



Pregnancy Hormone Metabolite Patterns, Pregnancy Symptoms, and Coffee Consumption

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Because of contradictory reports of pregnancy outcomes and coffee intake, this study was designed to determine how hormone metabolite levels, symptoms, and coffee consumption patterns are related. Eligible subjects were recruited in Cincinnati, Ohio, from 1996 to 1998, aged 18–40 years, and nonsmokers; drank at least 18 ounces (1 ounce = 29.6 ml) of coffee per week (including decaffeinated) at the last menstrual period; and were enrolled by 9 weeks from the last menstrual period. Beverage consumption and pregnancy symptoms were recorded daily. Weekly, first-morning urine samples were collected to assess human chorionic gonadotropin, estrone-3-glucuronide, and pregnanediol-3-glucuronide. A time-dependent, repeated measures analysis was performed to test several associations. Data from 92 subjects were analyzed with the following results. 1) Coffee consumption was significantly, inversely associated with weekly levels of estrone-3-glucuronide and human chorionic gonadotropin. 2) Weekly hours of nausea were significantly, directly associated with human chorionic gonadotropin and inversely with estrone-3-glucuronide and pregnanediol-3-glucuronide. 3) Weekly coffee consumption was significantly associated with vomiting but not with nausea or appetite loss. 4) Weekly levels of pregnanediol-3-glucuronide were 32.2% lower in subjects who drank at least 8 ounces of coffee/day at the last menstrual period, though above what was necessary to maintain those pregnancies. This study shows the significance of these important variables to be considered in future research. *Am J Epidemiol* 2002;156:428–37.

caffeine; coffee; estrogens; gonadotropins, chorionic; nausea; pregnancy; progesterone

Abbreviation: SD, standard deviation.

Because of the high prevalence of caffeine consumption during pregnancy (1) and the contradictory reports of adverse pregnancy outcomes with coffee consumption (2–12), the current study was designed to investigate underlying methodological issues that may explain some of the controversy. Many of these previous studies failed to account for food or beverage aversions and symptoms of pregnancy, such as nausea (3, 5, 6, 9, 12), and of those that did, most did not adjust for the severity of symptoms and/or changes in consumption over time (2, 4, 7, 8, 11). A meta-analysis of previous studies of nausea showed that this symptom is significantly more prevalent in full-term pregnancies than in those ending in spontaneous abortion (13). Three studies have reported a specific aversion to coffee during pregnancy (10, 14, 15). It has been postulated that women having healthier pregnancies may have higher pregnancy hormone levels and more severe pregnancy symptoms. These women

with increased pregnancy symptoms may reduce their coffee consumption because of coffee aversion more than do women who have a weaker pregnancy “signal” (16). Thus, previous findings of an association between increased coffee consumption and pregnancy loss may be explained, in part, by women with less viable pregnancies who have lower levels of hormones and, therefore, fail to experience coffee aversion. The current study collected repeated measures of pregnancy symptoms, coffee consumption, and urinary hormone metabolites to determine how these are related. Four research questions were evaluated (figure 1):

1. Are higher levels of urinary hormone metabolites related to a concurrent decrease in coffee consumption?
2. Are higher levels of urinary hormone metabolites related to a concurrent increase in pregnancy symptoms?

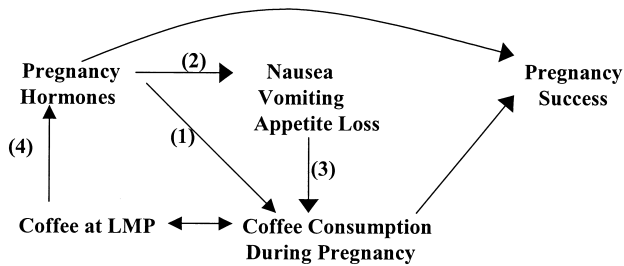


FIGURE 1. Study hypothesis comprising four main analyses: 1, pregnancy hormone metabolites and concurrent coffee consumption; 2, pregnancy hormone metabolites and concurrent pregnancy symptoms; 3, pregnancy symptoms and concurrent coffee consumption; and 4, coffee consumption at the last menstrual period (LMP) and subsequent pregnancy hormone metabolites, Cincinnati, Ohio, 1996–1998.

3. Are increased pregnancy symptoms related to a concurrent decrease in coffee consumption during pregnancy?
4. Is higher coffee consumption at the last menstrual period associated with lower subsequent levels of urinary pregnancy hormone metabolites?

MATERIALS AND METHODS

Study subjects

Subjects were eligible if they were 18–40 years of age, enrolled by the ninth week from the last menstrual period, consumed a minimum of 18 ounces (5.3 dl) per week of coffee at the last menstrual period, and were nonsmokers for at least 4 weeks prior to enrollment. Women with any of the following conditions were ineligible: hyperemesis gravidarum in a previous pregnancy that resulted in hospitalization, chronic spontaneous abortion (more than three consecutive miscarriages), diabetes mellitus, or chronic hypertension, renal disease, thyroid illness, rheumatoid arthritis, lupus, or human immunodeficiency virus positive status. The study proposal was approved by the University of Cincinnati Institutional Review Board.

Subjects in the greater Cincinnati area were recruited through advertising in community papers, fax newsletters, public transportation systems, and flyers/pamphlets at schools, hospitals, doctors' offices, and pharmacies. Advertisements described the study as "a noninvasive pregnancy/beverage study for women in early pregnancy" who did not smoke. A sample size of 110 was targeted for detecting a difference of 18–27 percent in urinary hormone metabolite levels ($\alpha = 0.05$, $\beta = 0.20$) (17, 18). The study was voluntary, and all participants signed an informed consent statement.

Data collection

An in-person, baseline questionnaire was administered at enrollment to assess beverage consumption and pregnancy

symptoms. A daily diary, suitable for direct computer entry, was returned weekly in postage-paid envelopes. On the diary subjects recorded all beverage intake and the timing and severity of nausea, vomiting, and appetite loss. Beverage increases or decreases were assessed on a weekly questionnaire in which subjects listed their reasons for changing consumption. Coffee aversion was then defined as a decrease in consumption due to a physical reaction to the sight, smell, or taste of coffee. A combination of retrospective (from the date of enrollment back to the last menstrual period) and prospective (from enrollment to week 14 from the last menstrual period) data was collected. Study staff checked all questionnaire data for errors by entering and comparing data in duplicate and then by performing logic checks and a manual sight check. Data were collected from September 1996 through February 1998.

Questions designed to assess specific information on beverage consumption included the following: caffeine-containing and decaffeinated beverages, weekday and weekend day consumption, the size of the container (accounting for ice), the method of brewing coffee or tea, and the portion of the container consumed. The following sources of caffeine were included: coffee, tea, sodas, hot chocolate, chocolate snacks, and medications (19). To blind subjects to the purpose of the study, data were also collected on the consumption of other beverages that commonly changes during pregnancy, such as milk and alcohol. At the initial interview, examples of container sizes and pictorial representations of brewing methods were displayed.

Assay analysis

Weekly, first-morning urine samples were collected by study subjects, starting the morning after enrollment and then every Wednesday thereafter, continuing through the 12th week from the last menstrual period. Samples were stored in glass vials (containing 7 percent glycerol for preservation) and then frozen within 1 hour of collection. Assays measured human chorionic gonadotropin, estrone-3-glucuronide, and pregnanediol-3-glucuronide. Estrone-3-glucuronide and pregnanediol-3-glucuronide are downstream urinary metabolites of estrogen and progesterone, respectively. An assay kit developed by Diagnostic Systems Laboratories, Inc. (Webster, Texas), was used to measure human chorionic gonadotropin in urine. Estrone-3-glucuronide and pregnanediol-3-glucuronide were analyzed using an enzyme-linked immunosorbent assay and normalized by creatinine levels to adjust for differences in volume (20). With this method, measurements of estrone-3-glucuronide and pregnanediol-3-glucuronide in early morning urine samples parallel the parent hormones in plasma (21, 22). The minimal detection limits for these measures were 10 ng/ml for estrone-3-glucuronide, 60 ng/ml for pregnanediol-3-glucuronide, 10 mg/dl for creatinine, and 0.5 mIU/ml for intact, serum human chorionic gonadotropin (similar to urinary human chorionic gonadotropin). The intra- and inter-assay variation/precision, as expressed by the coefficient of variation, was 4/13 for estrone-3-glucuronide, 6/12 for pregnanediol-3-glucuronide, 4/3 for creatinine, and 5/7 for human chorionic gonadotropin (intact, serum).

Statistical analysis

Descriptive statistics were generated to characterize the study population, pregnancy symptoms, coffee and caffeine consumption, and hormone metabolite levels and to describe the relations among measurements made on the same subject at consecutive timepoints. Plots of individual values and sample means of weekly coffee consumption and hormone levels were obtained to investigate the dynamics of individual and population trends. Distributions of continuous outcome variables were tested for normality. A parametric approach to describe the relations among human chorionic gonadotropin, pregnanediol-3-glucuronide, estrone-3-glucuronide, coffee/caffeine consumption, nausea, and appetite loss was assumed. Inferences were based on model parameters after variables were scaled appropriately.

Four main analyses corresponded to the four research questions (figure 1), each using a time-dependent, repeated measures analysis. To evaluate if hormone levels were associated with coffee consumption (question 1), we analyzed each hormone metabolite separately as the predictor variable, with weekly coffee consumption (including decaffeinated) as the outcome variable. Because weekly levels of coffee consumption were somewhat dependent on initial consumption, coffee intake at the time of the last menstrual period was a covariate. Coffee consumption patterns might have also been affected by study subjects' reducing intake for perceived health reasons, including advice from a doctor. However, because consumption patterns differed according to coffee aversion status, consumption was considered a valid measure of coffee aversion.

For evaluation of whether hormone levels were related to pregnancy symptoms (question 2), nausea (hours/day), appetite loss (days/week), and vomiting (episodes/week) were regressed separately on each hormone metabolite. For evaluation of whether pregnancy symptoms were related to concurrent coffee consumption (question 3), nausea, appetite loss, and vomiting were regressed separately on weekly coffee consumption. For evaluation of whether coffee consumption at the last menstrual period was associated with subsequent pregnancy hormone levels (question 4), the amount of coffee intake at the last menstrual period was analyzed as the predictor variable, with subsequent weekly hormone metabolites as the outcome.

All analyses used the SAS Institute, Inc. (Cary, North Carolina), procedure, Proc Mixed, in which the subject and time were modeled as random effects, and covariates were considered fixed effects. Hormone values were log transformed to linearize model parameters, and hormone analyses were limited to weeks 6–12. Because coffee and caffeine consumption were highly correlated (the weekly correlation coefficient (r) ranged from 0.31 to 0.73), and because aversion was reported for coffee but not for other caffeine-containing substances, the results shown are primarily for analyses of coffee consumption. Variables were screened for evidence of confounding and effect modification by assessing their association with the dependent and independent variables. The following variables were evaluated for confounding in each analysis by using a backward elimination and by assessing whether removal of each variable

significantly changed the regression coefficients for the independent variables of interest: former smoker, age, first pregnancy, coffee at the last menstrual period, income level, previous miscarriage, race, and education. Variables were tested as categorical and also, where possible, as continuous.

RESULTS

There were 838 subjects screened for eligibility to obtain 114 (13.6 percent) participants. Approximately half of those screened (51.7 percent, $n = 433$) were ineligible because of advanced gestation, 30.8 percent ($n = 258$) were of eligible gestation but were not regular coffee drinkers, and 3.9 percent ($n = 33$) were ineligible for all the other criteria combined. The mean time of enrollment was 6.5 weeks from the last menstrual period (range, 3.6–8.9 weeks). Women whose pregnancies ended in a miscarriage during the study period ($n = 11$) and those with multiple births ($n = 4$) or incomplete data ($n = 7$) were excluded from the analyses.

The final study population of 92 was mostly middle class, well educated, and Caucasian, with an average age of 30.3 years (table 1). Over half (54 percent) had never smoked, and the remaining women were former smokers. Twenty-eight percent were primigravidas, and, of the multigravida group, 26 percent had a prior miscarriage.

The mean consumption of coffee (including decaffeinated) at the last menstrual period was 106.7 (standard deviation (SD), 72.2) ounces/week (31.5 dl) with a range of 18–387 ounces (5.3–114.4 dl). The mean caffeine consumption from coffee alone at the last menstrual period was 872 (SD, 743) mg/week (range, 11–4,018 mg) (23–25). The mean caffeine consumption from all caffeine-containing beverages (coffee, tea, cola, and chocolate milk) was 1,214 (SD, 955) mg/week (range, 17–4,406 mg). Almost all of the study population reported that they had decreased or quit drinking coffee during the first trimester (93 percent, $n = 86$). Moreover, 59 percent ($n = 54$) reported a physical aversion to coffee at least once during the study period. Coffee was second only to meat as the most prominently rejected food or beverage.

There was a high prevalence of first-trimester pregnancy symptoms, with a mean onset between 5 and 6 weeks from the last menstrual period. Nausea was the most prevalent symptom, with 89.5 percent of subjects reporting at least 1 hour during week 8 and 40.2 percent prevalence at week 14. The mean number of hours of nausea per week peaked at 27.8 (SD, 25.7; range, 0–105) at week 7 and was 6.9 (SD, 12.3; range, 0–52) at week 14 (figure 2). Vomiting was less prevalent, with 31.4 percent of subjects reporting at least one episode during week 8 and 16.1 percent during week 14. The mean number of vomiting episodes per week ranged from 1.5 (SD, 3.5; range, 0–20) at week 8 to 0.7 (SD, 2.0; range, 0–11) at week 13. At least one day of appetite loss was reported by 77.9 percent of the subjects during week 8, decreasing to 27.6 percent during week 14. The mean number of days of appetite loss per week (ranging from 0–7 days) was 3.7 (SD, 2.7) during week 8 and 1.3 (SD, 2.4) during week 14. During the first trimester, 92 percent ($n = 85$) reported an aversion to a specific food or beverage.

TABLE 1. Characteristics of 92 eligible participants, Cincinnati, Ohio, 1996–1998

	%	Mean (SD*)	Median	Range
Education				
High school	13			
Some college	28			
College graduate	59			
Pregnancy history (no.)				
Gravidity		2.4 (1.3)	2.0	1–7
Previous miscarriages†	26			
Primigravidas	28			
Annual income (\$)				
≤20,000	3			
20,001–40,000	32			
40,001–60,000	30			
>60,000	33			
Unknown	2			
Smoking history				
Former smoker	46			
Never smoker	54			
Age (years)				
Caucasian	90	30.3 (3.8)	30.5	20–40
Married	96			
Coffee consumption at last menstrual period (ounces‡/week)				
		106.7 (72.2)	86.0	18–378
Caffeine from coffee consumption at last menstrual period (mg/week)§				
		872 (743)	670	11–4,018
Caffeine from all beverage consumption at last menstrual period (mg/week)§				
		1,214 (955)	1,030	17–4,406

* SD, standard deviation.

† Among multigravidas.

‡ One ounce = 0.296 dl.

§ The following values were used to calculate caffeine content (mg/ounce): coffee (drip brewed, 10.6; instant, 9.4; decaf, 0.4; espresso/cappuccino, 40; percolated, 10.6); tea (brewed, 6.2; instant, 3.5; iced, 3.1); sodas (cola, 3.5; flavored cola, 3.4; yellow soda, 4.5; Barq's root beer (Coca-Cola North America, Atlanta, GA), 1.9); other (hot chocolate, 0.8; chocolate milk, 0.6; solid dark chocolate, 20.0; solid milk chocolate, 6.0; and chocolate cake/brownie, 6.4). References: Calorie, nutrient and ingredient content of major types of soft drinks. Washington, DC: National Soft Drink Association, 1999 (www.nstda.org/softdrinks/ingredients.html); Coffee science source. New York, NY: National Coffee Association, 2001 (<http://www.coffeescience.org>); Everything you need to know about ... caffeine: what products contain caffeine and how much? Washington, DC: International Food Information Council Foundation, July 1998 (http://www.ific.org/proactive/newsroom/release_vtml?id=17784).

Figure 2 illustrates the mean weekly hormone metabolite values for the participants in this pregnant cohort. These patterns are consistent with those reported previously in the literature (26). Estrone-3-glucuronide increased steadily from week 6 to week 12; the mean value at week 12 (163 ng/mg of creatinine) is nearly fourfold that of week 6 (44 ng/mg

of creatinine). Pregnanediol-3-glucuronide increased from 9,795 ng/mg of creatinine at week 6 to 13,391 ng/mg of creatinine at week 12. Human chorionic gonadotropin had a bell-shaped curve, increasing from 13,799 mIU/ml at week 6 to 60,344 mIU/ml at week 9 and decreasing to 31,722 mIU/ml at week 12. As shown in figure 2, the pattern of human

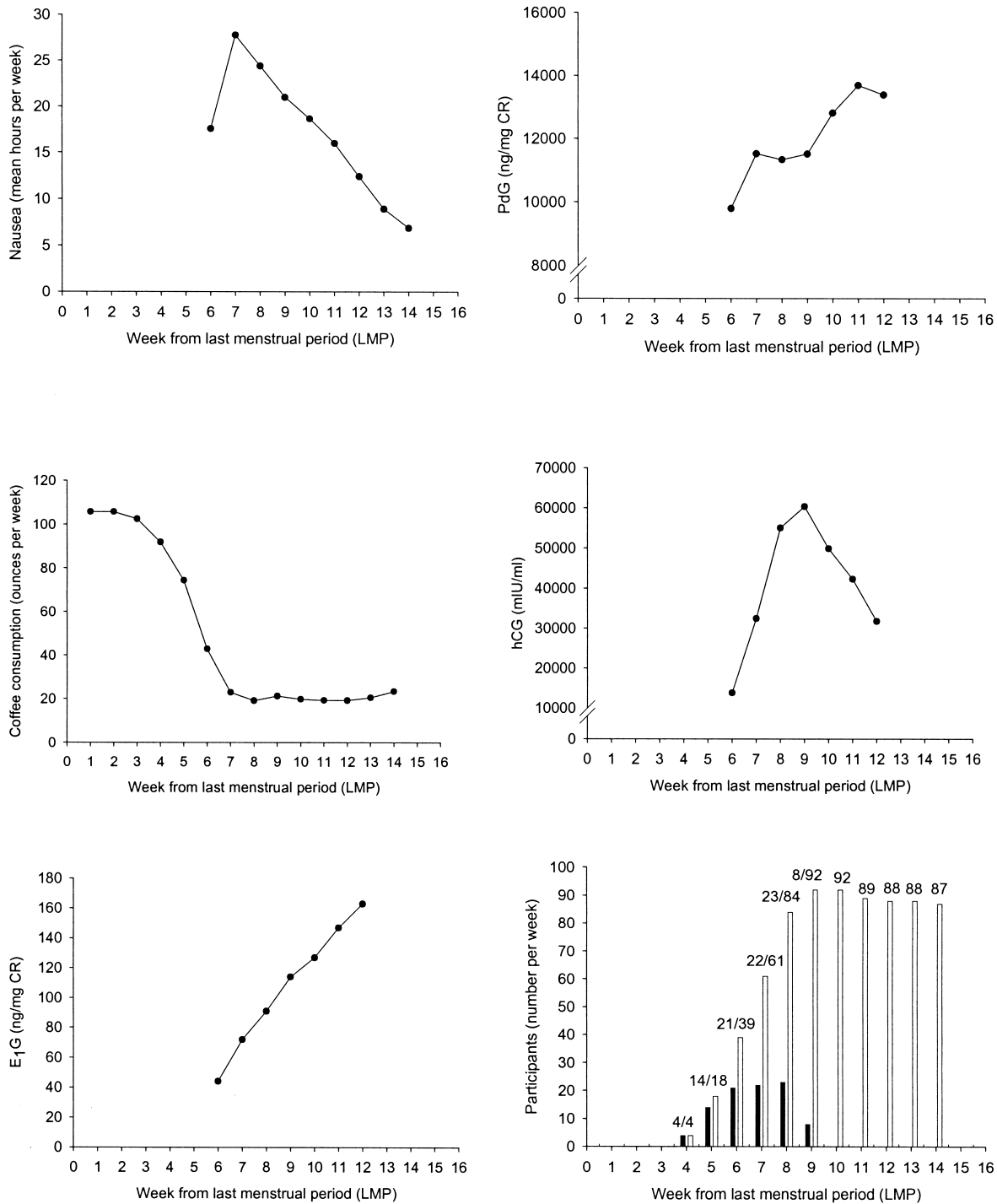


FIGURE 2. Weekly nausea, coffee consumption, hormone metabolite levels, and participation levels by week from last menstrual period in 92 participants, Cincinnati, Ohio, 1996–1998. CR, creatinine; E₁G, estrone-3-glucuronide; hCG, human chorionic gonadotropin; PdG, pregnanediol-3-glucuronide. One ounce = 0.296 dl. Black bar, number enrolling each week; white bar, number of participants each week.

chorionic gonadotropin is similar to that of nausea while being inverse to the pattern of coffee consumption. Figure 2 also illustrates weekly enrollment and participation.

The results of research questions 1–3 are shown in table 2. Weekly coffee consumption was inversely and significantly associated with weekly human chorionic gonadotropin

TABLE 2. Statistical results of research questions 1, 2, and 3: weekly coffee consumption and concurrent hormone metabolite levels, weekly symptoms and concurrent hormone metabolite levels, and weekly coffee consumption and concurrent pregnancy symptoms for weeks 6–12 from the last menstrual period in 92 participants, Cincinnati, Ohio, 1996–1998

Outcome variable, main independent variable(s), and covariates	Regression coefficient†	95% confidence interval
Weekly coffee consumption (ounces‡/week)		
Weekly human chorionic gonadotropin (log)	-5.8***	-7.9, -3.8
Coffee at last menstrual period (ounces/week)	0.1**	0.05, 0.2
Former smoker (yes/no)	11.3	-0.4, 22.9
First pregnancy (yes/no)	-11.2	-23.8, 1.3
Weekly estrone-3-glucuronide (log)		
Coffee at last menstrual period (ounces/week)	0.1**	0.04, 0.2
Former smoker (yes/no)	12.1*	0.5, 23.6
First pregnancy (yes/no)	-8.6	-21.1, 3.9
Weekly pregnanediol-3-glucuronide (log)		
Coffee at last menstrual period (ounces/week)	0.1**	0.03, 0.2
Former smoker (yes/no)	12.9*	1.1, 24.6
First pregnancy (yes/no)	-11.0	-23.7, 1.7
Weekly nausea (hours/week)		
Weekly human chorionic gonadotropin (log)	2.6***	1.2, 3.9
Weekly estrone-3-glucuronide (log)	-3.0*	-5.5, -0.4
Weekly pregnanediol-3-glucuronide (log)	-5.0*	-9.1, -0.9
Appetite loss (days/week)		
Weekly human chorionic gonadotropin (log)	0.1	-0.1, 0.3
Weekly estrone-3-glucuronide (log)	-0.7***	-1.1, -0.4
Weekly pregnanediol-3-glucuronide (log)	-0.7*	-1.2, -0.2
Vomiting (episodes/week)		
Weekly human chorionic gonadotropin (log)	0.1	-0.1, 0.3
Weekly estrone-3-glucuronide (log)	-0.2	-0.5, 0.2
Weekly pregnanediol-3-glucuronide (log)	-0.3	-0.9, 0.3
Weekly coffee consumption (ounces/week)		
Nausea (hours/week)		
Coffee at last menstrual period (ounces/week)	-0.1	-0.2, 0.1
Coffee at last menstrual period (ounces/week)	0.1***	0.06, 0.2
Appetite loss (days/week)		
Coffee at last menstrual period (ounces/week)	-0.1	-1.1, 0.9
Coffee at last menstrual period (ounces/week)	0.1**	0.04, 0.2
Former smoker (yes/no)	13.0*	1.3, 24.8
First pregnancy (yes/no)	-11.1	-23.8, 1.6
Vomiting (episodes/week)		
Coffee at last menstrual period (ounces/week)	-1.0*	-1.9, -0.01
Coffee at last menstrual period (ounces/week)	0.1**	0.04, 0.2
Former smoker (yes/no)	13.4*	1.8, 25.0
First pregnancy (yes/no)	-10.0	-22.6, 2.6

* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

† A regression coefficient in the log scale translates in the following manner: every 2.5- to 3-fold increase in human chorionic gonadotropin is related to an average decrease in coffee of 5.8 ounces (1.7 dl) per week. Similarly, a 2.5- to 3-fold increase in estrone-3-glucuronide is related to an average decrease in coffee of 7.6 ounces (2.2 dl) per week.

‡ One ounce = 0.296 dl.

levels ($p < 0.0001$), after adjustment for coffee at the last menstrual period, being a former smoker (yes/no), and first pregnancy (yes/no). Coffee consumption was also inversely and significantly associated with weekly estrone-3-glucuronide levels ($p < 0.001$), after adjustment for confounding variables. Pregnanediol-3-glucuronide was inversely associated with coffee consumption, although not statistically significant. To determine if extreme values were driving the results, we excluded the top 10 percent of the coffee consumers and then the top and bottom 10 percent; the results did not differ.

Research question 2 asked if the levels of hormone metabolites were associated with pregnancy symptoms. The weekly number of hours of nausea was directly and significantly associated with human chorionic gonadotropin (table 2; $p < 0.001$) and inversely and significantly associated with estrone-3-glucuronide ($p < 0.05$) and pregnanediol-3-glucuronide ($p < 0.05$). Each metabolite was examined separately. No covariates were significant in these models, so the results are presented unadjusted. Thus, every 2.5- to 3-fold increase in human chorionic gonadotropin was associated with an increase in the frequency of nausea of 2.6 hours per week. Analysis of appetite loss as the outcome gave similar results, though human chorionic gonadotropin was not significant (table 2). The frequency of vomiting (number of episodes per week) was not significantly associated with any of the hormones.

To analytically address the question of how well nausea could be used as a surrogate for pregnancy hormones, we calculated the ratio of the log likelihood, explained by the regression of each (untransformed) hormone metabolite on the weekly hours of nausea, to the log likelihood that can be explained by a model that included multiple predictor variables (27). The full model included the weekly number of hours of nausea, first pregnancy (yes/no), ever smoker (yes/no), and the weekly number of ounces of coffee consumption. The proportion of the variance in human chorionic gonadotropin, estrone-3-glucuronide, and pregnanediol-3-glucuronide that was explained by nausea was between 67 and 69 percent.

Results of research question 3 are shown in table 2. Of the pregnancy symptoms, only vomiting frequency was significantly related to a decrease in coffee consumption, adjusted for coffee consumption at the last menstrual period, first pregnancy, and ever smoker.

These first three analyses (research questions 1, 2, and 3) were repeated to test the associations between milligrams of caffeine instead of ounces of coffee. The results were similar, though stronger for coffee than caffeine in some cases.

Coffee consumption at the last menstrual period was analyzed for an association with subsequent weekly hormone metabolites (research question 4). Coffee intake at the last menstrual period (more than 8 ounces/day (2.4 dl)) was not associated with subsequent weekly levels of estrone-3-glucuronide or human chorionic gonadotropin. However, pregnanediol-3-glucuronide levels were significantly lower by an average of 32.2 percent for weeks 6–12 in the higher consumption group ($p = 0.005$), adjusted for age. Table 3 shows weekly pregnanediol-3-glucuronide levels, stratified

by the two consumption groups. Adjusting for weekly coffee consumption during pregnancy did not change the effect estimates. When coffee consumption at the last menstrual period was stratified into tertiles, a dose-response relation with pregnanediol-3-glucuronide was observed. Nausea severity (greater than 7 hours per week vs. less) was not a significant covariate. When coffee consumption was dichotomized into more than 12 ounces per day (3.5 dl) versus less, the higher consumption group had 22.5 percent lower levels of first trimester pregnanediol-3-glucuronide ($p = 0.03$), adjusted for age. When the results were stratified into four groups combining former/never smoker and high/low coffee consumption at the last menstrual period, no change in the level of pregnanediol-3-glucuronide was observed for prior smoking. A repeated measures analysis showed that prior smoking was not significantly associated with any of the three hormones. Caffeine consumption from all beverage sources at the last menstrual period was not significantly associated with any of the three hormone metabolite levels.

Nine study miscarriages that were excluded from the analyses had hormonal data available for some weeks, with pregnanediol-3-glucuronide levels ranging from 8,800 to 10,700 ng/mg of creatinine (weeks 7–10). The median amount of coffee consumed at the last menstrual period for those who miscarried was 84 ounces/week (24.8 dl), similar to that of the main analysis cohort. Unfortunately, because of the variation in and timing of miscarriages, there were too few data to allow for a meaningful analysis of the patterns of coffee consumption, hormones, and miscarriage.

DISCUSSION

Our analyses show that the levels of early pregnancy hormone metabolites, coffee consumption patterns, and pregnancy symptoms are significantly associated with each other. It has been postulated that viable pregnancies have higher hormone levels, causing women to experience greater nausea and coffee aversion and a subsequent decrease in coffee consumption (16). In this study, coffee consumption decreased significantly with rising human chorionic gonadotropin and estrone-3-glucuronide levels, supporting such a hypothesis.

Nausea and appetite loss were significantly associated with all three hormone metabolites. Although the inverse relation between these symptoms and estrone-3-glucuronide and pregnanediol-3-glucuronide seems unexpected, it is likely because the majority of hormone data are from weeks 8–12, a time when estrone-3-glucuronide and pregnanediol-3-glucuronide were increasing, nausea was decreasing, and human chorionic gonadotropin was decreasing (figure 2). Although nausea accounted for 67–69 percent of the variance in these three hormone metabolites, nausea and appetite loss were not significantly related to a concurrent decrease in coffee consumption, though figure 2 suggests such a relation. The decrease in consumption due to coffee aversion may be a phenomenon that is independent of nausea and appetite loss and, thus, nausea might not be a good surrogate for hormones as they relate to coffee consumption. We feel strongly that future studies of coffee consumption and preg-

TABLE 3. Weekly pregnanediol-3-glucuronide levels (ng/mg of creatinine) by amount of coffee consumption (ounces/week) at the last menstrual period in 92 participants, Cincinnati, Ohio, 1996–1998

Week from last menstrual period	Pregnanediol-3-glucuronide levels of high coffee consumers (>8 ounces*/day at last menstrual period)			Pregnanediol-3-glucuronide levels of low coffee consumers (≤8 ounces/day at last menstrual period)		
	Mean (SD)†	Range	No. of samples/week‡	Mean (SD)	Range	No. of samples/week
6	8,395 (3,947)	3,388–17,357	21	13,062 (8,087)	5,239–29,987	9
7	9,022 (4,341)	2,672–20,247	32	16,856 (17,290)	4,764–75,797	15
8	9,351 (3,864)	2,692–17,704	54	16,438 (13,391)	4,061–63,481	21
9	10,679 (5,384)	1,750–38,764	71	14,086 (8,718)	5,444–46,147	23
10	11,436 (5,304)	2,355–24,422	66	16,605 (11,085)	4,503–55,997	24
11	12,394 (5,816)	2,870–27,328	66	17,580 (10,718)	5,140–52,611	22
12	12,821 (6,577)	2,605–30,525	51	15,102 (7,292)	6,102–32,059	17

* Approximately one mug (2.4 dl).

† SD, standard deviation.

‡ Subjects supplied their first urine sample the morning after enrollment, followed by weekly samples given every Wednesday. Thus, the first two samples are not necessarily 7 days apart and, in some cases, led to two samples in the first week of collection. Weeks 10–12 show less than 92 total samples per week because of dropouts or incomplete data.

nancy outcome should measure coffee aversion along with symptoms.

“Coffee aversion,” per se, has been examined sparingly in previous studies. A study by Hook (15) reported that 21 percent of coffee drinkers cited a physical aversion as a reason for decreasing, compared with 59 percent reported in our study. That study collected data retrospectively after delivery (15), however, likely explaining this difference. In both the current study and the study by Hook (15), coffee was one of the most prominently rejected food items during early pregnancy. Cnattingius et al. (10) reported that 21 percent of subjects having spontaneous abortions and 41 percent of control subjects experienced an aversion to coffee, though they did not describe how the data were measured.

Another significant finding of the current study was a marked decrease in subsequent gestational levels of pregnanediol-3-glucuronide associated with higher levels of coffee consumption at the last menstrual period. When the association was tested using various cutpoints, the findings were robust. Because the 92 pregnancies analyzed in our study did not end in fetal loss, it is difficult to determine what biologic or clinical significance these findings have, though the levels of pregnanediol-3-glucuronide appear to be above what was necessary to maintain these pregnancies. We could find no previous literature assessing a “critical” level of first trimester urinary pregnanediol-3-glucuronide for pregnancy maintenance, though there is some information using other media or assay methods (28–33). There is no animal literature showing that coffee consumption prior to pregnancy is related to subsequent pregnancy hormone levels, and no studies in humans have examined this research question. It is plausible that coffee is related to a lower pregnanediol-3-glucuronide level prior to pregnancy that remains lower during the first trimester. It was interesting that the decrease in pregnanediol-3-glucuronide was not related to caffeine consumption, but only to coffee, even though 72 percent of the caffeine consumed in this study was from coffee.

There are limitations to measuring urinary metabolites instead of plasma levels of circulating hormones. It is possible that differences in pregnanediol-3-glucuronide between low and high coffee drinkers may be due to metabolism and not to hormonal differences. Estrogen and progesterone follow the same metabolic pathway, resulting in estrone-3-glucuronide and pregnanediol-3-glucuronide, respectively. In the current study, pregnanediol-3-glucuronide was decreased while estrone-3-glucuronide was not affected, suggesting that the mechanism is acting on the production and not the metabolism of progesterone. It is also possible that unmeasured factors, such as the storage of steroid hormones in adipose tissue, may explain some of the differences by influencing the clearance of hormone metabolites. Clearly, there is a need for additional research of progesterone levels in early pregnancies in order to elucidate the significance of these findings.

Many previous studies of pregnancy outcome and coffee/caffeine consumption did not account for nausea (3, 5, 6, 9, 12), and most that did account for nausea or coffee aversion did not collect detailed, prospective, or repeated measurements of these factors (2, 4, 7, 8). Two recently published studies attempted in their study design to test whether nausea or coffee aversion is confounding an association between coffee consumption and pregnancy loss (10, 11). The study by Cnattingius et al. (10) found, in an analysis of nonsmokers stratified by coffee aversion status, that a positive, dose-response relation between caffeine consumption and spontaneous abortion was more significant among those who did not report an aversion. Similarly, the study by Wen et al. (11) found a positive, dose-response relation to be more marked in those who continued to consume coffee after nausea started, corroborating a previous study by Fenster et al. (7). Of these studies, only the study by Cnattingius et al. (10) collected repeated, weekly measures of coffee and caffeine and the severity of symptoms, though a repeated measures statistical analysis was not used.

One limitation to our study is that we collected few urine samples before week 6. Hence, we recommend that future biologic sampling be conducted as early as the last menstrual period. Even though our analyses included data only after week 6, when coffee consumption had already decreased by more than 50 percent, we still found significant associations between coffee consumption and hormone metabolite levels. It is difficult, however, to assess whether hormonal changes preceded coffee aversion.

The current results were analyzed in a small sample of healthy pregnancies and are not necessarily generalizable to other populations, such as smokers. Interestingly, former smoking was significantly associated with coffee consumption during early pregnancy. On weekly questionnaires, no one in our cohort reported smoking during the study period; however, smoking and coffee drinking have been shown to be correlated lifestyle habits (34), and the correlation may also exist for former smokers and coffee drinking.

We recommend that the methods used in this study be repeated in a larger population that will allow for analyses of miscarriage and other pregnancy outcomes. As was postulated by Stein and Susser in 1991 (16), fetal demise may be accompanied by decreasing hormone levels, a decline in symptoms, and an increase in coffee consumption before the clinical miscarriage occurs. Such a scenario may lead to an artifactual association between coffee consumption and miscarriage. Thus, future studies of coffee consumption and pregnancy outcomes might include repeated measures of fetal viability, for example, heartbeat or ultrasound, to indicate the timing of fetal demise. It is also recommended that future studies adjust for the severity of symptoms over time, because hormones in spontaneously aborted pregnancies can still rise (28, 29). In addition, the authors recommend that coffee aversion and coffee consumption be considered important variables along with symptoms and caffeine consumption and that researchers not only collect repeated measures but also analyze data using longitudinal methods.

In summary, hormone metabolite levels, pregnancy symptoms, and coffee consumption were significantly associated with each other, indicating the need to adjust for the effects of coffee aversion when evaluating the relation between coffee/caffeine and pregnancy loss. In addition, moderate to high levels of coffee consumption at the last menstrual period were associated with lowered subsequent weekly levels of pregnanediol-3-glucuronide, which was not significant for pregnancy loss in our study, but which may have implications for women who have infertility or insufficient pregnancy hormone levels. This study has the strengths of repeated, detailed measurements of symptoms, consumption, and hormone metabolites, and it shows the significance of these important variables.

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