Peer Review File

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Reviewer A

Comment 1. How many days did it take for the newly biodegradable stent to be completely degraded? The authors should show the changes of degrading rates and the fluoroscopic images.

Reply 1: According to our follow-ups between 5 and 6 weeks 91.7% of the BraidStent®-H were completely degraded. Only one of the BUS had not fully degraded and it was at the next follow-up (at 12 weeks) that complete degradation was demonstrated. The BraidStent® is designed to degrade between 4 and 6 weeks.

Changes in text: On the reviewer's advice we have included new images of BraidStent®-H degradation by ureteroscopy. Figure 4.

Comment 2. Did the authors observe the changes of BUS hydrated fragments in urine analysis?

Reply 2: Indeed, during the 3-week and 6-week follow-ups, the floating fragments of the stent can be seen in the urine collected for analysis. This allows us to measure them and check that their dimensions are very small.

Changes in text: No changes.

Comment 3. The authors should describe the changes of blood chemistry and urine analysis in detail.

Reply 3: Thank you for your comment. The blood biochemistry values have not been included because there are no changes and the amount of data is too large to include. The urinalysis values are included in Table 1, the asymptomatic bacteriuria and we have included in Table 1 another row where the urinary pH changes throughout the different phases of the study are included. This is the parameter of greatest interest with respect to urinalysis in the evaluation of a BUS.

Changes in text: Table 1 (pH parameters). A sentence is included stating that there are no significant changes throughout the study. Page 8.

Comment 4. The Internal ureteral diameter usually changes according to the ureteral location. Which location of the ureter did the authors measure?

Reply 4: In this study, site identification is very easy, as we always assess the lesion area (the stricture) and its progression after endoureterotomy. It always

presents a different morphology due to the stricture, its treatment and follow-up. Since the morphology is altered by the treatment. In addition to the fact that at the beginning of the study its location and the relationship with the lumbar vertebrae is measured exactly.
Changes in text: NA.

Comment 5. How did the authors evaluate the ureteral peristalsis? The authors should describe the median data with IQR.
Reply 5: We assessing ureteral peristalsis through Excretory Urography. We follow this protocol to favor ureteral peristalsis visualization: assessment is performed by means of continuous videourography (C-arm) and the number of waves/minute is counted. Bowel are always prepared 24 hours before surgery in order to remove waste and urinary bladder is emptied to favor the pressure gradient between renal pelvis and urinary bladder. Consequently, and given the fact that this study stage takes a long time and there is exposure to ionizing radiation, all assistants leave the OR and the operator works behind a mobile X-ray barrier, which we fortunately have in our experimental OR.
To the second question, the values expressed in Table 1 with respect to peristalsis are expressed in % of animals that showed ureteral peristalsis. We consider that this is the clearest way to assess the results of this study, as in some cases only 50 or even 0% of the animals showed peristalsis and expressing these values by waves/minute may lead to confusion among the readers. The purpose of the assessment of peristalsis is to describe whether or not peristalsis occurs, the quantification of peristalsis waves is, in our opinion, not so relevant. Although such data is available as it is measured at each follow-up
Changes in text: No changes.

Comment 6. The authors stated that the pathological study of ureteral wall at the endoureterotomy area showed between-group statistical significance in the "lamina propria fibrosis" and "serosal alterations" parameters. The authors should describe the reasons as well. Why did the BUS affect to better ureteral heeling after endoureterotomy?
Reply 6: Thank you for your comment. A comment to this regard has been added in the Discussion of the manuscript.
The reasons that could explain this difference in the better healing caused by the BraidStent®- H are mainly based on several factors: Firstly, the length of time is that although both stents remain in place for 6 weeks, the BraidStent®-H is undergoing continuous degradation, which is more evident during the fourth and

sixth weeks, decreasing its effect on the incised ureteral wall compared to the standard ureteral stent. This should result in less friction with the ureteral wall, causing less oedema and less compressive effect on the ureter section that is second intention healing. Also, as the BraidStent®-H does not show any VUR, this should favour ureteral healing, unlike the standard ureteral stent group.
Changes in the text: Included in the Discussion of the manuscript (Page 12).

Reviewer B
Comment 1: Biodegradable ureteral stent (BUS) is an attractive idea. But, the material by which BUS is constructed is very important to keep the patients safe. Are the co-polymer A and B suitable to human body??
Reply 1: Both polymers are biocompatible and have already assessed their biocompatibility in patients, as they are used daily in surgery as suture materials.
Changes in the text: "Two biocompatible and biodegradable copolymers with different degradation rates were used". (Page 5)

Comment 2: In methods, the author curried out the retrograde endoureterotomy for ureteral stricture by using laser. The setting is 1.2J-10Hz. Why did the author choose this setting? High J might become critical problem for ureteral mucosa, which cause the recurrent stricture.
Reply 2: We understand the reviewer's concern. But we use standard settings for Holmium laser endoureterotomy, as described in a large number of scientific papers.
-(1J-10Hz.). *Hibi H, Ohori T, Taki T. Long-term results of endoureterotomy using a holmium laser. Int J Urol.2007;14:872–874.*
-(0.8-1.2-8-12Hz). *Gdor Y, Gabr AH, Faerber GJ, Wolf JS Jr. Holmium:yttriumaluminum- garnet laser endoureterotomy for the treatment of transplant kidney ureteral strictures. Transplantation.2008;85:1318–1321.*
-(1.2J-10Hz). *Lin CM, Tsai TH, Lin TC, Tang SH, Wu ST, Sun GH, Cha TL. Holmium: yttrium- aluminum-garnet laser endoureterotomy for benign ureteral strictures: a single-centre experience. Acta Chir Belg.2009;109:746–750.*
- (1J-8-15Hz). *Corcoran AT, Smaldone MC, Ricchiuti DD. Management of benign ureteral strictures in the endoscopic era. J Endourol. 2009. 23:1909–1912.*
- (1.2J-10Hz). *Gnessin E, Yossepowitch O, Holland R. Holmium laser endoureterotomy for benign ureteral stricture: a single center experience. J Urol. 2009;182:2775–2779.* -(1.5-2.5J-10-15Hz). *Geavlete P, Georgescu D, Mirciulescu V. Ureteroscopic laser approach in recurrent ureteropelvic junction stenosis. Eur Urol.2007;51:1542–1548.*

- (1.2J-10Hz). *Elabd SA, Elbahnasy AM, Farahat YA, Soliman MG, Taha MR,Elgarabawy MA, Figenshau R. Minimally-invasive correction of ureteropelvic junction obstruction: do retrograde endoincision techniques still have a role in the era of laparoscopic pyeloplasty? Ther Adv Urol.2009;1:227–234.*
- (1J-10Hz). *Wu Z, Feng C, Ding Q, Jiang H, Zhang Y. Ureteroscopic holmium: YAG laser endopyelotomy is effective in distinctive ureteropelvic junction obstructions. Wideochir Inne Tech Malo Inwazyjne. 2011; 6:144–149.*
-(1.2J-10Hz). *Shao Y-H, Wu S-H, Cha T-L, et al. Endoureterotomy for ureteral stricture: A retrospective study of Holmium verus Thulium laser. Int Surg 2017. 102:496-503.*
Changes in the text: No changes.

Comment 3: In methods, what kind of laser duration in laser setting did you select? I mean which you choose the long, or short, or with Moses or not? Because the temperature is different when doing laser cutting into ureteral mucosa.
Reply 3: Unfortunately, the laser equipment we use in the Experimental Unit does not allow us to choose between the pulses to be used, and even less so does it have the Moses effect. It is a Storz Calculase II SCB 20W.
Changes in the text: No changes.

Comment 4. In methods, did you use saline irrigation? If so, you should describe the details of procedure methods.
Reply 4: Thank you for your comment. We have added in the Methods section the irrigation system used. (Ureteroscopy irrigation system by Cook Medical). The irrigation liquid is normal saline solution.
Changes in the text: We have modified our text as advised **"**Ureteroscopic evaluation was carried out by intermittent irrigation with normal saline solution through the Cook Medical irrigation system". (Page 6).

Comment 5. In methods, the author mentioned the placing time in 6mo after endoureterotomy. Why did you select this duration (6mo) of placing stent?
Reply 5: We mention in the manuscript that "A 5Fr polymeric ureteral double pigtail stent was placed in Group-I (Control group) for a 6-week period (Universa® Soft, 22 cm, Cook® Medical)". The average time for stenting after endoureterotomy according to the scientific literature is 4-6 weeks, although there is no consensus due to lack of scientific evidence.
- *Hibi H, Ohori T, Taki T. Long-term results of endoureterotomy using a holmium*

laser. Int J Urol 2007;14:872–874.
- Shao Y-H, Wu S-H, Cha T-L, et al. Endoureterotomy for ureteral stricture: A retrospective study of Holmium versus Thulium laser. Int Surg 2017. 102:496-503.
Changes in the text: No changes.

Comment 6: In results, global success rate by groups, Groups-1 and 2 was higher in 91.6% and 87.5%. This means the difference of procedure methods is not related with success rate of treatment for ureteral stricture. Therefore, does this BUS have the efficacy for care of ureteral stricture?
Reply 6: Indeed, in our study we did not find any parameter evaluated that would lead us to believe that the BraidStent®-H is inferior to the Double Jota ureteral stent. Moreover, with regard to the healing of the ureteral wall that underwent the stricture and subsequent endoureterotomy, we found better results in Group-II.
Changes in the text: No changes.

Comment 7: In results, the author described "Significance was actually found in the assessment of distal ureteral peristalsis, since up to 50% of animals in Group-2 maintained it, whereas no animals in Group-1". I am wondering if how you investigate the degrees of this peristalsis in ureter.
Reply 7: We assessing ureteral peristalsis through Excretory Urography. We follow this protocol to favor ureteral peristalsis visualization: assessment is performed by means of videourography (continuous fluoroscopy C-arm) and the number of waves/minute is counted. Bowel are always prepared 24 hours before surgery in order to remove waste and urinary bladder is emptied in order to favor the pressure gradient between renal pelvis and urinary bladder. Consequently, and given the fact that this study stage takes a long time and there is exposure to ionizing radiation, all assistants leave the OR and the operator works behind a mobile X-ray barrier, which we fortunately have in our experimental OR.
Changes in the text: No changes.

Comment 8: BUS is so interesting manufacture. And then, we knew that this BUS was degraded depending on placing duration. These pieces of degraded BUS run out through the ureteral lumen spontaneously. However, if ureteral stricture is present in distal ureter, can this BUS use?
Reply 8: Thank you for your interesting comment. The design we are assessing in this experimental study, it can only be used when the ureteral stricture is at

least 2 cm above the UVJ. Since it is an intraureteral stent to avoid VUR and stent material in the urinary bladder, it must always be 2 cm above the ureteral orifice. It is true that when ureteral strictures exist in the two 2 cm of the distal ureter, the conventional ureteral stent design will be used but with biodegradable materials.

Changes in the text: "It is important to highlight that due to the design of the BraidStent®-H, treatment of ureteral strictures in the two cm above the ureteral orifice are not indicated. Since it is an intraureteral stent to avoid VUR and stent material in the urinary bladder, it must always be 2 cm above the ureteral orifice". Page 11.

Comment 9. In results, the author described "In Group-2, 75% of the animals showed distal ureteral peristalsis versus just 8.3% in Group-1". Why is Group-2 higher rate?

Reply 9: As the design of the BraidStent®-H is intraureteral and leaves 5-6 cm of the distal ureter unintubated. This allows recovery of the peristaltic wave transmission by allowing coaptation of the ureter walls without the presence of a foreign body. The transmission of the peristaltic wave, although interrupted in the proximal ureter by the stent, is adequately transmitted in the distal ureter. Moreover, at the 6-week follow-up, 91% of the BraidStents® had already degraded, which facilitates the transmission of the peristaltic wave compared to Group-I in which the biostable stent remains in place.

Changes in the text: No changes.

Reviewer C

Comment 1. Authors commented that SVCUG was performed at baseline, 3, 6, and 12 weeks. Does SVCUG conducted under general or local anesthesia? Why do you perform SVCUG frequently?

Reply 1: Unfortunately, it is not possible to perform a routine voiding cystourethrography study in a porcine animal model. Therefore, the animals must be under general anaesthesia and we performed a SVCUG to evaluate the appearance of VUR in the different phases of the experimental study. SVCUG is a technique validated by different research groups to evaluate the appearance of VUR.

-Lumiaho J, Heino A, Aaltomaa S, et al. A short biodegradable helical spiral ureteric stent provides better antireflux and drainage properties than a double-J stent. Scand J Urol Nephrol. 2011;45:129-133.

- Soria F, de la Cruz JE, Budia A, et al. Experimental Assessment of New Generation

of Ureteral Stents: Biodegradable and Antireflux Properties. J Endourol 2020;34:359-365.
Changes in the text: No changes.

Comment 2. "The BraidStent®-H were coated with a layer 8 of 72 μm of sodium heparin 5000 UI/mL." How can you decide the concentration of sodium heparin?
Reply 2: We fully understand the reviewer's concern, this paper is part of a research project that started in 2016, and it includes some *in vitro* studies, firstly, in order to determine the ability of degradable biomaterials to be coated with heparin, and to assess the quantity of heparin that stents can release, as well as the release rate of the heparin coating the stent. All the above is conducted in a laboratory, in artificial urine first and in porcine urine later, by urinalysis and ELISA (Heparin Sodium HS-ELISA). All these studies allow us to perform the study in an animal model, but it is impossible to include all previous year-long development phases of a stent in just one limited-length paper.
BraidStent®-H are homogenously covered by a 233-mg heparin dose whose coating is 72 microns thick. Heparin is not bacteriostatic or bactericidal, but only has a strong anti-adhesive effect. This effect is not influenced by heparin concentration nor does it act with a minimum inhibitory concentration, but is realized by means of a homogeneous stent coating. The aim is to avoid bacteria from adhering to the stent, and from beginning biofilm formation, by means of heparin's electrostatic forces. No research groups report a minimum inhibitory ability, since this is not necessary because of heparin's mechanism of action. Coating thickness is not related to its anti-adhesive ability, but to its release rate, which makes it possible to modify coating time, and that the stent remains biodegradable after the loss of the heparin coating, as we have designed for it.
Changes in the text: No changes.

Comment 3. How can you evaluate the distal ureteral peristalsis?
Reply 3: We assessing ureteral peristalsis through Excretory Urography. We follow this protocol to favor ureteral peristalsis visualization: assessment is performed by means of videourography (continuous fluoroscopy C-arm) and the number of waves/minute is counted. Bowel are always prepared 24 hours before surgery in order to remove waste and urinary bladder is emptied in order to favor the pressure gradient between renal pelvis and urinary bladder. Consequently, and given the fact that this study stage takes a long time and there is exposure to ionizing radiation, all assistants leave the OR and the operator works behind a mobile X-ray barrier, which we fortunately have in our

experimental OR.
Changes in the text: No changes.

Comment 4. What is the definition of global success rate?
Reply 4: We describe in the manuscript that "*Success was strictly defined as the following: relief of signs and ultrasound and fluoroscopic resolution of US and obstructive uropathy at the end of the study*".
Changes in the text: The "global success rate" is included at the end of the Methods section. (Page 7).

Comment 5. If stent was not degraded completely, how can you treat this?
Reply 5: We thank the reviewer for pointing this out. Honestly, we have never had this problem in the more than 62 BUS of this design that we have evaluated in recent years in animal model. Because of their biomaterials, this is very unlikely, since their degradation is exclusively by hydrolysis and all biomaterials have an in vitro study phase previously. But to answer your question, if there is a failure in degradation and it remains in the upper urinary tract, it would be treated in the same way as when there is a fragmented (or encrusted) standard double-jota ureteral stent by retrograde ureteroscopy or a percutaneous approach. Obviously, being an intraureteral stent, its removal is more demanding of endourological skills if there are any complications (like migration, which is probably more common than any degradation failure).
Changes in the text: No changes.

Comment 6. It is needed to be improve the resolution of the figure.
Reply 6: Thank you very much for your comment. We have increased the resolution of the images.
Changes in the text: The resolution of all images has been improved.

Reviewer D
Comment 1: Line 2 - delete "unfortunately".
Reply 1: The word "unfourtunately" has been deleted.
Changes in the text: The word "unfourtunately" has been deleted. Page 2.

Comment 2: line 8 - change to "Then, animals were randomly assigned to..."
Reply 2: the suggested changes have been made.
Changes in the text: "Then, animals were randomly assigned to". Page 2.

Comment 3. Methods can be more robust. How was follow up performed?
Reply 3: The Methods section of the abstract has been rewritten.
Changes in the text: The Methods section of the abstract has been rewritten. Page 2.

Comment 4. Delete line 23 end of sentence "in view of the disappointing results..."
Reply 4: We have made the change.
Changes in the text: " in view of the disappointing results..." has been deleted. Page 2.

Comment 5. Clarify what the authors mean by "high morbidity." Increased number of ER visits?
Reply 5: Issues related to the occurrence of pain in 80% of the patients, work leave, and the impact on sex life related to ureteral stents are included in the manuscript.
Changes in the text: (Page 3). Issues related to the development of pain, medical leave, and sexual impairment associated with ureteral stents are included in the manuscript. Reference 5.
Joshi HB, et al. Indwelling ureteral stents: evaluation of symptoms, quality of life and utility. J Urol 2003;169:1065-9.

Comment 6. There is limited data regarding efficacy of heparin coating of stents to reduce bacterial adhesion. In fact, the primary outcome is prevention of encrustation.
Reply 6: We fully agree with the reviewer that there are few studies, mainly at the clinical level, that have evaluated heparin for ureteral stent coating. However, the use of heparin as ureteral stent coating has been *in vitro* investigated both for the prevention of biofilm formation and encrustation. Heparin is highly electronegative, rendering it repulsive bacterial attachment, as both Gram-positive and Gram-negative bacteria have an overall negative charge at their surfaces (*Ros SF, et al. Bacterial adhesion to phosphorylcholine-based polymers with varying cationic charge and the effect of heparin preadsortion. J Mater Sci Mater Med 2005;16:1003- 1015*). It has been shown that heparin-coated ureteral stents can remain biofilm and encrustation free up to 12 months (*Tenke P, et al. Bacterial biofilm formation on urologic devices and heparin coating as preventive strategy. Int J Antimicrob Agents 2004;23 Suppl 1:S67-74*).

Biofilm formation on the stent surface has been implicated as an important step in the process of stent associated bacteriuria, UTI, stent encrustation and stent-related symptoms. The aim of heparin coating stents to prevent both bacterial colonisation and encrustation is quite similar. Because, although it has been shown that encrustation can be initiated in "sterile" urine (although that is a term that should be discussed, due to the presence of a microbiome in the urine), the main trigger for encrustation is due to bacterial colonisation. Urease producing bacteria in the biofilm and lithogenic characteristics of urine in stone formers seem to be the most likely culprits influencing encrustation of the stent surface (*Sighinolfi MC et al. Chemical and mineralogical analysis of ureteral stent encrustation and associated risk factors. Urology 2015. Broomfield RJ, et al. Crystalline bacterial biofilm formation on urinary catheters by urease-producing urinary tract pathogens: a simple method of control. J Med Microbiol 2009.*)
Heparin is a very safe glycosaminoglycan that has been previously evaluated, that can easily coat the BraidStent®, and is relatively inexpensive procedure. It has also been used in a commercial ureteral stent (Radiance, Cook Medical) and has been little evaluated in in vivo studies in animal models.
It should be borne in mind that in our research for an anti-adhesive coating agent we have to take into account its short-term release in order not to affect the biodegradability of our BUS, a fact that heparin fulfilled perfectly in our *in vitro* studies.
Changes in the text: No changes.

Comment 7. *Which antibiotic was used for prophylaxis?*
Reply 7: We use enrofloxacin for antibacterial prophylaxis.
Changes in the text: The antibiotic used for prophylaxis has been added to the manuscript. Enrofloxacin. (Page 4).

Comment 8 Were any urine parameters (pH, osmolality) besides bacteria measured during this study?
Reply 8: Yes, we perform a urine chemistry analysis: Blood, bilirubin, urobilinogen, ketones, protein, nitrite, glucose, pH, specific gravity, leukocites, creatinine, microalbumin, ascorbic acid, Albumin to Creatinine ratio.
But there are no interesting alterations in these parameters. The pH parameter has been included in Table 1 as it is the most relevant parameter in this study as it can be affected by the BUS degradation reaction.
Changes in the text: Include pH in Table 1.

Comment 9. ureteral stricture animal model section - line 11 change "were" to "underwent"; line 15 change "retrogradely placed" to "placed in a retrograde fashion".
Reply 9: We have made the change.
Changes in the text: Suggested changes were made. (Page 5).

Comment 10. 3 week follow up line 8 change "placement" to "placed".
Reply 10: Suggested changes were made.
Changes in the text: suggested changes were made. (Page 5).

Comment 11. Table 1 - there is a significant drop off in bacteriuria from week 6 to week 12 in group II. Why do the authors believe this to be the case? Perhaps the degradation products of the stents were aspirated and artificially removed from the bladder, thereby reducing bacterial load. Should be mentioned in discussion.
Reply 11: We thank the reviewer for pointing this out. Firstly, it should be considered that what we are determining is bacteriuria, which in all cases was always asymptomatic. That is, the CFU/ml count, which is different from other tests such as urine culture or stent culture.
From 6 to 12 weeks there is no traces of BUS in the urinary tract (except in one animal where it was partially degraded at 6 weeks), so this could not be the cause. Our idea, and the one we are working on for future studies, is that the residual degradation of the BUS is the cause of the free bacteria. As the BUS degrades, the polymeric components begin to break down in an orderly fashion. Similar to all urinary stents, after three weeks a biofilm layer has developed and colonises all stents. In the case of BUS, when this biofilm fragments, it releases the bacteria that were embedded and isolated in the biofilm and become planktonic. For this reason, standard stents do not show this high bacteriuria, as most of the bacteria remain protected and isolated inside the biofilm. This has been demonstrated when stent culture and urinalysis do not coincide and different bacterial species are identified. "*Bacteria within the biofilms differ both in behavior and in phenotypic form from the planktonic, free-floating bacteria*" *Tenke P. Bacterial biofilm formation in urologic devices and heparin coating as preventive strategy. Int J Antimicro Agents 2004, S67-S74".*
Changes in the text: No changes are made to the manuscript as this is already described in the original manuscript. (Page 14).

Comment 12. The use of heparin for stent coating has not been described in detail in the literature. The article cited is a case series of 5 patients, and as such I do not think heparin had a chance at reducing bacteriuria (though arguably there is no good substance to use or it would be widespread on the market).
Reply 12: We fully agree with the reviewer, but our aim as researchers is to look for effective strategies for the prevention of stent-associated infections to interrupt the process of biofilm formation in the new BUS. The prevention of bacterial adherence to ureteral stents surface is an extremely difficult task to achieve, because a high number of adhesion mechanisms exits that may even vary between different species of bacteria (bacterial adhesins). The complexity of the adhesion mechanisms is the true reason why none of the current anti-adherence strategies researched achieve it becomes effective. We believed, like the Yang L et al group, that further studies need to be conducted to determine whether stents with heparin coating have true potential as long-term devices able to resist both encrustation and biofilm formation in vivo. (*Yang L, et al. Ureteral stent technology: drug-eluting stents and stent coatings. Asian J Urol 2015;2:194-201*).
After our study results, we will continue to work on the coating with new agents, perhaps AMP (antimicrobial peptides) against biofilm to reduce BraidStent® colonisation.
Changes in the text: We add to the references a new manuscript with a clinical evaluation of a heparin-coated stent. *Bacteria within the biofilms differ both in behavior and in phenotypic form from the planktonic, free-floating bacteria*" *Tenke P. Bacterial biofilm formation in urologic devices and heparin coating as preventive strategy. Int J Antimicro Agents 2004, S67- S74.*

Comment 13. Overall: The authors show non-inferiority of Braidstent to double JJ stent for ureteral stricture disease, but I would like to see more about specific urine parameters of these animals. What was the baseline pH, and did that change with the dissolution of the stent? What bacteria was present in the urine, and was it the same for all animals? Does it differ from the comparison group, or from other typical porcine urinary contaminants?
Reply 13: Thank you very much for your interesting questions. To answer them, we have included the information you requested in the manuscript and in Table 1. The bacteria identified in both groups are included, a row with pH changes throughout the different phases is included in Table 1, and the occurrence of bacteria in urine in pigs with ureteral stents is discussed with other experimental studies.

Changes in the text: Table 1 (pH), Table 1-identification of bacteria species (*E.coli, Enterococcus sp, Enterobacter sp*) and comparison with other ureteral stents bacteria in porcine model. "We found no differences between the bacteria identified in urine in both experimental groups (*E.coli, Enterococcus sp, Enterobacter sp*), compared to other studies in a swine model, nor between the most commonly identified bacteria associated with indwelling ureteral stents in patients (32-35)". (Page 14).

(32).Soria F, et al. Comparative study of ureteral stents following endoureterotomy in the porcine model: 3 vs 6 weeks and 7F vs 14F. Cardiovasc Intervent Radiol. 2005;28:773-8.

(33). Soria F, et al. Endourologic techniques for ureteropelvic junction obstruction therapy. Comparative animal study. J Pediatr Surg. 2008;43:1528-32.

(34).Al KF, et al. Ureteral stent microbiota is associated with patient comorbidities but not antibiotic exposure. Cell Rep Med. 2020; 22;1(6):100094.

(35).Kehinde EO, et al. Bacteriology of urinary tract infection associated with indwelling J ureteral stents. J Endourol. 2004;18(9):891-6.

Reviewer E

Comment 1. Please provide more information on the sex, initial weight of the pigs at the start and at conclusion of 5 month study. Porcine model tends to grow very significantly and ureteric diameter may be affected.

Reply 1: The weights of the animals at the different follow-ups of the experimental study are included in Table 1. The sex of the animals are all females (already in the original manuscript).

Indeed, such a long-term study in an animal model (5 months) implies that the animals are still growing. But during the 6 weeks of stenting the weight did not increase significantly. But this is always a handicap, the comparative study design aims to reduce the effects of these limitations.

Changes in the text: Table 1**,** weights of the animals at the different follow-ups.

Comment 2. P2;L2: this statement has no bearing on the title or the nature of the study.

Reply 2: The sentence has been removed from the manuscript. "Unfortunately, there are no biodegradable ureteral stents available to date for clinical use".

Changes in the text: The sentence has been removed from the manuscript.

Comment 3. P2L6: please remove "were".

Reply 3: We have made the change.

Changes in the text: The word has been removed from the abstract.

Comment 4. P3L20: does not match title. Consider revising title to show conclusion of study.
Reply 4: In agreement with the reviewer's advice, the title of the manuscript has been changed.
Changes in the text: "Heparin coating in biodegradable ureteral stents does not decrease bacterial colonization. assessment in ureteral stricture endourological treatment in animal model".

Comment 5. P5L19: please describe how many F was BraidStent expanded to in the study. Ureteric ischaemic can occur at 36Fr.
Reply 5: We fully understand the reviewer's concern. It has been one of our research focuses in recent years with respect to the design of this stent. Only the distal end (the anti-migration system) is self-expanding until it contacts with the ureteral wall, up to a maximum of 36Fr. The other part of the BraidStent has a thickness of 3Fr. This distal end is composed of only 4 threads of polymer A in a conformation identical to a Dormia basket. Therefore, the radial force caused by this distal end is minimal, and it is designed to adapt and anchor to the diameter of the ureteral lumen, never to dilate this ureteral lumen. In no case does it have sufficient mechanical force (previously evaluated by *in silico* studies through computational and biomechanical simulation) to distend the ureteral lumen. We have included a description of the stent in Fig. 1.
Changes in the text: Figure 1 and Footnote.

Comment 6. P6L17 Please justify that need for additional intervention in Grp 2 on week4/5. This make interfere with the healing of the stricture.
Reply 6: We fully agree with the reviewer that the assessment at 4 and 5 weeks is a challenging decision and should be undertaken with great precaution. However, it is necessary to evaluate the rate of BraidStent-H degradation in animal models very closely. It is essential to determine when this BUS degrades and how this degradation affects the mechanical properties of the stent. Leaving the macroscopic degradation period unassessed seems to us to weaken the results of this study. It should be borne in mind that for ethical reasons we need to extract as much information as possible from the animal study, obviously without compromising the objectives of the experimental study. According to previous studies by our research group, despite the risk, after 3 weeks, healing of the ureter by second intention is enough. Ureteroscopic evaluation was only

carried out until the distal end of the BUS was evaluated, never progressing over the stent.

- *Soria F, Rioja LA, Blas M, Duran E, Uson J. Evaluation of the duration of ureteral stenting following endopyelotomy: Animal study. Int J Urol. 2006;13:1333-8.*
- *Soria F, et al. Comparative study of ureteral stents following endoureterotomy in the porcine model: 3 vs 6 weeks and 7F vs 14F. Cardiovasc Intervent Radiol. 2005;28:773-8.*

Changes in the text: "In order to closely assess the degradation rate, the size of degradation fragments and the loss of mechanical properties of the BraidStent®-H, intermediate follow-ups at 4 and 5 weeks by ureteroscopy and contrast fluoroscopy were carried out exclusively in Group-II". (Page 6).

Comment 7. P6L19: To elaborate the basis of pathological assessment? Was it graded by an animal pathologist?

Reply 7: Thank you very much for your comment. We agree with you that the assessment by an experienced animal model expert is essential for our line of research. Fortunately, we have this profile in our research. The pathologist evaluating the study has extensive experience in assessment the porcine urinary tract as she has been collaborating with our research line for more than 8 years. She is also a veterinarian and lecturer in the Department of Comparative Anatomy and Pathology (Faculty of Veterinary Sciences. Murcia University. Spain).

Changes in the text: No changes.

Comment 8. P2L18 vs P11L5 vs P14L5=10: just align conclusion, Highly vs as efficacious.

Reply 8: The manuscript is amended to include the reviewer's comment. "BraidStent®-H has been shown to be as efficacious as current ureteral stents in the treatment of benign ureteral strictures following laser endoureterotomy. In addition, it reduces the morbidity associated with current stents and has a homogeneous and predictable degradation rate of about 6 weeks, with no obstructive fragments. Future studies are required to improve the antibacterial coating to reduce BraidStent®-H contamination in view of the results obtained with the heparin coating".

Changes in the text: The Conclusions are amended to include the reviewer's comment.

Comment 9. P9,L16: The increase in size of the porcine model may have an influence.
Reply 9: Totally agree, this clarification is added to the manuscript.
Changes in the text: "However, it may also be due to the fact that the animals continue to grow and increase in weight throughout the study (Table 1)". (Page 10).

Reviewer F
Comment 1. Please change the title to reflect the primary outcome. For example, "Coating biodegradable stents with heparin does not improve bacterial colonization."
Reply 1: Thank you for your comment.
Changes in the text: New title: "Heparin coating in biodegradable ureteral stents does not decrease bacterial colonization.Aassessment in ureteral stricture endourological treatment in animal model".

Comment 2. To study the advantage of heparin coating on biodegradable stents, it may have been more appropriate to compare the Braidstent-H with the regular Braidstent rather than with the standard double-J alone. The authors should comment on why this wasn't the case.
Reply 2: Thank you for your comment. This experimental study is part of a research project since 2016. The uncoated BraidStent study was already published by our group before, the Animal Experimentation Ethics Committee does not allow to repeat experimental groups already evaluated. Nor does the publishers' policy allow the use of experimental groups that have already been described in the scientific literature. In the present study our aim is to evaluate the heparin-coated BraidStent after endourological treatment of benign ureteral strictures, since the use of this BUS to ensure correct ureteral healing by second intention and being an intraureteral stent constitutes an unevaluated challenge. In the Discussion we have compared our results between both BraidStents with respect to bacterial colonisation, finding no differences.
Changes in the text: No changes.

Comment 3. It would strengthen the methods to include representative images of some of the study parameters, such as retrograde pyelography or endoscopic images demonstrating the ureteral strictures, SVCUG to show reflux, and excretory urography to show peristalsis.

Reply 3: Thank you for your comment.
Changes in the text: We added 5 new figures. Following the policy of Trans Androl Urol: Tables/Figures, 10 figures are deemed sufficient.
Figure 3. BraidStent®-H assessment under ultrasound control (3 weeks).
Figure 4. Ureteroscopic view of the non-obstructing BraidStent®-H fragments at 5 weeks.
Figure 5. Non-obstructing urothelial hyperplasia. Distal anchoring system location.
Figure 7. SVCUG. VUR Assessment in Group-I (6 weeks). *-Catheter to control intravesical pressure. **-Double pigtail ureteral stent. ***-Urine bladder.
Figure 8. Ureteroscopic assessment of the ureteral healing in Group-II (5 months).

Comment 4. The authors should indicate which types of bacteria were predominantly found on urine culture at each of the follow-up time points. It would be interesting to see whether the different stent types favor different types of bacterial colonization.
Reply 4: Thank you for your comment.
Changes in the text: The different species of bacteria found in the urine tests (*E.coli and Enterococcus sp, Enterobacter sp*) have been included in the Table 1.

Reviewer G
Comment 1. Please describe the morphology of Illustration of the BraidStent -H in more detail.
Reply 1: Thank you for your comment.
Changes in the text: The morphology of the BraidStent-H is described in depth in Figure 1.

Comment 2. What kind of bacteria are there at the time of Bacteriuria?
Reply 2: Thank you for your feedback.
Changes in the text: The different species of bacteria found in the urine tests (*E.coli, Enterococcus sp, Enterobacter sp*)) have been included in the Table 1.