mildly elevated value was not repeated, and the moderately elevated 
ALT value normalized at six months. Both raised ALT values occurred in HIV-exposed 
infants receiving IPT and NVP.
Discussion

This is the first study describing the pharmacokinetics of INH and correlating with \textit{NAT2} genotypes in LBW- and premature infants. INH plasma concentrations in LBW-infants, administered at a dose of 10 mg/kg, compared well to published target adult values\textsuperscript{(20-22)}. An increased, although variable half-life was observed in LBW-infants, cautioning against higher INH dosing strategies. Marked reduced clearance was present in smaller infants and in slow acetylators. The most important pathway for INH metabolism in humans is dependant on the trimodal \textit{NAT2} acetylation\textsuperscript{(18)} already apparent in this young age group.

Optimal and safe INH dosing for TB prevention and treatment is especially relevant for LBW-infants. We administered INH at the lower end of the WHO recommended dosage of 10-15 mg/kg\textsuperscript{(23)}. Desirable pharmacokinetic targets for children include either a 2-hour plasma concentration of 3-5 µg/ml\textsuperscript{(20)}, or a 3-hour value of >1.5 µg/ml\textsuperscript{(21)}, both correlating with good clinical response in adults. We found good absorption of INH in all infants and adult pharmacokinetic target values were achieved. All but one infant achieved a 2-hour drug plasma concentration of ≥3 µg/ml. For the latter, the 2-hour value was 2.9 µg/ml. All infants achieved a 3-hour value of ≥1.5 µg/ml. The effect of routine 2-3 hourly feeding of LBW-infants did not influence the \textit{C}_{\text{max}}. However, food intake is known to decrease the bioavailability of INH\textsuperscript{(24)} and we were unable to evaluate pharmacokinetics in the absence of feeding. Concomitant medicines frequently used in
LBW-infants included theophylline and NVP, neither of which should impact on pharmacokinetic parameters of INH (25, 26).

In our study, we observed an increased, but variable half-life (1.45-14.25 hours) using 10 mg/kg of INH, in line with an earlier study of two neonates, in whom the half-lives were 7.8 and 19.8 hours, respectively (27). This is not surprising, since most drugs in newborns have a prolonged elimination half-life in general. This finding however cautions against using a high dose of INH in small infants. Furthermore, a study describing the INH pharmacokinetics according to phenotype, performed in 34 children (two infants < 1 month of age), showed a definite decrease of half-life with increasing age, suggesting slower elimination of INH in young children (28). Although desirable INH pharmacokinetic targets are essential for efficacy in infants, caution should be applied when dosing at the higher range to prevent potential toxicity.

Markedly reduced INH clearance was noted in the smaller and younger LBW-infants in the study. INH has a significant first-pass metabolism, with the rate of drug metabolism depending largely on the maturation of hepatic enzymes. Impaired elimination in the smaller and younger LBW-infants is therefore probably due to immature hepatic enzymes. The development of drug metabolizing enzymes vary widely between neonates and may be prolonged in premature infants (29). Pharmacokinetic studies show that the grade of maturation of enzymes is the most important factor in determining the rate of metabolism of a drug, with most liver enzymes maturing after the first year of life (30, 31). The effect of reduced INH elimination was even more pronounced in breastfeeding.
infants. However, this was confounded by the fact that smaller infants were more likely to receive breast milk. Anti-tuberculosis drugs, including INH, are secreted in the breast milk of women on TB treatment, ranging between 0.05-28% (32). Levels of anti-tuberculosis drugs in breast milk are inadequate to prevent or treat infants, but may increase the exposure to these medications. Therefore, Tran et al., recommend dosing at the lower end of the therapeutic range (i.e. 10 mg/kg/day of INH), to decrease the risk of potential toxicity (32).

Age and acetylator status influence INH pharmacokinetics in children (28, 33). INH is acetylated to acetylisoniazid by a hepatic and intestinal enzyme N-acetyltransferase 2, which is coded for genetically. In our study, trimodal clearance of INH as a function of NAT2 genotype was already apparent, even in this young age group. Our results showed that all compared parameters were significantly different for all three acetylator groups, slow acetylators having decreased elimination compared to intermediate and fast acetylators. Recent data from an IPT trial conducted in children aged 3 – 24 months, illustrated not only the difference for each genotype group, but also immature NAT2 activity, particularly in fast acetylators with acquisition of activity to adult values occurring over the first 2 years of life (34). This is in keeping with the maturation of genetically determined NAT2 activity over time. The impact of enzyme maturity on the INH dosing for LBW-infants requires further studying.

Transient elevation of serum transaminases occurs commonly with INH, but clinically manifested hepatotoxicity is rare. No formal relationship has been demonstrated between
INH plasma concentrations and hepatotoxicity. Previous observations indicate that hepatotoxicity may be dose-related (35, 36). In our study only two ALT results were slightly raised at month 3, with the thrice-elevated value returning to normal at month 6. The limited safety data collected was reassuring and no jaundice was observed in any infant. A limitation of the study was the few pharmacokinetic sampling points, mainly because of the small blood volume available in LBW-infants. An earlier time point may have assisted with determining the $C_{\text{max}}$ more accurately, as previous pharmacokinetic studies in children indicate that the $T_{\text{max}}$ occurs any time between 1 and 2 hours (28, 37).

In conclusion, a sound understanding of the pharmacokinetic properties of currently used anti-tuberculosis drugs is essential for optimal use in newborns and infants. LBW-infants receiving 10 mg/kg of INH had desirable drug concentrations, comparable to adult target values. However, a prolonged half-life and reduced elimination of INH was noted in smaller and younger infants, especially in genetically determined slow acetylators. Therefore, we caution against exceeding a dosage of 10 mg/kg in this population.

Although no serious adverse effects were observed, more data on safety is needed. More research is needed on appropriate dosing requirements for TB drugs in newborns and infants.

**Acknowledgements**

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The authors thank the study team, specifically Ms E Playandi, for their assistance. Lastly, we thank the parents who permitted us to include their infants in the study.
References


<table>
<thead>
<tr>
<th>Clinical characteristics</th>
<th>N = 20 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (Male)</td>
<td>14 (70)</td>
</tr>
<tr>
<td>Ethnicity (Black)</td>
<td>12 (60)@</td>
</tr>
<tr>
<td>HIV-exposed</td>
<td>16 (80)</td>
</tr>
<tr>
<td>Exclusive breastfeeding</td>
<td>9 (45)</td>
</tr>
<tr>
<td>Formula feeding</td>
<td>11 (55)</td>
</tr>
<tr>
<td>Birth weight in grams, median [IQR]</td>
<td>1575 [1190-2035]</td>
</tr>
<tr>
<td>- &lt; 2500 grams &amp; &gt;1500 grams</td>
<td>10 (50)</td>
</tr>
<tr>
<td>- &lt; 1500 grams</td>
<td>10 (50)</td>
</tr>
<tr>
<td>Gestational age in weeks, median [IQR]</td>
<td>35 [34-38]</td>
</tr>
<tr>
<td>- Term (≥38 weeks)</td>
<td>3 (15)</td>
</tr>
<tr>
<td>- Premature (&lt;38 weeks)</td>
<td>17 (85)</td>
</tr>
<tr>
<td>Weight on day of PK sampling in grams, median [IQR]</td>
<td>1874 [1361-2120]</td>
</tr>
<tr>
<td>Age in days at time of PK sampling, median [IQR]</td>
<td>14 [9-31]</td>
</tr>
<tr>
<td>Concomitant medications ^</td>
<td></td>
</tr>
<tr>
<td>- Nevirapine</td>
<td>15 (75)§</td>
</tr>
<tr>
<td>- Theophylline</td>
<td>8 (40)</td>
</tr>
<tr>
<td>- Combination anti-TB drugs (RMP, PZA, ETH, ofloxacin)</td>
<td>4 (20) *</td>
</tr>
</tbody>
</table>
- cART (abacavir, lopinavir/ritonavir, lamivudine) and co-
trimoxazole | 1 (5)*
- Hydrochlorothiazide and spirinolactone | 1 (5)*

§ Unless indicated otherwise
@ 7 of mixed race, 1 white infant
^ Infants could be on a different combination of listed concomitant medication
‡ 2 infants on NVP and INH had a raised ALT at 3 months
* Infants on RMP, PZA (n=2); RMP, PZA and ETH (n=1); Ofloxacin and ETH (n=1)
§ One infant, day 59 of life, was HIV-infected and treated with cART and co-trimoxazole
* To control mild cardiac failure in infant with a ventricular septal defect

HIV=human immunodeficiency virus; IQR=interquartile range; NVP= Nevirapine;
INH=isoniazid; ALT=Alanine aminotransferase; RMP=Rifampicin; PZA=Pyrazinamide; ETH=
Ethionamide; cART= combination anti-retroviral therapy
Table 2. Summary of pharmacokinetic parameters in low birth weight infants (n=20)

<table>
<thead>
<tr>
<th>Pharmacokinetic parameters</th>
<th>Median (IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$C_{\text{max}}$ (μg/ml)</td>
<td>5.63 (4.86 - 7.53)</td>
</tr>
<tr>
<td>$T_{\text{max}}$ (h)</td>
<td>2 (2 - 2)</td>
</tr>
<tr>
<td>AUC$_{2-5}$ (μg.h/ml)</td>
<td>13.56 (11.75 - 19.10)</td>
</tr>
<tr>
<td>$k_e$ ( h$^{-1}$) $^\wedge$</td>
<td>0.15 (0.10 - 0.23)</td>
</tr>
<tr>
<td>$t_{1/2}$ (h) $^\wedge$</td>
<td>4.69 (3.08 - 7.60)</td>
</tr>
</tbody>
</table>

$^\wedge$ $k_e$ and $t_{1/2}$ not calculated for 1 patient due to continuous high drug plasma concentrations

IQR=interquartile range; $C_{\text{max}}$= maximum drug concentration; $T_{\text{max}}$=time to $C_{\text{max}}$; h= hours; AUC$_{2-5}$ = area under the concentration time curve; $k_e$= first order elimination rate constant; $t_{1/2}$ = half-life
Table 3. Effect of clinical and other characteristics on isoniazid pharmacokinetic parameters in low birth weight infants (n = 20)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>C&lt;sub&gt;max&lt;/sub&gt; (µg/ml)</th>
<th>AUC&lt;sub&gt;2-5&lt;/sub&gt; (µg·h/ml)</th>
<th>t&lt;sub&gt;1/2&lt;/sub&gt; (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N Median(IQR)</td>
<td>N Median(IQR)</td>
<td>N Median(IQR)</td>
</tr>
<tr>
<td><strong>NAT2 genotyping</strong></td>
<td></td>
<td>p-value</td>
<td>p-value</td>
</tr>
<tr>
<td>SS- Slow</td>
<td>5 6.54 (5.60 - 8.05)</td>
<td>5 16.93 (14.59 - 21.87)</td>
<td>5 6.56 (4.86 - 9.57)</td>
</tr>
<tr>
<td>FS- Intermediate</td>
<td>11 6.41 (5.03 - 7.66)</td>
<td>11 13.21 (12.03 - 21.26)</td>
<td>11 4.52 (3.48 - 6.27)</td>
</tr>
<tr>
<td>FF- Fast</td>
<td>4 3.90 (2.95 - 4.86)</td>
<td>4 6.78 (4.96 - 11.31)</td>
<td>3 1.78 (1.45 - 2.04)</td>
</tr>
<tr>
<td><strong>Birthweight</strong></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Very low birthweight</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(&lt;1500 g)</td>
<td>10 6.58 (5.60 - 8.05)</td>
<td>10 15.35 (13.21 - 21.62)</td>
<td>10 4.56 (3.74 - 6.27)</td>
</tr>
<tr>
<td>Low birthweight</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(≥1500 - &lt;2500 g)</td>
<td>10 4.99 (4.11 - 6.41)</td>
<td>10 12.61 (8.14 - 14.59)</td>
<td>9 4.52 (1.95 - 6.56)</td>
</tr>
<tr>
<td><strong>Weight at PK time</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;1750 g</td>
<td>9 7.66 (5.60 - 8.05)</td>
<td>9 21.26 (13.21 - 21.62)</td>
<td>9 3.93 (4.12 - 9.15)</td>
</tr>
<tr>
<td>Gestational age in weeks</td>
<td>≥1750 g</td>
<td>Gestational age in weeks</td>
<td>≥38 weeks</td>
</tr>
<tr>
<td>--------------------------</td>
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</tr>
<tr>
<td></td>
<td>11</td>
<td>10</td>
<td>9</td>
</tr>
<tr>
<td></td>
<td>5.03 (4.11 - 6.54)</td>
<td>13.19 (8.14 - 14.59)</td>
<td>3.60 (1.95 - 5.09)</td>
</tr>
<tr>
<td>&lt;38 weeks</td>
<td>17</td>
<td>16</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>5.66 (5.03 - 7.66)</td>
<td>14.48 (13.19 - 21.26)</td>
<td>4.69 (3.61 - 6.42)</td>
</tr>
<tr>
<td>≥38 weeks</td>
<td>3</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>4.11 (3.02 - 6.41)</td>
<td>11.46 (5.41 - 12.03)</td>
<td>2.04 (1.95 - 8.64)</td>
</tr>
</tbody>
</table>

**Gender**

- Female: 6
  - 5.14 (3.02 - 8.05)
  - 14.54 (5.41 - 21.62)
  - 6.27 (2.04 - 6.56)

- Male: 14
  - 6.04 (5.03 - 7.40)
  - 13.27 (12.03 - 16.93)
  - 4.39 (3.48 - 5.93)
  - 0.4579
  - 0.1837
  - 0.1837
  - 0.8690
  - 0.7812

**HIV status**

- Exposed: 16
  - 5.45 (4.45 - 6.97)
  - 13.49 (10.19 - 16.91)
  - 4.52 (2.04 - 6.27)

- Negative: 4
  - 7.34 (6.14 - 9.39)
  - 17.60 (13.27 - 25.08)
  - 6.53 (3.08 - 11.91)
  - 0.0472
  - 0.0472
  - 0.1306
  - 0.4237

**Feeding**

- Breastfeeding: 9
  - 7.66 (5.66 - 8.05)
  - 5.93 (4.12 - 9.15)
| Formula | 11 | 4.94 (3.35 - 6.41) | 0.0027 | 11 | 12.03 (8.14 - 14.59) | 0.0135 | 10 | 4.13 (1.95 - 5.09) | 0.1208 |

* $t_{1/2}$ not calculated for 1 patient due to continuous high drug plasma concentrations

$C_{\text{max}}$ = maximum drug concentration; $\text{AUC}_{2-5}$ = area under the concentration time curve; $t_{1/2}$ = half-life; IQR = interquartile range;
Figure 1: Individual isoniazid drug concentrations in low birth weight infants. (n=20)
Figure 2: Isoniazid concentrations in relation to NAT-2 genotyping (n=20)
Figure 3: Isoniazid concentrations relative to current weight in low birth weight infants (n=20)