Drug Excretion into Breast Milk: Are all Drugs Contraindicated for Breastfeeding?

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INTRODUCTION

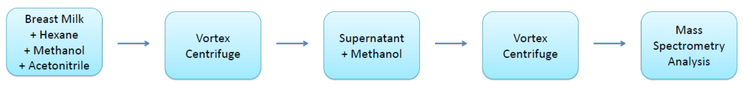
Current epidemiological research provides strong evidence for health benefits associated with breastfeeding, including reductions in infant mortality, reduced chances of infection or the development of chronic diseases, and positive impacts on cognitive development.[1-3] Current Centre for Disease Control Breastfeeding Report Card data demonstrate more women (~ 80%) are breastfeeding and for longer.[4] However, studies have also shown that 66 - 80% of women are on some kind of medication during the postpartum period.[5] Consequently, this leads to an increased likelihood of infant drug exposure through breast milk. Although the dosages are considered to be small, there have been reports of adverse events and even fatality in infants exposed to drugs through breast milk. Although not all drugs may be contraindicated while breastfeeding, there remains little data on this topic.

Methotrexate (MTX) is the first line of treatment for rheumatoid arthritis (RA), which has a high incidence in women of childbearing age. Disease activity of RA often decreases during pregnancy but tends to flare following delivery, necessitating the reestablishment of treatment. Only one case report on the excretion of MTX into breast milk has been published. Due to the paucity of data on MTX levels in breast milk, the potential risk of toxicity and drug accumulation in the infant remains largely unknown. We have developed a sensitive and specific LC-MS/MS method to quantitate MTX and its metabolite in human milk and applied it to patient samples. We have also calculated the relative infant dose of MTX to determine the risk to the infant. The objective of this study is to investigate the risk of drug exposure in nursing infants by determining drug concentrations in breast milk, with an aim to developing pharmacokinetic profiles for drugs excreted into breast milk.

METHOD

Sample Preparation

To investigate infant drug exposure through breast milk, we established a drug safety monitoring program, Drugs in Lactation Analysis Consortium (DLAC), to measure several drugs commonly used by women breastfeeding, beginning with methotrexate. Breast milk is a complex lipid and protein-rich matrix, with drugs partitioning to either the aqueous or lipid phase, thus requiring meticulous sample processing before analysis. Differences in the lipid and protein composition of milk exists between fore and hind milk. We first investigated whether drugs partition to the aqueous or the lipid phase of breast milk. We then worked to create a simplified drug extraction method using hexane, methanol and acetonitrile to facilitate efficient drug extraction from breast milk, as shown below.



Then 5µL of supernatant were loaded on an analytical column and eluted with an LC cycle time of 3.5 minutes. The signal is detected by an IONICS 3Q 220 triple quadrupole mass spectrometer. All solvents are HPLC grade.

Instruments

A Shimadzu Prominence UFLCxr system was coupled to the IONICS 3Q 220 mass spectrometer. The detailed LC conditions are shown below and the time program is listed in Table 1.

Column:	Imtakt Unison UK-C8, 75x3mm, 3µm
Flow Rate:	0.6 mL/min
Injection Volume:	10 µL
Column Temperature:	30◦C

Time (min)	Solvent B %
0.3	20
1.8	95
2.0	95
2.1	10
3.5	10

Table1: LC gradient, 3.5 min total runtime

RESULTS

Calibration Curves

a) Methotrexate b) 7OH-Methotrexate

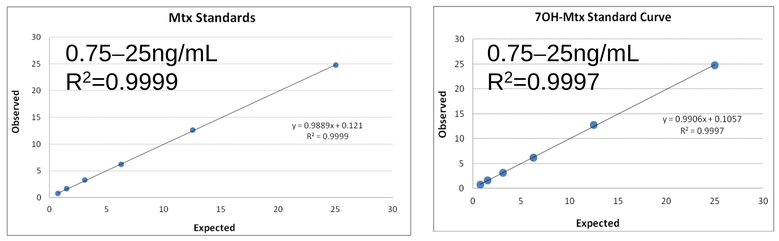


Fig.1a–b. Calibration curves for (a) Methotrexate and (b) 7OH-Methotrexate in breast milk .

Good linearity (with $R^2 > 0.999$) was obtained for methotrexate and its metabolite 7OH-methotrexate. Multiple replicates (8 in several runs) were performed. Examples of the calibration curves generated for methotrexate and 7OH-methotrexate are shown in Figures 1a–b, respectively. Excellent linearity was obtained for a concentration range of 0.75 to 25ng/mL for methotrexate and 7OH-methotrexate. The accuracy obtained is between 90.0% to 106.3% and the corresponding CV% is less than 11.3% within a run.

A between day precision study was also carried out using 3 levels of a QC sample injected multiple times over multiple days. The mean, standard deviation and CV were determined. The results in Table 2 (below) show that for QC levels 2 & 3, the CV% is less than 6%.

Methotrexate	Mean (ng/mL)	Stdv	% CV
QC Level 1	1.03	0.24	23.6
QC Level 2	7.49	0.40	5.3
QC Level 3	16.77	0.33	2.0

7-OH Methotrexate	Mean (ng/mL)	Stdv	% CV
QC Level 1	1.21	0.27	22.3
QC Level 2	8.20	0.37	4.5
QC Level 3	17.62	0.50	2.8

Table 2a–b: Mean, standard deviation, and CV for three levels of QC for methotrexate and 7OH-methotrexate.

Method Comparison

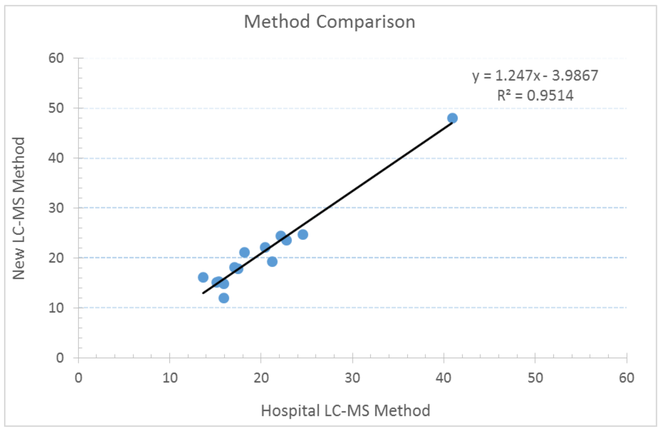


Fig.2. Method comparison between one sample preparation for serum and the method developed for breast milk in this study.

Fourteen serum samples from patients on methotrexate, who had drug concentrations determined by another LC-MS method, were also analyzed using the method developed for this study. The plot in Figure 2 shows good agreement between methods with an R^2-value of > 0.95 and a percent difference between results of < 20%.

Pharmacokinetic profiling

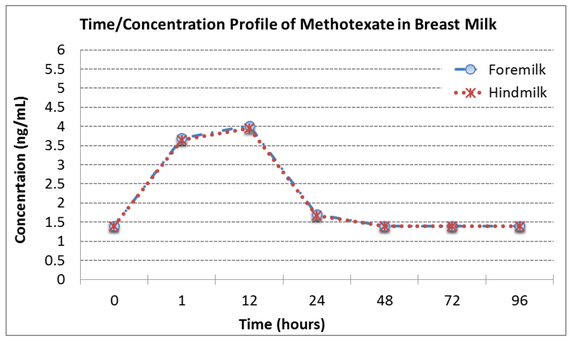


Fig.3. Pharmacokinetic profiling of methotrexate in breast milk following a subcutaneous dose of 25 mg/mL. Both foremilk and hindmilk were measured.

Pharmacokinetic profiling of methotrexate in breast milk following a subcutaneous dose of 25 mg/mL was examined. Both foremilk and hindmilk were analyzed. As illustrated in the Figure 3, methotrexate concentration in breast milk peaks between 1 – 12 hours post dose and quickly decreases within 24 hours post-dose. By 48 hours post-dose, the concentrations excreted in breast milk are negligible. Interestingly, methotrexate is excreted into breast milk with no notable differences in drug concentrations between foremilk and hindmilk. These data provide the foundation to establish a TDM system for measuring drug concentrations in breast milk. We hope to carry out population-based pharmacokinetic analysis to establish safety guidelines on drug excretion into breast milk.

CONCLUSION

Good linearity and reproducibility were observed for methotrexate and its metabolite 7OH-methotrexate in both serum and breast milk. The results indicate that this ESI-LC-MS/MS method using an IONICS 3Q 220 mass spectrometer providing the best ionization and sampling efficiencies is robust and ideal for this clinical research study.

This study highlights the issue of infant risk for toxicity though drug exposure in breast milk. Methotrexate is excreted into breast milk at significant concentrations within the first 24 hours post-dose. However, no notable differences in drug concentrations between foremilk and hindmilk were observed. Due to the difficulty in obtaining foremilk and hindmilk, this is the first study to measure and compare drug levels in this sample type. These data provide the foundation to establish a TDM system for measuring drug concentrations in breast milk, with the aim to carry out population-based pharmacokinetic analysis to establish safety guidelines regarding drug excretion into breast milk as well as breast feeding guidelines.

References:

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