Meconium Fatty Acid Ethyl Ester, Ethyl Glucuronide, and Ethyl Sulfate Sensitivity and Specificity to Detect Maternal Drinking During Pregnancy

Sarah K. Himes¹, Kimberly A. Dukes², Tara Tripp², Julie Petersen², Cheri Raffo², Larry Burd³, Hein Odendaal⁴, Amy J. Elliott⁵, Dale Hereld⁶, Caroline Signore⁷, Marian Willinger⁷, and Marilyn A. Huestis¹, for the Prenatal Alcohol in SIDS and Stillbirth (PASS) Network

¹Chemistry and Drug Metabolism Section, Intramural Research Program, National Institute on Drug Abuse, National Institutes of Health, Baltimore, MD 21224
²DM-STAT Inc., Malden, MA 02148
³Department of Pediatrics, University of North Dakota School of Medicine, Grand Forks, ND 58203
⁴Department of Obstetrics and Gynecology, Faculty of Medicine and Health Science, Stellenbosch University, Tygerberg, South Africa
⁵Center for Health Outcomes and Prevention Research, Sanford Research, Sioux Falls, SD 57104
⁶Division of Metabolism and Health Effects, National Institute on Alcohol Abuse and Alcoholism, National Institutes of Health, Rockville, MD 20892
⁷Eunice Kennedy Shriver National Institute of Child Health and Human Development, Bethesda, MD 20892

Introduction: We investigated agreement between self-reported prenatal alcohol exposure (PAE) and objective meconium alcohol markers to determine the optimal meconium marker and threshold for identifying PAE. Meconium ethyl glucuronide (EtG) and ethyl sulfate (EtS) quantification may be superior to fatty acid ethyl esters (FAEE) quantification due to improved stability in meconium and lack of maternal olive oil diet bias. More research is needed to validate meconium EtG and EtS cutoffs against reliable self-report measures.

Methods: Nine FAEE, EtG, and EtS were quantified by liquid chromatography-tandem mass spectrometry in 0.1 g meconium from infants of Safe Passage Study participants. Lower limits of quantification were 25-50 ng/g for FAEE, 5 ng/g for EtG, and 2.5 ng/g for EtS (Himes et al., 2014). Sample preparation involved methanolic homogenization and solid phase extraction.
Detailed PAE information was collected from women with a validated timeline follow-back interview four times during pregnancy and once postpartum (Dukes et al., 2014). As meconium formation begins during weeks 12-20 (Burd and Hofer, 2008), maternal self-reported drinking at or beyond 19 weeks was our exposure variable.

Results: Of 107 women, 33 reported no alcohol consumption in pregnancy, 16 stopped drinking by week 19, and 58 drank beyond 19 weeks (including 45 3rd trimester drinkers). There was moderate-substantial agreement between self-reported PAE at or beyond 19 weeks and meconium EtG ≥30 ng/g (kappa: 0.57, 95% CI 0.41-0.73). This biomarker and associated cutoff was superior to a 7 FAEE sum ≥2 nmol/g (the currently recommended cutoff by Chan et al., 2003 and 2004, summing ethyl linolenate, palmitoleate, arachidonate, linoleate, palmitate, oleate, and stearate) and all other individual and combination marker cutoffs. With meconium EtG ≥30 ng/g as the gold standard and maternal self-report at or after 19 weeks gestation as the test condition, 82% sensitivity (95% CI: 71.6-92.0) and 75% specificity (95% CI: 63.2-86.8) were observed. A significant dose-concentration relationship between self-reported drinks per drinking day after gestational week 19 and meconium EtG ≥30 ng/g also was observed (P<0.0001).

Conclusions: We assessed meconium EtG, EtS, and FAEE concentrations in the same meconium sample and compared concentrations to detailed self-reported PAE data. Maternal alcohol consumption ≥19 weeks was better represented by meconium EtG ≥30 ng/g compared to currently utilized FAEE cutoffs. Confirmation of these findings should be explored in other populations and larger samples. We recommend meconium EtG as a better alcohol marker than FAEE for identifying PAE. Our data demonstrate the clinical diagnostic capabilities of mass spectrometry for PAE identification and quantification in a complex matrix such as meconium.

References:

Chan D, Klein J, Karaskov T, Koren G. Fetal exposure to alcohol as evidenced by fatty acid ethyl esters in meconium in the absence of maternal drinking history in pregnancy. Ther Drug Monit 2004;26:474-81.


Himes SK, Concheiro M, Scheidweiler KB, Huestis MA. Validation of a novel method to identify in utero ethanol exposure: simultaneous meconium extraction of fatty acid ethyl esters, ethyl glucuronide, and ethyl sulfate followed by LC-MS/MS quantification. Anal Bioanal Chem 2014;406:1945-55.