The ChorioAnchor: Design and Testing of a Novel Chorioamniotic Anchoring Device to Enable Percutaneous Fetoscopic Surgery

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Mini Summary

What does this study add to current knowledge?
- The study presents the design of the ChorioAnchor, a percutaneously delivered novel resorbable anchoring system for the chorioamniotic membranes for use in fetoscopic surgery. The study also describes the process of elucidating the functional strength requirements for this device and the testing conducted on the device.

What are the main clinical implications?
- Percutaneous fetoscopic surgery, while significantly less invasive than open in utero surgery, carries an increased risk of chorioamniotic membrane separation and preterm prelabor rupture of membranes (PPROM). ChorioAnchor is a percutaneously delivered device that is designed to reduce the risk of PPROM by anchoring the chorioamniotic membranes to the uterine wall and providing mechanical support to the membranes prior to uterine port placement.

Keywords
Preterm prelabor rupture of fetal membranes · Fetal surgery · Fetoscopy

Abstract

Introduction: Percutaneous fetoscopic surgery is hampered by an increased risk of preterm prelabor rupture of membranes (PPROM). Recent surgical techniques have shown that suturing the chorioamniotic membranes following laparotomy and uterine exteriorization is associated with a lower risk of PPPROM compared to percutaneous in utero surgery. This study presents the ChorioAnchor, a novel resorbable device that percutaneously anchors the chorioamniotic membranes to the uterine wall. Methods: Human factors testing and peel tests were used to simulate the worst-case in-use loading conditions, establishing the device strength requirements. Tensile testing was used to measure the time-zero strength of the device. Porcine cadaver testing was used to examine ultrasound visibility and acute handling.

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characteristics. Short-term host response was examined through an acute 7-day implantation study in a rabbit model. Results: With a time-zero tensile strength of 47 N, the ChorioAnchor exceeded the established 4 N strength requirement. Both the ChorioAnchor and delivery device were seen to be clearly visible under ultrasound imaging. Short-term host response to the device was well within the range expected for this type of device. Conclusion: The ChorioAnchor meets its engineering requirements in the early stages of implantation. Future studies will examine the kinetics of degradation of the device in vitro and in vivo.

Introduction – Clinical Background and Unmet Need

Congenital birth defects affect approximately 3% of babies born in the USA each year and remain a major cause of neonatal morbidity and mortality. While some congenital anomalies can be successfully treated after birth, there is evidence that in utero interventions for fetal spina bifida [1], congenital diaphragmatic hernia [2], lower urinary tract obstruction [3], severe pleural effusions, and twin-to-twin transfusion syndrome [4] improve pregnancy outcomes [1, 5]. Open fetal surgery is the current standard of care for in utero spina bifida closure [1]. This procedure involves a lower abdominal wall incision as well as a 6–8 cm incision in the uterus to gain access to the fetal back (Fig. 1a). Open fetal surgery is associated with significant long-term obstetrical risks including a uterine rupture rate of almost 10% in subsequent pregnancies [6] and mandatory cesarean sections for all subsequent deliveries [1, 7, 8].

Fetoscopic surgery is a less invasive procedure performed through 3–4 mm laparoscopic cannulas inserted into the uterus through which low-profile tools and fetoscopes are introduced into the uterine cavity to perform the surgical intervention typically under ultrasound and direct visualization (Fig. 1b). Fetoscopic surgery has been adopted for several in utero interventions including meningomyelocele closure [9–11], enabling a longer gestation period for in utero development of the fetus and an increased likelihood of vaginal birth [5, 9, 10, 12]. However, the adoption of fetoscopic surgery is hindered by preterm prelabor rupture of membranes (PPROM). Often described as the “Achilles heel of fetal surgery,” PPROM can lead to pregnancy complications including preterm delivery and ascending infection [13]. Preterm deliveries associated with PPROM are also associated with a higher likelihood of major neonatal morbidities, including necrotizing enterocolitis, respiratory distress syndrome, and bronchopulmonary dysplasia [14]. Chorioamniotic separation is a key failure mode preceding PPROM [15], attributed to the insertion and manipulation of surgical instruments, which can detach the chorioamniotic membranes. The membranes are considered to be largely avascular, making reattachment and healing processes difficult to naturally achieve.

To reduce the risk of PPROM, fetal surgeons have piloted a method for fetal repair involving suturing the chorioamniotic membranes to the uterine wall around each operative port to preserve the mechanical apposition of the chorioamniotic membranes (Fig. 1c) to the decidual uterine surface. The sutures are placed in a box-like configuration (box stitch) around each fetoscopic port site prior to surgical port insertion into the amniotic cavity [16]. This method, applied during spina bifida closure procedures, is associated with lower rates of PPROM than those reported in percutaneous fetal spina bifida closure [17] and has led to a lower rate of premature delivery [10, 12, 16] than the reported rates for open [1] or percutaneous spina bifida closure [17]. However, this box stitch method requires exteriorization of the uterus following a large abdominal wall incision in order to provide access to the uterus for the placement of the box stitch. Laparotomies are associated with increased risk for bleeding or infection and a longer hospital stay, and thus there has been an interest in developing a membrane-securing method that would not require exteriorization of the uterus, particularly when the placenta is located in the posterior aspect of the uterus.

In some fetal surgery centers, fully percutaneous fetoscopic surgeries have been performed without membrane stitching (Fig. 1b); these procedures are accompanied by PPROM rates ranging from 80% [18] to 100% [17] with percutaneous fetoscopic spina bifida closure. In contrast, the box stitch method is associated with a PPROM rate of 26.7% in laparotomy-assisted fetoscopic spina bifida closure [16]. Thus, in order to secure the membranes during fetoscopic surgery while avoiding the complications associated with laparotomies and uterine exteriorization, there is a need for a novel percutaneous device for anchoring the chorioamniotic membranes.

In this paper, we describe the design, prototyping, bench testing, and pilot in vivo evaluation of ChorioAnchor™, a degradable polymeric fixation device that anchors the chorioamniotic membranes to the uterine wall via a percutaneous deployment system. The use of this device is expected to be associated with lower rates of PPROM and premature labor, thus rendering in utero, percutaneous fetal surgery safer for mothers and their unborn babies.
Design

Design Concept

The ChorioAnchor is composed of two cylindrical anchors connected by a loop of suture with a locking-sliding knot (e.g., modified Meltzer knot, Weston knot, etc.) that can be tightened to close the separation between the anchors (Fig. 2a). The device is deployed through a delivery tube that ejects the distal anchor inside the amniotic cavity under ultrasonography guidance and the proximal anchor outside the uterine wall under direct visualization with the use of commercially available optical trocars. A knot pusher is then used to tighten the locking-sliding knot, approximating the chorionic membrane to the uterine wall (Fig. 3). ChorioAnchors are deployed around the fetoscope insertion site to provide mechanical support to the chorionic membranes prior to vascular cannula placement during percutaneous in utero surgery (Fig. 2b). By mimicking the support pattern of the box stitch currently used in laparotomy-assisted fetoscopic spina bifida closure, the device allows fetal and pediatric surgeons to leverage their experience and familiarity with the box stitch when using the ChorioAnchor. The implant is designed to be resorbable and fully degrades after its functional lifespan.
Clinical immersion and interviews with fetal surgeons (J.E., M.A.B.) were conducted to develop the engineering requirements for the anchor device. The anchor spacing needs to be long enough to facilitate delivery across the thickness of the uterine wall at the time of the procedure (6–12 mm [19]). The device needs to withstand the forces of delivery and the maximal forces required to approximate the chorioamniotic membranes against the uterine wall. In addition, it needs to maintain adequate mechanical strength for the remaining duration of pregnancy post-surgery (at least 20 weeks). This must be balanced against the requirement that in the post-delivery setting, the device must fully degrade in the shortest possible time to ensure restoration of normal uterine wall morphology.

**Anchor Design and Materials**

To aid in pivoting the anchors to lie flat against the uterine wall post-ejection, the cylindrical anchors have a high aspect ratio (1.8 mm diameter; 10 mm length) and a...

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*Fig. 3.* ChorioAnchor deployment sequence. **a** The delivery tube is introduced into the amniotic cavity. **b** The distal anchor is ejected. **c** The delivery tube is withdrawn into the space between the uterine and abdominal walls; the suture pull induces a moment rotating the anchor against the uterine surface. **d** The proximal anchor is ejected. **e** The pusher tube is used to advance the knot approximating the chorioamniotic membranes to the uterine wall.

*Fig. 4.* **a** Anchor component features. **b** The suture path through the ChorioAnchor aids in rotating the anchor flat against the uterine wall. **c** Fabricated ChorioAnchor made from PLG 8218 and TephaFLEX P4HB suture.
suture path that generates a couple force system to enable rotation of the anchor when pulled (Fig. 4b). Early testing showed that the portion of suture that is routed adjacent to the anchor needed to be constrained as it was pushed through the delivery tube in order to avoid bunching of the suture and the resultant jamming of the device assembly. The anchors are hence engineered to fully occlude the delivery tube lumen and feature longitudinal slots running the length of the anchor to constrain/contain the suture during deployment (Fig. 4a). Rounded atraumatic tip features are incorporated in the anchors to minimize potential harm to the uterine wall, surrounding organs, chorioamniotic membranes, or the fetus (Fig. 4a). The anchors are made of a resorbable copolymer of L-lactide and glycolide (Corbion PLG 8218) chosen for its degradation time of 12–18 months. The suture is made from poly-4-hydroxybutyrate (P4HB), a flexible polymer that retains 20–30% of its mechanical strength in the 20–25-week window and substantially degrades within 64 weeks [20, 21]. The suture is knotted with a modified Meltzer sliding knot that can be advanced with a knot pusher to cinch the device.

Delivery Device Requirements
The delivery device needs to have sufficient length to reach the uterine cavity in a typical patient at the 20th to 24th weeks of pregnancy. Beck et al. [22] reported an increase in the risk of PPROM with an increase in the size of the instrument that penetrates the uterine cavity. Hence, the diameter of the delivery system shaft that achieves access to the uterine cavity via penetration of the uterine wall must be minimized to reduce the risk of PPROM. Nevertheless, mitigating this risk is the fact that the sites where the delivery instrument penetrates the membranes are also the regions with maximal mechanical support provided by the ChorioAnchor. Through conversations with fetal surgeons (J.E., M.A.B.) and published data on the risk of PPROM, a maximum outer diameter of 2.11 mm (14 Birmingham gauge) was established as a requirement for the delivery system. The delivery device needs to be able to penetrate the abdominal and uterine walls with minimal tissue trauma and minimal risk of harming the fetus. The design of the delivery device must ensure that each anchor is deployed separately in the desired anatomic position (chorioamniotic-uterine junction and serosa-uterine junction), as any inadvertent ejection of an anchor in the wrong location would result in the need for an extended surgical procedure to retrieve the anchor device. One of the challenges when sutures are used to anchor the chorioamniotic membrane in laparoscopic-assisted fetoscopic spina bifida closure is that a curved suture needle is hard to visualize by ultrasonography making it difficult to identify when the needle has reached the uterine cavity. Therefore, the delivery device must have a linear design for the entire device to be visualized by ultrasonography in order to improve the safety of percutaneous in utero procedure.

Prototype Delivery Device Design
The delivery device (Fig. 5) is composed of concentric tubes that contain the ChorioAnchor, advance the anchors into position, and actuate the locking-sliding knot. Atraumatic or minimally traumatic access is facilitated using a sharp obturator to penetrate the abdominal and uterine walls to position the delivery tube within the uterine cavity under ultrasound guidance. The delivery tube is constructed with thin-walled 316L stainless steel enabling maximizing of the inner diameter (and consequently, anchor size) while keeping the outer diameter and hence the maximum size of all penetrating elements at 2.11 mm. Future design iterations will aim to reduce
this diameter further, but such efforts are limited by the mechanical strength required of the anchoring device. After positioning the delivery tube, the obturator is removed leaving only the blunt-tipped delivery tube in the uterine cavity, which eliminates the possibility of accidental harm to the fetus from the sharp obturator. The ChorioAnchor is loaded into an anchor tube which is mated with the delivery tube and locked in position. The central pusher tube then ejects the distal anchor into the amniotic cavity. The pusher tube and anchor tube have a peg and slot system with the lengths of these features tuned to provide the mechanical logic system that ensures full ejection of each anchor separately with each actuation. The delivery tube is then retracted outside the uterus, and the second anchor is deployed under direct visualization facilitated via a commercially available 5 mm optical trocar. The pusher tube is then used to advance the locking-sliding knot, thereby approximating the chorioamniotic membranes to the uterine wall. The prototype delivery device featured an all-metal construction to facilitate repeated cleaning and autoclave sterilization. Production devices will likely use a combination of plastic and metal components.

**Device Testing – Methods**

**Device Bench Testing**

The maximum strength needed for the ChorioAnchor was established by measurement of the forces experienced by the device in two loading scenarios. In the first scenario, we measured the force applied by the surgeon when tightening the suture knots in the device during delivery. This surgical maneuver represents the initial peak when tightening the suture knots in the device during delivery. Measurement of the forces applied by surgeons was accomplished on a test bed designed to measure the compressive forces applied between two layers of soft silicone (Smooth-On Dragon Skin™ FX-Pro™; Shore Hardness: 2A) soaked in soap water, using a 2 kgf load cell (LC61SP-2KG; Omega Engineering Inc.) connected to a bridge amplifier and meter (DPS20; Omega Engineering Inc.). The hardness of the silicone layers was chosen by having surgeons use the delivery device obturator to penetrate silicone test phantoms with a range of hardness values and select the phantom that best matched the feel of the uterine wall. Three fetal surgeons at Texas Children’s Hospital were trained to deploy the ChorioAnchor across the two silicone layers using the prototype delivery device and tighten the locking-sliding knot while simulating the anticipated surgical procedure. The surgeons also placed a stitch across the two silicone layers replicating one leg of the box stitch currently used to mitigate the risk of PPROM [12]. The compressive load applied between the two layers is reacted by the tensile load in the stitch loop or ChorioAnchor. The steady state load after deployment of the ChorioAnchor or stitch was recorded with each surgeon completing 3 deployments of each. A Student's t-test was used to compare the applied compressive load between the stitch and ChorioAnchor groups.

**Chorioamniotic Membrane-Device Peel Strength**

Measurement of the force required to separate the membranes from the uterine wall was accomplished through a peel test. Third trimester bovine amniotic membranes and 32mm wide collagen casing (LEM Products) were used as analogs for the chorioamniotic membranes. The bovine amniotic membranes and collagen sheets were subjected to tensile testing matching the protocol used by Gremare et al. [23] to measure their relative strength compared to human amniotic membranes. Soft silicone (Smooth-On Dragon Skin™ FX-Pro™; Shore Hardness: 2A) with hardness chosen as before to best represent the uterine wall was used to cast a 15 mm thick block to serve as the uterine wall analog. A composite test phantom comprising the silicone block (uterine wall analog) and the bovine membrane/collagen casing (chorioamniotic membrane analog) was used for all peel force testing of membrane-device constructs (Fig. 6a, b). ChorioAnchor devices were deployed across the test phantom and fixtured to a mechanical test frame (MTS Insight 1) as shown in Figure 6. The membrane analog was clamped in the upper jaw of the test frame, and the fixture was aligned such that the membrane was parallel to the test
frame axis. The ChorioAnchor was aligned to ensure that the peel front contacts the anchor over the long edge to generate larger detachment forces resulting from the larger contact area. This configuration represents the worst-case scenario in terms of forces on the ChorioAnchor. The membrane was pulled upward at 10 mm/min while recording load and test frame crosshead position at 100 Hz until failure of the membrane analog was observed. The test was conducted 5 times each using bovine amniotic membranes and collagen sheets as the chorioamniotic membrane analog. The test was also repeated with the membrane analog anchored by a 2 cm stitch placed with 3-0 Monomax suture tied with a double overhand knot to replicate the current surgical standard for anchoring the membranes prior to fetal surgery [12]. Student’s t-tests were used to compare the failure load between the ChorioAnchor and the stitch. It is important to note that the current test phantom does not model the fusion between the chorioamniotic membrane and uterine wall. As a result, the model does not mimic the force to separate

Fig. 6. Chorioamniotic membrane-device peel strength test. a Test configuration schematic. b Peel test in-progress evaluating the ChorioAnchor using a collagen sheet as the chorioamniotic membrane analog. c Representative load-displacement curve indicating failure point.

Fig. 7. Anchor mechanical strength testing. a Test setup photograph. 3D-printed fixtures are used to connect the anchors to the mechanical test frame. The fixtures center the anchors, aligning the device axis with the test frame axis while providing a path for the suture loop between the anchors. b Representative load-displacement curve indicating failure point.
the 2 layers, but the results represent the isolated peel strength of the anchor compared to the box stitch as the current control, also evaluated under the same conditions, i.e., no membrane adhesion to uterine wall.

ChorioAnchor Mechanical Strength
The mechanical strength of the ChorioAnchor was measured through a pull-apart test loading the device in tension and replicating the physiologic load condition. The anchors were connected to a mechanical test frame (MTS Insight 1) through 3D-printed fixtures that self-center the anchors when loaded and provide a path for the suture (Fig. 7). The devices were then pulled apart at 5 mm/min until failure while recording load and crosshead position at 100 Hz. Five ChorioAnchors were tested, recording peak load and failure mode.

Usability and Ultrasound Visibility Testing
Usability and ultrasound guidance capability of the ChorioAnchor and delivery system were evaluated in a porcine cadaver bladder model. Briefly, a fetal surgeon was trained in the use of the ChorioAnchor and delivery system and asked to deliver two ChorioAnchors in the porcine bladder under ultrasound guidance (Fig. 8a). Simulating current surgical access techniques, a small incision was made in the top layer of skin to facilitate entry with the obturator, with the rest of the delivery procedure performed as designed. The surgeon was asked to provide feedback on the usability of the device. Ultrasound images were captured to examine the visibility of the delivery device and anchor.

Short-Term Host Response Pilot Testing
Short-term tissue response to the ChorioAnchor device was evaluated in a pilot open-surgical in vivo study in a pregnant rabbit model under a research protocol approved by Baylor College of Medicine Institutional Animal Care and Use Committee (AN-7503). This study was conducted prior to final material selection with the anchors being injection molded from Corbion PG S (polylactic acid), and the suture loop fabricated with Ethicon PDS II (polydioxanone). The Corbion PG S is similar in chemistry and degradation products to the PLG 8218 used in the final device design and bench studies. This material was chosen at a stage in the project when the desired degradation profile was not known, and an early preclinical pilot was needed to provide insights into the tissue response to an implant. Future studies have since then focused solely on the final chosen PLG 8218 and P4HB suture materials.

Device Testing – Results

Device Bench Testing
Deployment Tension
Surgeon-applied tension on the ChorioAnchor was measured at 1.4 ± 0.6 N with the corresponding load for the stitch measured at 0.6 ± 0.2 N. Loads applied with the stitch were significantly lower ($p = 0.004$) than those applied by the ChorioAnchor.
Fig. 9. a ChorioAnchor in rabbit uterus at necropsy. b Histology section locations. c Normal uterine serosa (left); minimal reactive fibroplasia in serosa apposed to anchor (right, black ink). d Normal endometrium (left); attenuation of endometrial glands at anchor site (right, arrow). e Normal uterine wall thickness (left); 60% reduction in wall thickness at anchor site (right). f Foreign body reaction at suture track (note multinucleate giant cells; arrow indicates suture track perpendicular to uterine wall). g Focal pressure necrosis, center of anchor at suture track.
Chorioamniotic Membrane-Device Peel Strength
Tensile testing measured the failure load for the collagen sheets at 3.6 ± 0.8 N and the bovine amniotic membranes at 3.5 ± 1.7 N. Peak forces to separate the membrane analog from the uterine wall analog are summarized in Table 1. When approximated by the ChorioAnchor, failure was seen to originate at the hole in the membrane through which the anchor was delivered. When anchored by the stitch (clinical control), failure was seen to originate at the two insertion holes (needle entry and exit) for the stitch. Failure forces were not significantly different between the membrane analog anchored by the stitch and those anchored by the ChorioAnchor (p > 0.05).

ChorioAnchor Mechanical Strength
The tensile load to failure for the ChorioAnchor device was measured at 46.7 ± 7.7 N. Failure in all samples occurred in the suture loop connecting the anchors while the anchor body was intact in all these tests.

Usability and Ultrasound Visibility Testing
Testing on the porcine cadaver model verified the ability of the prototype delivery system to deploy the ChorioAnchor percutaneously across internal body layers. The distal end of the delivery tube was seen to be clearly visible under ultrasound (Fig. 8b). In addition, the anchors were also easily visualized, allowing the surgeon to confirm deployment of the anchors through ultrasound imaging. Surgeons were able to use the prototype delivery device with minimal training or practice but reported some degree of dissatisfaction with the ergonomics of the device, specifically with the large initial motion required to move the anchors from the anchor tube to the tip of the delivery tube as the first anchor is ejected. This human factors/ergonomics issue was noted as a desirable area of focus for future optimization of the delivery system design.

Short-Term Host Response Testing
Histologic evaluation showed that the ChorioAnchor was well tolerated with levels of inflammation within the range expected of implanted sutures used for uterine wall repair. Specifically, tissue reaction was minimal to mild in all cases. On the external surface of the uterus, the serosa apposed to the anchor showed minimal reactive fibroplasia (Fig. 9c) with a slight increase in number and size of fibroblasts. On the mucosal surface, some attenuation of endometrial glands was noted (Fig. 9d). Mild to moderate attenuation of the uterine wall was noted in all cases, presumably due to compression (Fig. 9e) which ranged from 5% to 60% of the original uterine wall thickness measured adjacent to the area of compression. At the centers of the anchors, where suture material penetrated the uterine wall, minimal to mild inflammation and tissue reaction were noted, typically granulomatous/foreign body type (Fig. 9f). The level of tissue damage and inflammation was within the range expected for any implanted suture. At the center of the anchors, where suture penetrated the uterine wall, there was evidence of focal pressure necrosis in 2 out of 5 anchors (Fig. 9g). These lesions were focal and limited to the sections immediately adjacent to the suture tracks.

Discussion/Conclusion

Device Bench Testing
Deployment Tension
Measuring surgeon applied forces using the stitch and ChorioAnchor showed that surgeons apply greater loads on the tissue with the ChorioAnchor than with the stitch. This is most likely a result of surgeon familiarity with the box stitch technique as well as the more direct tactile feedback available during conventional suturing. Nevertheless, these test results enabled the development of a key engineering requirement for the strength of the anchoring system in terms of the peak tensioning forces generated during placement. These tests indicate that from the perspective of loads applied at the time of deployment, the device should withstand a peak tensile force of at least 3.2 N (mean ± 3 standard deviations). While this is the engineering design requirement, it is anticipated that with training, surgeons will develop the tactile feel that will allow for lower tensioning forces to be generated similar to the current box stitch. Deployment tension directly impacts the extent of uterine wall compression and the associated risk of pressure necrosis.

Chorioamniotic Membrane-Device Peel Strength
Tensile testing of the uterine membrane analog measured the failure load for the collagen sheets at 3.6 ± 0.8 N.

Table 1. Peel test failure loads for two chorioamniotic membrane analog (collagen sheet and bovine amniotic membrane) anchored by the ChorioAnchor and stitch methods; n = 5 for each group

| Failure load (mean ± standard deviation) (N) | ChorioAnchor | Stitch |
|---------------------------------------------|-------------|--------|
| Collagen sheet                              | 6.8±3.0     | 8.0±3.1 |
| Bovine amniotic membrane                    | 3.4±1.0     | 4.2±1.6 |

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N and the bovine amniotic membranes at 3.5 ± 1.7 N. Gremare et al. [23] reported the strength of human membranes as 0.8 ± 0.4 N, indicating that our chosen analog for the peel test would report failure loads more than four times greater than would be seen in practice.

The data from the peel tests show no difference in anchoring ability between the stitch and the ChorioAnchor, on both collagen sheets and the bovine amniotic membranes. The measured loads from this study enabled the development of the engineering requirement for the strength of the anchoring system in terms of the peak tensile load the device would experience as it anchors the membrane to the uterine wall. At loads greater than this level, the chorioamniotic membranes will have ruptured. With the greater of the two peel failure loads for the ChorioAnchor measuring at 6.8 ± 3.0 N and given that our test membrane analogs fail at forces more than 4 times greater than human membranes, the requirement for the peak tensile load the anchor must withstand during its functional period was established as 4 N (\[\text{mean} + 3 \text{ standard deviations}\]/4). Given that this force is greater than the result resulting from the deployment tension test, 4 N was established as the minimum tensile strength requirement for the ChorioAnchor.

ChorioAnchor Mechanical Strength

With a tensile failure load of 46.7 ± 7.7 N, ChorioAnchor significantly exceeds the engineering requirements for strength with a significant factor of safety. This testing was conducted on freshly molded (undegraded, time zero) material, and while failure always occurred in the suture component at time zero, this mechanism could shift to the anchor component as degradation progresses. While this study was limited to the time-zero properties of the device, future studies will assess the anchor component through its degradation lifespan examining changes to its strength, molecular weight, and mass. Poly-4-hydroxybutyrate (P4HB) sutures are known to have a strength retention under degradation of 20–30% in the 20–25-week window [20, 21]. Applying this reported percentage strength retention to the measured strength of the ChorioAnchor, it can be expected that the suture component will exceed the 4 N strength requirement through the functional life of the device until childbirth. This will be further verified through future degradation studies. It is also of note that the current box stitch is performed with the P4HB suture of similar diameter, with acceptable outcomes for reduced PPROM rates [16]. Based on this historical clinical evidence, it is reasonable to expect equivalent outcomes for the ChorioAnchor device which uses the same P4HB as the primary load-bearing mechanism.

Usability and Ultrasound Visibility Testing

Ultrasound visibility was identified as a key requirement for the delivery device, and the prototype delivery device was seen to be clearly visible in its entirety under ultrasonography, which in addition to ensuring proper positioning of the ChorioAnchor, is also critical in ensuring maternal and fetal safety when using the delivery device. The prototype device has room for improvement, with one of the primary critiques received being with the ergonomics of the device. Design approaches have been identified that can significantly shorten the actuation distance to eject the anchors and will be evaluated in future revisions. Another necessary avenue of refinement of the delivery system is the need to incorporate a mechanism that creates a space between the abdominal wall and the external uterine surface where the proximal anchor may be deployed, potentially under direct visualization (e.g., through a 5 mm optical balloon trocar), and freely rotated into position.

Short-Term Host Response Testing

The short-term rabbit model showed that while the implant was well tolerated, focal pressure necrosis was observed in 2 out of 5 anchoring sites adjacent to the suture tracks. This is likely due to the fact that in pregnant rabbits near term, the uterine wall is very thin and may be more susceptible to pressure than the uterine wall of a larger animal species. Moreover, in the pregnant rabbit model it is difficult to observe signs of tissue ischemia such as "blanching," which is frequently visualized in the human uterus when sutures are overtightened. Nonetheless, this highlights the need for the delivery system to provide good tactile feedback of the suture loop tension and, if possible, adjunctive direct visualization of the uterine surface to allow surgeons to accurately control the pressure applied to tissue captured by the ChorioAnchor. While this study involved only two animals, it was performed to provide early insight into the short-term inflammatory response while simulating the surgical technique to inform the acute handling of the device and related design changes. As such, the study did not justify the use of a large number of animals.

The conventional approach to performing in utero open spina bifida requires the use of commercially available surgical staplers, which help in extending the hysterotomy while achieving hemostasis and membrane affixation to the uterine wall. However, the use of absorbable
surgical staples leaves a much larger amount of foreign material in comparison with the ChorioAnchor anchors. Histologic evaluation by Ochsenbein-Kölble et al. [24] of 25 cases with stapled hysterotomy scars excised at the time of cesarean delivery following open spina bifida closure demonstrated that the intact layer of myometrium was less than 1 mm thick in 56% of cases. Moreover, the authors of this study reported myometrial necrosis adjacent to the scar as well as chorioamniotic membrane necrosis in 41% (9/22) of excised stapled hysterotomy scars [24]. This is analogous to the pressure necrosis observed in some anchoring sites adjacent to the suture tracks in our pregnant rabbits, but at a much larger scale given the larger dimensions of the staples compared to the Chorio-Anchors.

While this study illustrated a positive short-term response to the device, longer term bench (in vitro) and in vivo studies with a larger animal model such as pregnant sheep are needed to examine the degradation profile and by-products (in vitro), and the in vivo response past the acute inflammatory stage to determine the long-term uterine tissue response to the ChorioAnchor device. Future in vitro studies should evaluate the kinetics of degradation, i.e., mass loss, molecular weight changes, and strength reduction via testing in appropriate media under physiologic pH and temperature conditions. These in vitro studies are currently in progress and will be reported shortly in a forthcoming publication. Similarly, in vivo studies should study the inflammatory and healing response during the degradation of the ChorioAnchor device polymers (PLG 8218 and P4HB). Unfortunately, there is a lack of physiologically relevant nonprimate animal models for testing devices such as the ChorioAnchor since these mammals do not exhibit amnion and chorion fusion [25, 26]. Artificially anchoring the amniotic membranes with the device in these animal models would result in a loading scenario unlikely to be seen in human use of the device and may cause pregnancy complications in the animal. In addition, in sheep models, the presence of cotyledons restricts the available area for access to the amniotic cavity [26]. Fortunately, the ChorioAnchor device uses currently used suture materials for the primary transmural fixation/tensioning, along with very well-characterized biodegradable polymers (PLG 8218) with a long history of clinical use for the cylindrical anchor. Therefore, established nonprimate models such as a pregnant sheep would still be very useful in generating host response data with respect to the implant in contact with uterine tissue. Our future studies will focus on longer term evaluation of the host response to ChorioAnchor in a pregnant sheep model while using direct visualization to avoid local ischemia and ultrasound to identify safe regions between cotyledons for anchor deployment. This will allow us to evaluate the device through 10 weeks of gestation.

Future testing is also needed to evaluate the biological safety profile of the implant in terms of risks to both mother and fetus. When using ISO 10993 standards [27] to evaluate the biocompatibility of the ChorioAnchor, as well as preclinical studies directed at evaluating safety and efficacy, consideration will be given to the fact that while the device is in contact with maternal tissue throughout the life of the device, any degradation products released into the amniotic cavity will likely interact with and/or be ingested by the fetus. The anchors being comprised of poly(lactico-co-glycolic acid) will biodegrade into lactic and glycolic acids which have known pathways for metabolism and excretion [28]. While the expected degradation timeline of 12–18 months for PLG 8218 is fairly long, other materials considered for the anchor had degradation times that were too fast (polyglycolides – less than 1 month) or too slow (polycaprolactones and poly D/L-lactides – several years). It is also to be noted that lactic-glycolide co-polymers are used in commercially available staples such as the Medtronic Premium Poly CS™ Staplers, which are currently used in open fetal surgery to provide hemostasis and mechanical support to the chorioamniotic membranes [1]. These staplers leave behind a much larger mass of polymer compared to the ChorioAnchor. Anecdotally, in the clinical experience of JE and MB, after open fetal surgery using these staplers, the mass of polymer left behind can be felt under palpation during cesarean delivery. Yet, there is no evidence that the amount of foreign body left by these absorbable staples is associated with reduced fertility in the future [29]. Furthermore, the retained material has not been shown to pose a significant risk to the mother. With the significantly lower mass of retained polymer when using the ChorioAnchor, we expect similar or better outcomes. The suture, extruded from poly-4-hydroxybutyrate, biodegrades into 4-hydroxybutyrate which has known metabolism and excretion pathways [21]. While the timeline for substantial degradation of this suture is around 64 weeks, no other suture currently available is able to maintain the required mechanical strength over the remaining duration of pregnancy post-surgery (at least 20 weeks). For comparison, polydioxanone sutures show a 75% loss in tensile strength in only 6 weeks [30]. Additionally, the suture used is the same fiber that comprises the commercial Monomax suture that is currently used in the box

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chorionic membranes during laparotomy-assisted fetoscopic spina bifida closure and is cleared for clinical use by both the US FDA and EU regulatory agencies [20].

In summary, the ChorioAnchor device as designed meets all the engineering requirements in the early stages of implantation (acute response). Future work and publications will explore the ability of the device to meet the longer term requirements as the device degrades through its functional lifespan. If the device is proven in these future studies to meet the safety and efficacy requirements over the desired 20-week window with a safe degradation profile after childbirth, the ChorioAnchor could become a novel solution to expand the usage of percutaneous in utero surgery. Clinical studies will be required to determine if the use of ChorioAnchor in percutaneous fetal surgery is associated with lower rates of chorioamnion separation and PPROM, which could potentially translate into better pregnancy outcomes following percutaneous in utero surgery.

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Statement of Ethics

The research reported here does not include human subject data. The short-term implant study protocol was approved by the Institutional Animal Care and Use Committee, Baylor College of Medicine (Protocol Number AN-7503).

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Conflict of Interest Statement

Achu G. Byju, Michael J. Heffernan, Michael A. Belfort, Jimmy Espinoza, and Balakrishna Haridas are inventors on the patent around this device. Christine Luk and Michael J. Heffernan are employed at and own shares in Fannin Innovation Studio who are working to commercialize this device. Chester J. Koh has previously consulted with Intuitive Surgical and has received grants from Pfizer, Allergan, and Olympus. Balakrishna Haridas is a consultant for 3M, Johnson & Johnson (J&J), Sentire Medical Systems, and Shape Memory Medical, holds stock in Shape Memory Medical, and has received grants from J&J and Shape Memory Medical.

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Author Contributions

Achu G. Byju, Jimmy Espinoza, Balakrishna Haridas, Michael A. Belfort, and Michael J. Heffernan contributed to the design of the device. Achu G. Byju, Ashley Diemer, and Balakrishna Haridas led device testing activities. Brian W. Simons performed the histologic analysis of the short-term implant study. Jimmy Espinoza, Michael A. Belfort, and Chester J. Koh provided clinical insight on device design. Achu G. Byju, Christine Luk, and Balakrishna Haridas drafted the manuscript. All authors contributed to the editing of the document.

Data Availability Statement

All data generated or analyzed during this study are included in this article. Further inquiries can be directed to the corresponding author.
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