FATTY ACID ETHYL ESTERS IN MECONIUM: AN EMERGING BIOMARKER FOR *IN UTERO* ALCOHOL EXPOSURE

Joey Gareri, Graduate Student, Department of Pharmacology, University of Toronto

A CRITICAL REVIEW of "Agreement between maternal self-reported ethanol intake and tobacco use during pregnancy and meconium assays for fatty acid ethyl esters and cotinine." *Derauf et al. American Journal of Epidemiology* 2003;158(7):705-709.

Fetal Alcohol Spectrum Disorder afflicts approximately 1% of children born in North America[1]. This condition, encompassing a myriad of behavioural and developmental abnormalities, can be very difficult to diagnose in the absence of maternal drinking history[2]. This suggests the need for a reliable biological marker that is capable of confirming heavy exposure to alcohol *in utero*. It is estimated that up to 20% of women consume alcohol regularly while pregnant[3,4] and the prevalence of heavy drinking (>14 drinks/week) among pregnant women is estimated to be 0.1%-0.3% [5].

Fatty acid ethyl esters (FAEE) are non-oxidative ethanol metabolites found in the meconium (i.e. first stool) of neonates[6], as well as other organs and tissues of adult drinkers[7]. The presence of certain FAEE in meconium has been shown to represent fetal ethanol metabolism[8] during the second and third trimesters of pregnancy[9,10,11]. Several laboratories are currently developing FAEE meconium analysis as an objective test for prenatal alcohol exposure. The study by Derauf *et al* attempts to analyze the concordance between maternal self-reporting and meconium assays for FAEE and cotinine as biomarkers for maternal drinking and tobacco use.

This study was carried out at a large urban regional perinatal center in Hawaii. Pregnant mothers were questioned by a triage nurse in a standardized manner regarding their alcohol and tobacco use during pregnancy. Meconium was collected from each neonate and shipped to the U.S. Drug Testing Laboratories (USDTL) in Des Plaines, Illinois. Total and individual FAEE content were measured by gas chromatography-mass spectrometry.

The results showed moderate agreement between reported and measured tobacco use and no agreement between reported and measured alcohol intake in the third trimester. The authors concluded that the FAEE meconium test does not reflect maternal alcohol ingestion and requires additional refinement and validation for use as a biomarker.

In their analysis, the authors did not specify at what stage of pregnancy the mothers were interviewed. The fact that interviews were conducted by a triage nurse implies that the mothers were being admitted for delivery at the time. In addition to the fact that underreporting by pregnant alcohol-users is well documented[12], the interview itself was carried out by an unfamiliar individual in a high anxiety setting. These factors could serve to undermine attempts by the authors to provide an interview process conducive to minimizing false reports. It is highly unlikely, especially given this situation, that chronic problem drinkers would produce a truthful report.

The maternal histories obtained were documented by trimester of last use and drinks per day. Unfortunately, the ensuing meconium test results were only compared against third trimester use. It is known that the fetus begins producing meconium between 12-13 weeks of gestation; hence meconium analysis is capable of documenting fetal exposures in the second trimester as well as the third[13]. This dictates that both second and third trimester use should have been included in any comparisons between maternal reporting and meconium analysis.

In addition to the problems associated with the application of the maternal reports, there are

issues with the authors' definition of a positive test. The positive cut-off used in this study was 50ng cumulative FAEE per gram of meconium. Abstracts published by Chan *et al* as early as 2001 show preliminary findings of baseline cumulative FAEE levels in the meconium of certain neonates born to confirmed non-drinkers that far exceed this cut-off level[14]. In addition, the final results of this baseline study determined a positive cut-off of ~520ng/g as an indicator of heavy maternal drinking with high sensitivity and specificity[15].

Conversely, the USDTL team that carried out the analysis for this study found that 18/72 positive specimens produced an average FAEE level of 62,115ng/g[16]. They concluded that *significant amounts* of alcohol ingested during pregnancy would result in FAEE measurements of >10 000ng/g. This demonstrates the high likelihood of a subgroup of heavy drinkers in this population that did not report their alcohol use to the authors of this study.

Published data also shows that cumulative FAEE levels in the meconium of neonates born to social drinkers (<14 drinks/wk) are likely to be indistinguishable from abstainers[15]. By these standards, the four mothers who reported very mild alcohol intake in the third trimester could be expected to produce a negative or below-baseline result.

The authors were not mistaken in attributing a certain degree of unreliability to the FAEE meconium test. The test still requires further evaluation related to the dose-response relationship between alcohol ingestion and meconium FAEE levels. A recent study that interviewed problem-drinking women during pregnancy has shown a good dose-response curve with FAEE levels[17]. The authors were correct in remarking that maternal reporting is known to be unreliable, unfortunately, it does not appear that their methodology provided any solution to this problem. Cumulatively, several studies show that the FAEE meconium test requires quantitative interpretation when attempting to determine a neonate's exposure to alcohol *in utero* and can be quite effective in predicting fetal exposure to high levels of alcohol[15,16,17].

J. Gareri is funded by CIHR grant, Hospital for Sick Children, and the University of Toronto.

REFERENCES

1. Sampson PD et al. Incidence of fetal alcohol syndrome and prevalence of alcohol-related neurodevelopmental disorder. *Teratology* 1997;56:317-326.
2. Koren G, Loebstein R, Nulman I. Fetal alcohol syndrome: Role of the family physician. *Journal of FAS International* January, 1998.
3. Alcohol Use Among Women of Childbearing Age—United States, 1991-1999. MMWR Morb Mortal Wkly Rep. 2002;51(13):273-276.
4. Stewart DE, Streiner D. Alcohol drinking in pregnancy. *Gen Hosp Psychiatry* 1994;16(6):406-412.
5. Alcohol Consumption Among Pregnant and Childbearing-Aged Women—United States, 1991 and 1995. MMWR Morb Mortal Wkly Rep 1997;46(16):346-350.
6. Mac E *et al.* A marker of fetal exposure to alcohol by meconium analysis. *Pedriatric Research* 1994;35:238A.
7. Laposata EA, Lange LG. Presence of nonoxidative ethanol metabolism in human organs commonly damaged by ethanol abuse. *Science* 1986;231:497-499.
8. Chan D, Knie B, Koren G. Fatty acid ethyl esters transport across the human placenta. *Placenta* 2003;(24)A.41:P132.
9. Bearer CF, Swick A, Singer LT. FAEE: Biomarker for prenatal alcohol use. *Alcohol Clin Exp Res* 1996;2:139A.
10. Klein J, Karaskov T, Koren G. Fatty acid ethyl esters in meconium: Biomarkers for the detection of alcohol exposure in neonates. *Therapeutic Drug Monitoring* 1999;21:644-646.
11. Moore CM, Lewis D. Fatty acid ethyl esters in meconium: Biomarkers for the detection of alcohol exposure in neonates. *Clin Chim Acta* 2001;312: 235-237.
12. Russell M *et al.* Detecting risk drinking during pregnancy: a comparison of four screening questionnaires. *Am J Public Health* 1996;86:1435-1439.
13. Ostrea EM Jr. Testing for exposure to illicit drugs and other agents in the neonate: a review of laboratory methods and the role of meconium analysis. *Curr Probl Pediatr* 1999;29:37-56.

14. Chan D, Pellerin B, Paciorek C, Klein J, Koren G. Fatty acid ethyl esters: Baseline establishment in meconium. *Alcoholism Clinical and Experimental Research* 2001;25(5):48A.
15. Chan D *et al*. Population baseline of meconium fatty acid ethyl esters among infants of non-drinking women in Jerusalem and Toronto. *Ther Drug Monitor* 2003; 25(3):271-278.
16. Moore CM, Jones J, Lewis D, Buchi K. Prevalence of fatty acid ethyl esters in meconium specimens. *Clinical Chemistry* 2003;49(1):133-136.
17. Bearer C *et al.* Validation of a new biomarker of fetal exposure to alcohol. *J Pediatr* 2003;143(4):463-469.