Presence of Group B Streptococcus in Raw and Dehydrated Placentas

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ABSTRACT

Placentophagy is a growing global practice, as many mothers choose to have their placenta encapsulated after giving birth for postpartum wellness. A case report of an infant infected at birth with group B *Streptococcus* (GBS) whose mother consumed her encapsulated placenta has left some medical professionals concerned about the safety of the practice for mothers who are vaginally colonized with GBS during pregnancy. The purpose of this study is to determine if GBS is present in the placentas of mothers who test with GBS-positive results and to determine if encapsulation inhibits the survival of GBS in placentas infected with the bacteria. Twelve placentas were collected from mothers who delivered vaginally after testing with positive results for GBS during pregnancy. The raw placentas were swabbed and cultured for GBS using selective GBS media. Placentas were then cut into 55 separate samples and 46 were manually infected with a 0.5 McFarland of GBS culture. Each sample was dehydrated to simulate the encapsulation process and the dried powder was tested for GBS. Of the 12 original placentas, 2 had GBS-positive results following birth. Of the 46 manually infected placentas, 32 had negative results for GBS after dehydration, suggesting a relative risk reduction of 0.6957 and an absolute risk reduction of 69.57%. It was concluded that the percentage of raw placentas infected from the birth process is inherently low, and the encapsulation process significantly decreases the presence of GBS in infected placentas.

ABBREVIATIONS: ARR - absolute risk reduction, CDC - Centers for Disease Control, GBS - group B Streptococcus, IRB - Internal Review Board, QC - quality control, RR - relative risk, RRR - relative risk reduction.

Clin Lab Sci 2020;33(4):60–66

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INTRODUCTION

Over the past several decades, tens of thousands of new mothers have chosen to have their placentas encapsulated after giving birth.[1-5] Placenta encapsulation is the process of dehydrating the placenta, grinding it into a powder, and putting it in capsules to consume postpartum.[1-4] The reported benefits of placentophagy include improved mood, increased energy/decreased fatigue, and improved lactation.[2,5] There have been numerous studies on animal placentophagy but few studies on human maternal placentophagy.[2,6,7] These studies focused on the mothers' mood as well as hormones and nutrients found in the placenta.[8-10]

As with any emerging topic in the medical field, placentophagy has raised questions within the medical community concerning the safety of the practice for those involved—in this case, the mother and baby.[1-5] A single case study of late-onset group B *Streptococcus* (GBS) in a newborn, whose mother consumed her placenta capsules, led to concern of a link between the capsules and neonatal infection.[11] GBS colonizes the genital and gastrointestinal tracts and can cause early or late-onset neonatal disease in the mother, newborn, or fetus.[12] GBS is one of the leading causes of neonatal mortality and morbidity worldwide, causing meningitis, pneumonia, septicemia, and bacteremia in susceptible neonates.[12,13]

Following this case study, the Centers for Disease Control (CDC) and the American College of Obstetricians and Gynecologists made a sweeping recommendation against the practice of placentophagy, especially for mothers who had positive test results for GBS antepartum.[4,11] Health care professionals who advocate for placentophagy believe this recommendation to be hasty because there is neither clinical research to back up the claim of causality between placenta encapsulation and GBS in newborns, nor is there any reported increase in morbidity and mortality in newborns related to placenta encapsulation.[3,5] In fact, a systematic review of 23 000 births that compared neonatal outcomes between women who encapsulated their placenta and women who did not found that placentophagy was not associated with any adverse neonatal outcomes such as hospitalization or neonatal deaths.[5]

There is currently no research on whether GBS survives in the placenta after the encapsulation process.[1] In light of the continuing popularity of placenta encapsulation, the seriousness of GBS infections in newborns, and

the lack of literature on GBS in placentas, it is the goal of this study to determine whether GBS is present in the placenta of mothers who have GBS-positive test results after birth and after the encapsulation process. This study will look specifically at the presence of GBS in both raw and dehydrated placentas and allow mothers with GBS-positive test results and their care providers to better understand the actual risk of GBS contamination from consuming placenta.

Research Questions

This experiment set out to answer 2 questions. (1) Is GBS present in the raw placenta of mothers who had positive test results for GBS during pregnancy? (2) If GBS is present in the raw placenta, is the process of encapsulation sufficient to inhibit the survival of GBS in the dehydrated placenta?

GBS screening is now routinely performed on all pregnant women during their 35th–37th week of pregnancy,[14] and the universal administration of prophylactic antibiotics during labor for women who have positive test results for GBS has decreased the incidence of neonatal GBS infection by 80%.[15,16] Therefore, it is hypothesized that the placentas will have negative test results for GBS at birth. It is further hypothesized that in placentas contaminated with GBS, the process of dehydration will significantly reduce the survival of GBS.

MATERIALS AND METHODS

Placenta Donation and Collection

This study was approved by the Internal Review Board (IRB) and Biosafety Committee of Augusta University (Appendix 1). Verbal consent was obtained from each mother to use her placenta in adherence with IRB requirements (Appendix 2). All placentas came from the Atlanta Birth Center or an Atlanta area hospital. Twelve placentas were collected from postpartum mothers who had positive test results for GBS via a vaginal swab during pregnancy and had a vaginal birth. Exclusion criteria included mothers who have a condition that requires medical intervention during pregnancy, mothers with an active infection at the time of birth, and mothers delivering by cesarean section.

Preparation and Encapsulation

There are 2 common methods of placenta encapsulation used today. One method involves steaming and dehydrating the placenta (steamed-dehydrated method), while the other excludes the steaming step and involves just dehydration (raw-dehydration method). The raw-dehydration method was chosen over the steamed-dehydration method because previous studies have shown the steamed-dehydration method to destroy more microorganisms than raw dehydration, and it was advantageous to test the method that would pose the greatest risk.[1] Therefore, by extrapolation, it can be assumed that steamed-dehydrated placentas would show an even greater reduction in the presence of GBS.

The individual placentas were either frozen or processed within 48 hours of collection. Each placenta was stored and processed separately. Placentas were thoroughly rinsed, and blood clots were removed. The amnion, amniotic sac, and umbilical cord were removed and discarded. The placenta was sliced into thin strips using stainless steel scissors and placed on parchment paper on trays of a Nesco Snackmaster Pro dehydrator. The strips were thoroughly dried for 18–24 hours set at 160°F, measured by the dehydrator thermostat. After drying, the placenta strips were tested for dryness by snapping them in half. If they did not easily "snap," they were dehydrated further.[1] The dried strips were ground into a fine powder using an Oster blender. The placenta powder was the final product. In commercial encapsulation, this dried powder is then put into gelatin capsules for oral administration. All surfaces and equipment were cleaned and sanitized using a 10% bleach solution for 10–30 minutes between each sample preparation, according to Occupational Safety and Health Administration standards for sanitation for bloodborne pathogens.[17]

Testing for GBS

Before processing, both the maternal and fetal sides of the placenta were swabbed with a sterile cotton swab and the swab was immediately placed in a Hardy Diagnostics Strep B Carrot Broth to detect the presence of GBS in the raw placentas.[18-20] The Carrot Broth was incubated at 37°C for 24 hours and read the following day for a positive or negative result. An orange color indicated the sample was positive for GBS, while no color change indicated a negative result.[21] For further confirmation, a Hardy Diagnostics GBS Detect plate was then streaked with a loop of the Carrot Broth sample. The GBS Detect plates were incubated for 24 hours at 37°C and read the following day for growth or no growth.[22] Both media are selective for GBS. A quality control (QC) was run alongside each batch of samples, using a pure culture of commercial GBS grown on sheep blood agar.

Most of the placentas initially had negative test results for GBS. To test the second research question of whether the encapsulation process is sufficient to kill GBS, individual samples of placenta were manually contaminated with GBS. The placentas were split into 55 individual samples and processed each sample as a separate specimen. Using a 0.5 McFarland standard made from the QC-GBS culture, 1 to 4 drops of GBS was used to contaminate 46 of the 55 placenta samples. Each placenta sample was then retested using Carrot Broth and GBS Detect plates, as previously described, to ensure each sample had been successfully infected with the GBS. Each sample was then dehydrated and processed individually according to the

encapsulation method described in the previous section. After the placenta sample was fully dehydrated and ground into powder form, the dried powder was swabbed and tested using the same procedure as the previously described raw samples.

Statistical Analysis

For each placenta sample, the results were recorded of the raw-unadulterated placentas, the contaminated samples, and the dehydrated-placenta powder as positive results for GBS or negative results for GBS. Relative risk (RR), relative risk reduction (RRR), and absolute risk reduction (ARR) analyses were used to determine if the process of encapsulation reduced the amount of GBS present.[23] The RR in this case is the ratio of the probability of the dehydrated placentas having GBS vs. the probability of the raw contaminated placentas having GBS. The RRR is the ratio of the probability of the dehydration process reducing the presence of GBS compared to the raw contaminated placentas.

RESULTS

Out of the 12 placentas collected, 2 (16.7%) initially had positive test results for GBS. The placentas were cut up into a total of 55 samples with 9 of the samples left uncontaminated. These 9 samples remained negative after dehydration. Of the 46 manually infected samples, all 46 raw samples had positive results for GBS after contamination. After dehydration, 14 of the samples (30.4%) still had positive results for GBS and 32 (69.6%) had negative results for GBS (Table 1). The RR analysis calculated a RR of 0.3043 with a 95% CI of 0.1966 to 0.4711 and $P < 0.0001$ when the α value is set to 0.05 (Table 2). Therefore, the RRR is 0.6957 and the ARR is 69.57%.

Table 2. Statistical analysis of RR for contaminated placentas after dehydration process

RR	0.3043
RRR	0.6957
ARR	69.57%
95% CI for RR	0.1966–0.4711
Significance level	$P < 0.0001$
α value	$\alpha = 0.05$

ARR, absolute relative risk; RR, relative risk; RRR, relative risk reduction.

DISCUSSION

Interpretation of Results

Only 2 of the 12 original placentas initially had positive test results for GBS, indicating that a low percentage of placentas are colonized with GBS even from mothers who had positive GBS test results during pregnancy. The encapsulation process of dehydrating the placentas reduces the GBS present on the placentas an additional 69.6%. When the 2 research questions are considered together, there is approximately a 5.1% chance that a placenta from a mother who had a positive test result for GBS during pregnancy will have a positive test result for GBS after the encapsulation process.

Research Findings

It was hypothesized that no placentas would have positive test results for GBS after birth because of the universal administration of prophylactic antibiotics given during labor.[14-16] It was found that a low percentage of placentas had GBS-positive test results after birth, but not all had GBS-negative results. However, the 83.3% negative results correlate with the 80% reduction in newborn-GBS infections seen in previous studies since the implementation of universal antibiotics.[15] Furthermore, it could not be confirmed that all mothers who donated their placentas had received prophylactic antibiotics during labor, which could account for the 2 positive results. The only other placenta study that has performed a microbiological analysis of placentas found all raw placentas in their study to have negative results for GBS.[1]

For the manually infected placentas, the ARR of 69.57% with $P < 0.0001$ represents a statistically significant decrease in risk. This suggests that the dehydration process significantly reduces the survival of GBS on placentas that were infected in the raw state, which supports this study's second hypothesis. Similarly, the microbiological

Table 1. Results for original-raw placentas, uncontaminated-raw and dehydrated placentas, and raw and dehydrated placentas manually contaminated with GBS

	Total	GBS (+) Raw	GBS (–) Raw	Percentage (%) GBS(+) Raw	GBS(+) Dehydrated	GBS (–) Dehydrated	Percentage (%) GBS(+) Dehydrated
Original placentas	12	2	10	16.7	/	/	/
Uncontaminated samples	9	0	9	0	0	9	0
Manual contaminated samples	46	46	0	100	14	32	30.4

GBS, group B *Streptococcus*; –, negative; +, positive.

study by Johnson et al[1] showed a significant decrease in microorganisms after dehydration.

This study was limited by the relatively small sample size of placentas and the unknown status of whether the mothers received antibiotics during labor. In contrast, the samples that were manually contaminated most likely had a greater amount of GBS on them than naturally colonized placentas, which could cause the number of placentas with positive test results for GBS after dehydration in this study to be falsely elevated. Active infection of the mother or baby at the time of birth is a contraindication of placenta encapsulation, so placentas with an infectious dose of GBS would not traditionally be consumed.[1]

The findings of this study are important because they provide empirical data rather than speculation about the risk of placentophagy to the mothers with GBS-positive test results and their newborns. Many physicians have been using a single case study to discourage mothers from encapsulating their placentas, even if the mother does not have a positive GBS test result.[4,11] A single incident may warrant further research but does not provide sufficient data to make sweeping recommendations to an entire community. This is especially true when larger studies show no adverse neonatal outcomes in women who encapsulated their placentas.[5] While the CDC case study of the newborn infected with GBS found GBS in the placenta capsules, the physicians could not rule out other modes of transmission, such as direct contact with family members who might be colonized with GBS or that the antibiotics may not have eradicated the initial infection and could not establish a route of infection from the placenta capsules to the newborn.[11] This study provides the much-needed follow-up data that allow physicians and new mothers to know the statistical risk of their placentas containing GBS after birth and after the encapsulation process.

CONCLUSION

This study was an attempt to discover if GBS is present in raw placentas of mothers with positive GBS test results after birth, and if the dehydration process used in placenta encapsulation is sufficient to kill GBS. This was done by testing the raw placentas, inoculating placenta samples with GBS, and testing them before and after dehydration. The results conclude that the percentage of raw placentas infected from the birth process is inherently low, and that the encapsulation process significantly decreases the presence of GBS in infected placentas. The hope is that this research is a starting point for more studies on the presence of GBS in placentas, such as a quantitative study of how much GBS is present in the raw and dehydrated placentas or how long GBS can survive in the dehydrated powder.

Placentophagy tends to be responded to by the media and medical community in a sensationalized, rather than an evidence-based, manner. As the CDC case study illustrates, medical professionals often use lack of data to make assumptions regarding placentophagy's role in patient outcomes without giving equal weight to other contributing factors. The results of this study are significant to the clinical treatment of mothers with GBS-positive test results who wish to encapsulate their placentas. With this study, women and their care providers will better understand the risk involved in consuming their placenta for postpartum wellness. This study is one of several recent and ongoing studies about the benefits and risks of placenta encapsulation. Expecting mothers and their care providers should take into account all the information—such as the reported benefits of encapsulation, the levels of hormones, toxic elements, microorganisms found in dehydrated placentas, and the comparisons of neonatal outcomes between women who encapsulated their placenta vs. women who did not—and use these studies to make informed choices about their postpartum care.[1,2,5,10]

APPENDIX 1. IRB APPROVAL LETTER

Institutional Review Board Office
Augusta University
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Email: IRB@augusta.edu
Phone: 706-721-3110
http://www.augusta.edu/research/irboffice/

DATE:	September 5, 2019
TO:	Melanie Belk, BS
FROM:	Augusta University (AU) Committee A
PROJECT TITLE:	[1444887-2] Presence of Group B Streptococcus in Raw and Dehydrated Placenta
SUBMISSION TYPE:	New Project (Response/Follow-Up)
ACTION:	APPROVED
APPROVAL DATE:	September 4, 2019
NEXT REPORT DUE:	September 4, 2020
REVIEW TYPE:	Exempt Review
REVIEW CATEGORY:	Expedited review category # 3 3- Prospective collection of biological specimens for research purposes by noninvasive means.

Thank you for your submission of Response/Follow-Up materials for this New Project. The Augusta University (AU) Committee A has APPROVED your submission. This approval is based on an appropriate risk/benefit ratio and a project design wherein the risks have been minimized. All research must be conducted in accordance with this approved submission.

This submission has received Exempt Review based on applicable federal regulations:
45 CFR 46 (DHHS), 2018 Requirements
This project has been determined to be a MINIMAL RISK project.

- The IRB has determined this project does not require continuing review per 45 CFR 46.109(f) based on the following circumstance:

  Research eligible for expedited review in accordance with 45 CFR 46.110;

  You are required to ensure an Annual Report is submitted for this project when the Project Annual Report notification is sent to you. Please refer to the Next Report Due date listed at the beginning of this letter. Additionally, you remain responsible for the Principal Investigator responsibilities for this project listed in this letter to include, submission of amendments, reportable events, and closures.

The approval includes the following documents:

- Consent Waiver - Waiver of consent documentation (UPDATED: 07/10/2019)
- Consent Form- Belk Information Letter.docx (UPDATED: 07/31/2019)
- Letter - Stipulation Response Letter Template.docx (UPDATED: 07/10/2019)
- Protocol - Belk, Irby Updated protocol 7/26 (UPDATED: 07/26/2019)

APPENDIX 2. IRB VERBAL CONSENT LETTER

Approval Date: September 4, 2019	IRBNet ID: 1444887-2	AUGUSTA UNIVERSITY IRB APPROVED

Augusta University

Dear xxx,

You are being invited to participate in a research project to study Group B Streptococcus in raw and dehydrated placentas because you tested positive for Group B Streptococcus during your pregnancy. This research project is funded by Augusta University. Should you choose to participate, we will collect your placenta at the hospital after you give birth. I will work with hospital staff to package your placenta and pick it up after the birth.

The results of this project will be used for my graduate research project. Through your participation I hope to understand whether the encapsulation process is sufficient to destroy the bacteria of Group B Streptococcus in placentas. I hope that the results of the study will be useful for pregnant women and I hope to share my results by publishing them in a scientific journal and sharing them with birth professionals to help their patients make informed decisions.

There are no known risks to you if you decide to participate in this study and your placenta will not be identified with you personally. There is no direct benefit to you for participating in this study. You will not be compensated for your participation in this research. The alternative would be not to participate in the study. I will not share any information that identifies you with anyone outside my research group which consists of me, my research partner Ariana Irby, and my faculty mentor, Giti Bayhaghi.

Your placenta will be tested for Group B Streptococcus, and these results will be stored on a spreadsheet on my computer. The researchers will retain this data for the duration of the project. The researchers will dispose of your placenta after it has been tested. The data from this study will be made available to other researchers for other studies following the completion of this research study and will not contain information that could identify you.

I hope you will allow us to use your placenta for this study. Your participation is voluntary and there is no penalty if you do not participate. If you have any questions or concerns about the use of your placenta, about being in this study, or to receive a summary of my findings you may contact me at 404-824-8530.

If you have any questions or concerns about the "rights of research subjects", you may contact the Augusta University IRB Office at (706) 721-1483.

Sincerely,

Melanie Belk
1000 Medical Center Blvd.
Lawrenceville, GA 30046

Version date: July 31, 2019

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