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Short communication

Presence and concentration of 17 hormones in human placenta processed for encapsulation and consumption

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ABSTRACT

Human maternal placentophagy is a rare but growing practice in several industrialized countries among postpartum mothers seeking a variety of purported health benefits attributed to the practice. These postpartum mothers typically consume their placenta as a processed, encapsulated supplement. To determine whether free (unconjugated) steroid hormones and melatonin in placenta can survive the encapsulation process (namely steaming and dehydration), we analyzed 28 placenta samples processed for encapsulation using liquid chromatography tandem-mass spectrometry (LC-MS/MS) to evaluate the concentration of 17 hormones. The results revealed detectable concentrations for 16 of the hormones analyzed, some in concentrations that could conceivably yield physiological effects.

1. Introduction

The purported benefits of human maternal placentophagy, including improved maternal postpartum affect, energy, and lactation, are widely reported, although these are largely in the form of personal, anecdotal accounts in popular or social media [1], placentophagy advocacy literature [2] and online sources [3], or in self-reported research surveys [4]. To date, however, the practice has not been subjected to rigorous scientific investigation. The frequency of maternal placentophagy in the US and other industrialized countries, where it has been reported as a rare but established practice, is currently unknown, although one estimate based on client reports from a Portland, Oregon lactation consultant suggests as many as 50% of homebirth mothers and 10% of women delivering in birthing centers or hospitals engage in the practice (about 2000 mothers annually) in this US metropolitan area alone [5]. A survey of 189 placentophagic mothers suggests that ingestion of processed, encapsulated placenta is the most common form of the practice [4]. Many postpartum conditions, particularly depression, are thought to be caused by the precipitous drop in estrogens (estradiol and estriol) and progestogens (progesterone and its neuroactive metabolite, allopregnanolone) that occur at birth [see 6]. Placentophagy advocates claim that hormones retained in the placenta, such as estrogens and progesterone, likely provide a key source of the beneficial postpartum effects attributed to placentophagy, such as the relief of depressive symptoms and improved milk production [2,3]. Alternatively, some placentophagy researchers question whether such processing would destroy potentially beneficial biomolecular components [5,7–9]. In order to determine whether cooked and processed placenta retains potentially bioavailable hormones, we used liquid chromatography tandem-mass spectrometry (LC-MS/MS)[1] to analyze the concentration of 17 hormones: 11-deoxycortisol, 17-hydroxyprogesterone, 7-ketodehydroepiandrosterone, aldosterone, allopregnanolone, androstenedione, corticosterone, cortisol, cortisone, dehydroepiandrosterone (DHEA)[2], 5-alpha-dihydrotes-

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[1] LC-MS/MS: Liquid chromatography tandem mass spectrometry ionization.
[2] DHEA: Dehydroepiandrosterone.

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tosterone (DHT)[3], estradiol, estriol, estrone, melatonin, progesterone, and testosterone, in 28 placentas processed for encapsulation and consumption. Due to the exploratory nature of this study and because the hormonal content of encapsulated placenta has not been evaluated, these hormones were selected to provide the most comprehensive profile possible for encapsulated placenta. Additionally, the hormone classes included here are associated with the reported benefits of placentophagy (*e.g.*, effects of steroids on mood) [4,6].

2. Materials and methods

2.1. Placenta donors

All methods were approved by the Institutional Review Board and Institutional Biosafety Committee at the University of Nevada, Las Vegas, and written informed consent was obtained from all participants included in this study. Processed placenta samples were collected from 28 healthy female donors between the ages of 20–38 (mean age = 29.9 y) in the Las Vegas area who had previously decided to ingest their placenta postpartum. One placenta donor reported taking thyroid medication during pregnancy; however, thyroid hormones were not evaluated in the samples for this study. No other participants reported taking hormone supplements during pregnancy (see Table 1 [10]).

2.2. Sample collection

Placentas were processed in the donor's home within 4 days of birth through Placenta Benefits LTD, a Las Vegas based company that provides placenta encapsulation and training for encapsulation providers. Placentas were refrigerated or frozen (where processing occurred more than 24 h postpartum) prior to processing. Each placenta was rinsed in water, stripped of membranes, steamed with herb-infused water (internal temperature of 160 °F), thoroughly dehydrated using a food dehydrator (Excalibur 2400), and pulverized using a food processor (Magic Bullet MB1001C).

2.3. LC-MS/MS methods

Prior to LC-MS/MS analysis, hormones were extracted from placenta samples using QuEChERS methodology [12,13]. A 0.2 g sample was weighed into a 5-mL polypropylene tube and fortified with internal standards. Water (0.5 mL) was added and the tube was vortexed to mix. Acetonitrile (1.0 mL) was added and the tubes were shaken vigorously for 1 min. A mixture of salts (0.3 g Na_2SO_4 and 0.1 g $NaC_2H_3O_2$) was added and the tubes were again shaken for 1 min before entering the centrifuge for 10 min at 3000 rpm.

The upper acetonitrile layer was transferred for further clean-up with C18 SPE. Extracts were eluted from the SPE with 1:4 methanol/dichloromethane and dried under nitrogen. To the dried extract, equal parts sodium bicarbonate (50 mM) and pyridine-3-sulfonyl chloride (3 mg/mL in acetonitrile) were added and the mixture was heated at 60 °C for 10 min to allow for derivatization of the estrogens (estrone, estradiol, and estriol). Following derivatization, the solution was diluted with 1% formic acid for analysis by LC-MS/MS (AB Sciex Triple Quad 5500) using atmospheric pressure chemical ionization (APCI)[4] in the positive ionization mode. All sample analyses were run in singlet.

[3] DHT: 5-alpha-dihydrotestosterone.
[4] APCI: Atmospheric pressure chemical ionization.

Table 1
Clinical characteristics of pregnancies for placentas studied (N = 28) [7].

Parameter	Clinical characteristics[a,b]
Parity (median, 25–75%)	1, 1–2.25, Range = 1–4
Gestational age (weeks)	39.9 ± 1.26
Maternal age (years)	29.9 ± 4.7, Range = 20–38
Race	N = 28
Black	1 (3.6%)
White	22 (78.6%)
Other	5 (17.9%)
Ethnicity	N = 28
Hispanic/Latina	4 (14.3%)
Not Hispanic/Latina	24 (85.7%)
Prenatal medications	N = 28
Iron	4 (14.3%)
Prenatal multivitamin	25 (89.3%)
Other vitamin, mineral, or herbal supplements	13 (46.4%)
Albuterol	1 (3.6%)
Antacids	1 (3.6%)
Claritin	1 (3.6%)
Loratadine	1 (3.6%)
Zoloft	1 (3.6%)
Thyroid medication (unidentified)	1 (3.6%)
Drugs	N = 28
Cigarettes	0
Alcohol	0
Other	0
Previous prenatal admission(s)	N = 28
Yes	3 (10.7%)
Placenta previa	1 (3.6%)
Chorioangioma	1 (3.6%)
Preeclampsia	1 (3.6%)
No	25 (89.3%)
Antibiotics in labor	N = 28
None	24 (85.7%)
Unspecified	4 (14.3%)
Beta strep status	N = 28
Positive	1 (3.6%)
Negative	27 (96.4%)
Anesthesia	N = 28
Epidural	10 (35.7%)
Narcotics	1 (3.6%)
General	0
Other/none	9 (32.1%)
Unknown	8 (28.6%)
C-section	N = 5
Repeat, no labor	0
Repeat, with labor	2 (7.1%)
Primary, no labor	1 (3.6%)
Primary, with labor	2 (7.1%)
Maternal oxygen given at delivery	N = 28
Yes	1 (3.6%)
No	27 (96.4%)
Birth weight (grams)	3522.4 ± 491.8
Baby's sex	N = 28
Yes	14 (50.0%)
No	13 (46.4%)
Unknown	1 (3.6%)

[a] Values are expressed as means ± SD, or number (percentage) unless otherwise stated.
[b] Clinical characteristics were unknown for the following parameters: gravidity, blood pressures, screened for diabetes, antenatal steroids, magnesium sulfate, cervical ripening agent, labor, placental weight, and minutes from delivery to processing.

3. Results and discussion

Fifteen of the 17 hormones analyzed were detected in all 28 placenta samples. Melatonin was detected in only one third of the samples (n = 9, 32.1%), and DHT, the most active of the androgens, was below the detection limit (Table 2). Table 3 provides an overview of published hormone concentrations in unprepared placenta [15–20]. Variation between these values and our findings may

Table 2
Concentrations of 17 hormones in 28 dehydrated placenta samples.

Hormone	Number of detects	Concentration range (ng/g)	Concentration means ± SEM (ng/g)	Estimated intake from maximum recommended daily dose of placenta capsules (μg)[a]
11-Deoxycortisol	28	15.180–121.092	52.305 ± 4.16	0.173
17-hydroxyprogesterone	28	82.762–1105.969	265.611 ± 37.45	0.877
7-ketodehydroepiandrosterone	28	9.027–47.864	22.208 ± 1.899	0.073
Aldosterone	28	0.179–2.993	1.274 ± 0.151	0.004
Allopregnanolone[b]	28	37.898–181.745	111.147 ± 7.817	0.367
Androstenedione	28	97.501–1134.326	365.95 ± 40.002	1.208
Corticosterone	28	2.183–59.345	15.237 ± 2.432	0.050
Cortisol	28	9.829–205.696	85 ± 0.01	0.281
Cortisone	28	356.662–2171.952	1196 ± 0.074	3.947
Dehydroepiandrosterone	28	35.353–288.729	84.795 ± 9.67	0.280
Dihydrotestosterone	0	<DL	<DL	<DL
Estradiol[b]	28	44.612–172.959	103.46 ± 6.321	0.341
Estriol	28	453.480–926.433	752.187 ± 23.047	2.482
Estrone	28	172.527–582.218	343.472 ± 18.757	1.133
Melatonin	9	0.163–0.494	0.14 ± 0.026	<0.001
Progesterone[b]	28	4307.218–15508.879	11314.029 ± 393.895	37.336
Testosterone	28	5.078–119.290	30.503 ± 4.275	0.101

<DL: Below detectable limit.
[a] Based on maximum daily intake of placenta capsules of 3300 mg [2,14].
[b] Mean concentrations in 3300 mg of placenta capsules may potentially reach physiological effects thresholds.

result from differences in sampling site, tissue preparation, extraction, and analysis methods that impact hormone recovery.

Because many factors affect hormone bioavailability and bioactivity, such as delivery method and interaction between hormones, it is difficult to say conclusively whether the values reported here could elicit physiological effects in women taking placenta capsules. Despite this limitation, while concentrations of many of the selected hormones are relatively low, mean

Table 3
Published concentrations of hormones in unprepared placenta.

Hormone	Concentration means ± SEM (ng/g wet wt)	Concentration ranges (ng/g wet wt)	Sample location	References
11-Deoxycortisol	13.86 ± 2.08[a,c]	–	Homogenized from 3 locations	[15]
17-hydroxyprogesterone	42.96 ± 6.28[a,c]	–	Homogenized from 3 locations	[15]
Androstenedione	8.59 ± 1.72[a,c]	–	Homogenized from 3 locations	[15]
	47.8 ± 11.2[d]	–	Central basal surface (cotyledons)	[16]
Corticosterone	0.003 ± 0.001[c]	–	Homogenized from 3 locations	[15]
Cortisol	0.01 ± 0.001[c]	–	Homogenized from 3 locations	[15]
	–	0.9 ± 0.1–1.2 ± 0.2[c]	Core	[17]
	–	3.6 ± 0.4–5.6 ± 0.4[c]	Basal surface	[17]
Cortisone	0.72 ± 0.03[c]	–	Homogenized from 3 locations	[15]
	–	0.9 ± 0.1–1.2 ± 0.2[c]	Core	[17]
	–	73.3 ± 6.8–80.3 ± 3.0[c]	Basal surface	[17]
Estradiol	–	0.5–1.0[d]	Basal surface (cotyledons)	[18]
	150 ± 30[d]	–	Central basal surface (cotyledons)	[16]
	206 ± 16[d]	–	Central basal surface	[19]
	234 ± 23[d]	–	Central basal surface	[19]
Estriol	–	270–380[d]	Basal surface (cotyledons)	[18]
Estrone	–	30–50[d]	Basal surface (cotyledons)	[18]
	41 ± 5.2[d]	–	Central basal surface	[19]
	50 ± 6.9[d]	–	Central basal surface	[19]
	1410 ± 140[d]	–	Central basal surface (cotyledons)	[16]
Melatonin	–	0.50–0.80[b,d] (approximate)	Not specified	[20]
Progesterone	26.6 ± 3.8[d]	–	Central basal surface (cotyledons)	[16]
	–	80–110[d]	Basal surface (cotyledons)	[18]
	–	887.8 ± 117.4–919.3 ± 25.5[c]	Basal surface	[17]
	607.0 ± 26.12[c]	–	Peripheral chorionic surface	[17]
	700.0 ± 47.8[c]	–	Intermediate villous core	[17]
	818.5 ± 28.6[c]	–	Proximal villous core	[17]
	749.7 ± 33.3[c]	556.0–904.0[c]	Core	[17]
	907.4 ± 36.1[c]	684.0–1120.0[c]	Basal surface	[17]
	4340 ± 328[e]	–	Basal surface	[19]
	4940 ± 327[e]	–	Basal surface	[19]
Testosterone	0.577 ± 0.577[a,c]	–	Homogenized from 3 locations	[15]

SEM: Standard error of the mean.
[a] Value converted from nmol/g.
[b] Value converted from pg/g.
[c] Concentration analyzed using liquid chromatography tandem mass spectrometry (LC-MS/MS).
[d] Concentration analyzed using radioimmunoassay (RIA).
[e] Concentration analyzed using liquid chromatography (LC).

concentrations of estradiol, progesterone, and allopregnanolone could potentially reach physiological effect thresholds, given the maximum 3300 mg/day intake guidelines of some encapsulation providers [2,21] [5]. While our study is the first to use state-of-the-art methods (LC-MS/MS) to analyze hormone concentrations in human placenta processed for encapsulation, they are consistent with a Thai study which, using less sensitive and specific methods (chemiluminescent enzyme immunoassay), and analysis of fewer steroids, nevertheless detected progesterone, estradiol, and testosterone in heat-dried placenta, albeit at lower concentrations [22]. Given these results, future studies investigating possible dose-response effects and measurement of steroids and melatonin in body fluids (blood, urine, saliva) associated with this novel post-partum supplement are warranted.

Conflict of interest statement

This study was made possible, in part, by the collaboration between the study authors and Placenta Benefits LTD, a human placentophagy information and advocacy website and encapsulation service provider. Placenta Benefits LTD encapsulation providers were paid the standard fee for the placenta encapsulation services they provided to study participants. No Placenta Benefits LTD personnel were involved in any part of the study design, data collection, data analysis, or manuscript preparation. None of the study authors have any financial interest in Placenta Benefits LTD, or any other human placentophagy advocacy or services entity.

Contributors

Sharon M. Young: I declare that I participated in the study design, data and sample collection, interpretation of the data, and manuscript preparation, and that I have approved the final version. I declare no conflicts of interest.

Laura K. Gryder: I declare that I participated in the study design, data and sample collection, interpretation of the data, and manuscript preparation, and that I have approved the final version. I declare no conflicts of interest.

David Zava: I declare that I participated in the sample analysis, interpretation of the data, and manuscript preparation, and that I have approved the final version. I declare no conflicts of interest.

David W. Kimball: I declare that I participated in the sample analysis, interpretation of the data, and manuscript preparation, and that I have approved the final version. I declare no conflicts of interest.

Daniel C. Benyshek: I declare that I participated in the study design, interpretation of the data, and manuscript preparation, and that I have approved the final version. I declare no conflicts of interest.

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[5] The 3300 mg/day maximum dose is recommended by Placenta Benefits, LTD, whose encapsulation method was used in this study, by Cornelia Enning in *Placenta*: *the gift of life* [2], and is consistent with the Traditional Chinese Medicine (*Materia Medica*) recommendation of 1.5–4.5 g [14].