Development of a Therapeutic Capsule Endoscope for Treatment in the Gastrointestinal Tract: Bench Testing to Translational Trial

F.R. Stewart, I.P. Newton, I. Näthke
School of Life Sciences
University of Dundee
Dundee, DD1 5EH, UK

B.F. Cox
School of Medicine
University of Dundee
Dundee, DD1 9SY, UK

Z. Huang
School of Science and Engineering
University of Dundee
Dundee, DD1 4HN, UK

S. Cochran
School of Engineering
University of Glasgow
Glasgow, G12 8QQ, UK

***Abstract* — Video capsule endoscopy is a widely accepted clinical alternative to conventional endoscopy for examination of the gastrointestinal tract. Its advantages are that it can visualize the entire gastrointestinal tract including the anatomically remote small intestine and it is less invasive for the patient as compared to conventional endoscopy. However, video capsule endoscopy is suitable only for diagnosis, with little research into therapeutic capsule endoscopy. Like video capsule endoscopy, therapeutic capsule endoscopy has great potential to reach locations that would previously have been difficult, such as the small intestine. Ultrasound-mediated targeted drug delivery is a promising therapeutic capsule based modality as it may be scaled to size, provides temporary gut barrier disruption and power requirement is relatively low.**

This paper investigates the feasibility of a therapeutic capsule endoscope utilizing ultrasound-mediated targeted drug delivery. A prototype device, SonoCAIT, was built and tested. Further investigation of the drug delivery capabilities of the miniature focused US sources, testing was carried out on an *in vitro* model replicating the lining of the small intestine. This involved measuring the transepithelial resistance, a measure for barrier function, during insonation. A drop in transepithelial resistance occurred during insonation and returned to starting value post insonation. In anticipation of *in vivo* work, SonoCAIT, was reconfigured to operate within the small bowel of pigs.

Keywords* — *Capsule Endoscopy; UmTDD; SonoCAIT; Therapeutic Capsule Endoscope; GI Therapy

I. INTRODUCTION

Gastrointestinal (GI) diseases, such as inflammatory bowel disease (IBD), are an increasingly major health problem worldwide [1]. Crohn's disease, a subset of IBD, is a chronic, relapsing-remitting inflammatory disorder affecting the entire GI Tract.

Despite the role of the GI tract as a conduit for nutrient entry, GI epithelia can distinguish between beneficial and harmful agents. Current therapeutic agents used for treatment of Crohn's range from steroids to highly potent biological agents administered orally or intravenously and can be associated with unwanted side effects. Delivering drugs locally, to the diseased region presents a means for reduced systemic effects. In addition, delivering drugs beyond the GI epithelia could increase drug uptake and hence effectiveness.

Crohn's disease occurs most often in the small intestine, predominately at the ileocecal junction. However, Crohn's can manifest panenterically, making diagnosis via endoscopy more complex since this standard technique cannot visualize the entire small intestine. A promising alternative that allows visualization of the entire GI tract is video capsule endoscopy (VCE). VCE involves ingesting an autonomous capsule containing video imaging and wireless communications. Therefore, it can visualize the entire GI tract, reduce stress on the patient since it is less invasive than endoscopy, and reduce costs since it does not require a skilled operator to control [2].

There have been multiple examples of capsule endoscopes developed for therapy by acting as drug delivery vehicles. Prominent examples of such devices are Intelisite [3] and Enterion [4]. These capsules contained drug reservoir with capacity of 1 ml that can be emptied remotely. However, neither capsule can deliver drugs across the GI epithelium, limiting their efficacy. A capsule was developed that contained a microneedle for delivery through the mucosa [5] however, like the Enterion and Intelisite capsules, it cannot be positioned or localised and the microneedle poses a risk of perforating the mucosa. This could cause further problems, such as a breach in the GI epithelium, which would allow bacterial invasion.

US-mediated targeted drug delivery (UmTDD) is a means to overcome some of the challenges associated with therapeutic capsule endoscopy (TCE). UmTDD allows therapeutic agents to be delivered directly to the treatment site, reducing unwanted systemic effects, and uses focused US to direct, release, and enhance uptake of the therapeutic agents [6-8]. Typically, focused US transducers used for UmTDD are large arrays and extracorporeal. An example of UmTDD used in this configuration is for the treatment of uterine fibroids [9]. In this configuration, the US beam could be disrupted by gas pockets and bone, with guidance often required, using magnetic resonance for instance. This results in the procedure becoming highly complex and expensive. A potential solution to this would be to contain an UmTDD system in a capsule endoscope. UmTDD could solve some of the problems associated with other TCEs by allowing drugs to be delivered through the GI epithelium, potentially improving treatment. However, despite the immense potential of UmTDD capsules challenges remain that include: miniaturising the US transducers to fit within a capsule; ensuring the miniature transducers can enhance uptake; and positioning the capsules so they are directed towards the treatment site.

This paper sets out to address some of these challenges. A prototype therapeutic capsule, SonoCAIT, was constructed and benchtop testing has been carried out, discussed in section II. In section III, testing of the miniature focused US sources used in SonoCAIT on appropriate *in vitro* models of the small intestine to determine if they can enhance drug uptake is described. In section IV, the reconfiguration of SonoCAIT is described so that it is suitable for translational use and the next steps towards *in vivo* trials are discussed.

II. Prototype Therapeutic Capsule Endoscope: SonoCAIT

A. Capsule Construction

The Capsule has four main components necessary for UmTDD: focused US transducer, drug delivery channel, video camera and light source. The focused US source consisted of a self-focusing PZ26 (Meggitt Sensing Systems, Kvistgaard, Denmark) bowl with outer diameter OD = 5 mm, radius of curvature R_C = 15 mm, and frequency f = 4 MHz. The backing layer consisted of microballoon loaded epoxy (Epofix, Struers A/S, Denmark) and the transducer consisted of a case produced by additive manufacturing (Stratysis Ltd., MN, USA). The detailed fabrication process is discussed by Stewart et al [6]. The drug delivery channel consisted of a fine bore polythene tubing with outer diameter OD = 0.96 mm and inner diameter ID = 0.58 mm. Video imaging was provided by a CMOS camera (microScoutCam™, Medigus Ltd., Israel) with outer diameter OD = 1.2 mm and length L = 5 mm. The camera has an imaging area of 492.8 µm x 488.4 µm and corresponding resolution of 220 x 224 pixels. The camera was connected to a video processor which captures and saves imaging data. Illumination was provided for the camera by utilizing a printed circuit board (PCB) with four 40 mW (OSRAM Opto Semiconductors GmbH, Germany). The PCB has a 1.5 mm diameter hole in the center to allow the camera to pass through.

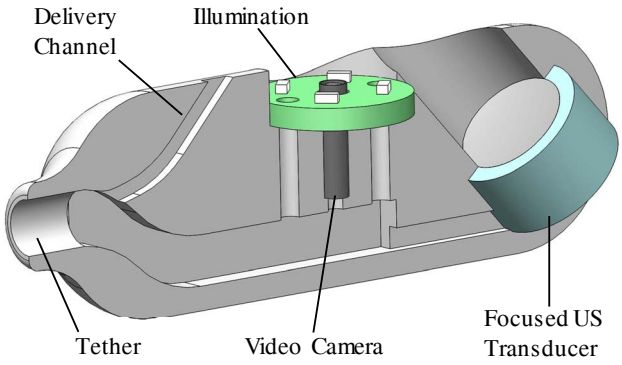

Fig.1. Computer aided cross sectional drawing of SonoCAIT. Capsule contains drug delivery channel, video camera and illumination, and focused US transducer.

The prototype capsule, SonoCAIT, was initially tethered to provide power and therapeutic agent delivery. The tether that was chosen for the prototype capsule has an outer diameter OD = 2.25 mm and inner diameter ID = 1.65 mm and houses power cables for camera and illumination and drug delivery channel. The capsule shell was designed in SolidWorks (Dassault Systemes SOLIDWORKS Corp., Waltham, MA) and produced by additive manufacturing using an Objet Connex 500 printer (Stratasys Ltd., Minnesota, USA). A computer-aided drawing of the prototype capsule and components is shown in Fig.1. below.

B. Benchtop Testing

Benchtop testing was carried out to characterize the ability of the miniature focused US sources to direct microbubbles (MBs) towards the target location (i.e. towards the US focus). The capsule was configured as shown in Fig.1. and MBs were passed through the delivery channel and into the US field. The behavior of the MBs was monitored via the video camera. It was found that the stream of MBs was redirected by more than 90^o by the acoustic radiation forces of the focused US. This result was important because it showed that the focused US transducer can direct MBs (that could be loaded together with therapeutic agents) towards the targeted location. Further details of the benchtop testing are described in [10].

III. Measuring the Effects of Focused Ultrasound on Small Intestine Cell Model

This section investigates the effectiveness of the miniature focused US sources at enhancing drug uptake through the GI barrier.

A. Materials and Methods

Cultured human colon cancer cells (Caco-2) form a model of the small intestine once differentiated and polarized. They mimic enterocytes lining the small intestine, and form cellular junctions and microvilli [11]. Consequently, Caco-2 cells are widely accepted in the literature and are a food and drug administration (FDA) approved model of the small intestine for drug transport studies.

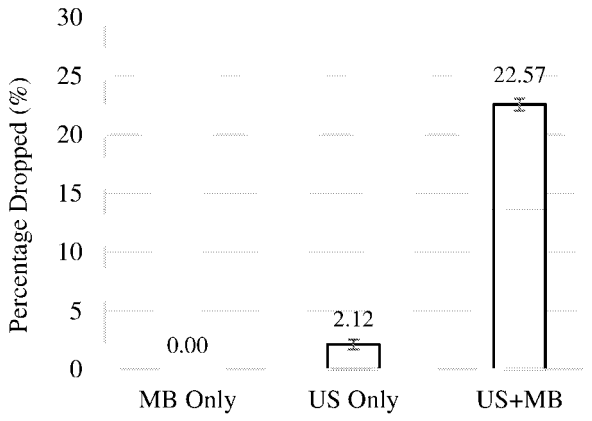

Fig. 2. TER drop, as a percentage of original resistance, of the Caco-2 cell model during insonation. Controls included MB only and US only. The MB+US sample showed a ten times greater drop in TER than the US Only.

Caco-2 cells were maintained in appropriate cell media and cultured under aseptic conditions. Cells were seeded on suspended membranes (ThinCert, Greiner Bio-One, Kremunster, Austria) at 500,000 cells per 12-well ThinCert. They were grown for 21-25 days until fully differentiated and polarized where the small intestinal model is established.

To perform experiments on the cell model, an insonation system was used [10] that matched transducers in SonoCAIT, with the exception that the delivery channel is now integrated and passes through the center of the transducer. This configuration minimizes the footprint of the UmTDD components and saves space in future iterations of SonoCAIT. Cells were insonated with a continuous sinusoidal waveform for 60s and an amplitude of 10 V_{pp}, corresponding to a pressure of 150 kPa. Transepithelial resistance (TER), a measure of cell barrier function [12], was measured during and post insonation.

Microbubbles (SonoVue, Bracco S.p.A., Milan, Italy) were introduced through the delivery channel of the transducer at a rate of 30 ml per hour at a concentration of $2x10^6$ MBs/ml diluted in cell media. Each experiment was performed in triplicate. Controls included: MBs introduced through the delivery channel, with no US present; and US only, with no MBs present.

B. *Results*

Results are displayed in Fig.2. The MB and US samples showed a drop in TER approximately ten times greater than controls. Since TER is a measure of epithelial barrier function, such increased drop in TER could translate into enhanced uptake of drugs through the mucosal barrier. However, further experiments are necessary to determine the exact relationship between reduced TER and increased drug uptake. In all samples, TER returned to starting values approximately 3 minutes post insonation. Such a temporary reduction in barrier function means the risk of bacterial invasion is minimal.

IV. Progress Towards *In Vivo* Trials

The next step involves moving toward testing the capsule *in vivo*. Fluorescent particles will be delivered using SonoCAIT and the effectiveness of the on-board transducers at enhancing uptake will be assessed. Fluorescent particles will be utilized because they allow quick post experimental analysis via fluorescent illumination and are cost effective.

The following section describes the reconfiguration of SonoCAIT to make it suitable for use *in vivo* and describes the future experiments it will be used for.

A. *SonoCAIT Reconfiguration and Construction*

The main components in SonoCAIT are the drug delivery channel and focused US transducer; however, it lacks a means to measure uptake of the fluorescent particles. This will be achieved using the fluorescent imaging capsule developed by Al-Rawhani et al [13]. Therefore, SonoCAIT only needs to contain drug delivery channel and focused US components and can remain tethered. This allows *in vivo* testing to be performed earlier than previously anticipated. Future versions of SonoCAIT will combine modalities from both capsules.

The capsule was designed in SolidWorks (Dassault Systemes SOLIDWORKS Corp., Waltham, MA) and printed using an Objet Connex 500 (Stratasys Ltd., Minnesota, USA). The capsule is printed in two halves to allow easier assembly and recessed grooves lock the two halves together. The transducer was constructed in the same process outlined by Stewart et al [10]. It was necessary that the tether is flexible enough so that it does not damage or deform the bowel but also have a large enough inner diameter that it can contain drug delivery and power cables. To achieve this, a GI feeding tube was used (CORPAK MedSystems, IL, USA) that has OD = 4 mm, ID = 2.65 mm, and L = 1.4 m. As they are designed for use *in vivo* they are flexible and CE marked. Finally, the capsules and tether were parylene coated to provide biocompatibility and to reduce friction. The reconfigured capsule is shown in Fig.3..

B. *Future Experiments Using SonoCAIT*

Experiments will be carried out as part of a study approved by the Animal Welfare and Ethical Review Board of the Roslin Institute, Roslin, Midlothian, EH25 9RG and are being carried out under Home Office (UK) License PPL 7008812.

The aim is to determine if UmTDD is feasible *in vivo* and if it can enhance uptake. The capsule will first be inserted into the

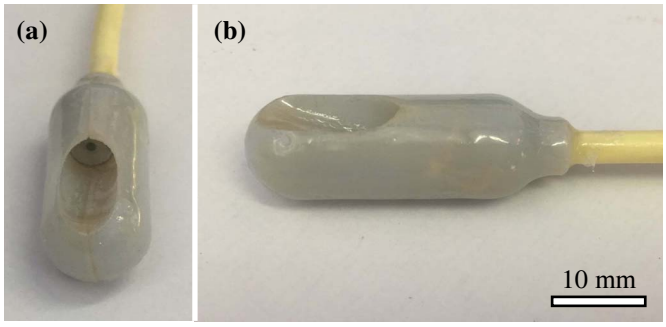

Fig. 3.(a) Front view of the *in vivo* reconfiguration of SonoCAIT. Transducer and drug delivery channel are shown, along with tether to provide power and delivery. (b) Side view. Capsule is 11 mm diameter by 30 mm in length, similar in size to commercial capsule endoscopes.

porcine small bowel. The small bowel will be insonated, and fluorescent particles will be introduced. The fluorescence imaging capsule will then be administered into the small bowel and it will assess uptake. Post mortem, the small bowel tissue will be investigated visually and by histology.

V. Conclusion and Future Work

This paper described the development and testing of a therapeutic capsule endoscope utilizing UmTDD.

In summary, the prototype capsule showed it was possible to fit an UmTDD system into capsule endoscope format. It was shown that this configuration could redirect a stream of MBs towards the treatment site. The effectiveness of the miniature focused US transducers at enhancing drug uptake was investigated on a small bowel cell model. Reduced barrier function was observed during insonation, which could translate to enhanced intercellular drug uptake. Finally, the reconfiguration of SonoCAIT to make it suitable for *in vivo* use was discussed. Future work involving this capsule was described and will include measuring the uptake of fluorescent particles delivered by the capsule into porcine small bowel and assessing their uptake into tissue with a fluorescence imaging capsule and post mortem via histology. This work shows that UmTDD as a modality for TCE is feasible could provide immense clinical benefit for treatment of GI diseases such as Crohn's disease.

Initially tethered, to facilitate delivery power and reagents, future versions will include wireless communications and power delivery. This could be achieved by embedding an antenna in the capsule shell to maximize space within the capsule [14]. In addition, future capsules will combine positioning and localization functionality to allow precise spatial targeting of reagents. This could be achieved by controlling the capsule with magnets, similar to Slawinski et al [15] who use a magnetic robotic arm to propel a capsule endoscope. Future work will aim to combine the UmTDD system described in this paper with fluorescence imaging, wireless communications and positioning/localization facilities to allow fully autonomous therapy in the GI tract.

Acknowledgment

Financial support from the UK Engineering and Physical Sciences Research Council, Grant EP/K034537 (Sonopill Programme) is gratefully acknowledged.

References

[1] G. G. Kaplan, "The global burden of IBD: from 2015 to 2025," *Nature reviews Gastroenterology & hepatology,* vol. 12, pp. 720-727, 2015.

[2] A. Van Gossum, M. Munoz-Navas, I. Fernandez-Urien, C. Carretero, G. Gay, M. Delvaux*, et al.*, "Capsule endoscopy versus colonoscopy for the detection of polyps and cancer," *New England Journal of Medicine,* vol. 361, pp. 264-270, 2009.

[3] N. J. Clear, A. Milton, M. Humphrey, B. T. Henry, M. Wulff, D. J. Nichols*, et al.*, "Evaluation of the Intelisite capsule to deliver theophylline and frusemide tablets to the small intestine and colon," *European journal of pharmaceutical sciences,* vol. 13, pp. 375-384, 2001.

[4] I. Wilding, "The enterion capsule: a novel technology for understanding the biopharmaceutical complexity of new molecular entities (NMEs)," *Drug Deliv Tech,* vol. 1, pp. 8-11, 2001.

[5] S. P. Woods and T. G. Constandinou, "Wireless capsule endoscope for targeted drug delivery: mechanics and design considerations," *IEEE Transactions on Biomedical Engineering,* vol. 60, pp. 945-953, 2013.

[6] F. Stewart, B. Cox, J. Vorstius, A. Verbeni, Y. Qiu, and S. Cochran, "Capsule-based ultrasound-mediated targeted gastrointestinal drug delivery," in *Ultrasonics Symposium (IUS), 2015 IEEE International*, 2015, pp. 1-4.

[7] Y.-Z. Zhao, Y.-K. Luo, C.-T. Lu, J.-F. Xu, J. Tang, M. Zhang*, et al.*, "Phospholipids-based microbubbles sonoporation pore size and reseal of cell membrane cultured in vitro," *Journal of drug targeting,* vol. 16, pp. 18-25, 2008.

[8] S. Hernot and A. L. Klibanov, "Microbubbles in ultrasound-triggered drug and gene delivery," *Advanced drug delivery reviews,* vol. 60, pp. 1153-1166, 2008.

[9] D. Gourevich, O. Dogadkin, A. Volovick, L. Wang, J. Gnaim, S. Cochran*, et al.*, "Ultrasound-mediated targeted drug delivery with a novel cyclodextrin-based drug carrier by mechanical and thermal mechanisms," *Journal of controlled release,* vol. 170, pp. 316-324, 2013.

[10] F. R. Stewart, Y. Qiu, H. S. Lay, I. P. Newton, B. F. Cox, M. A. Al-Rawhani*, et al.*, "Acoustic Sensing and Ultrasonic Drug Delivery in Multimodal Theranostic Capsule Endoscopy," *Sensors,* vol. 17, p. 1553, 2017.

[11] V. Meunier, M. Bourrie, Y. Berger, and G. Fabre, "The human intestinal epithelial cell line Caco-2; pharmacological and pharmacokinetic applications," *Cell biology and toxicology,* vol. 11, pp. 187-194, 1995.

[12] M. S. Balda, J. A. Whitney, C. Flores, S. González, M. Cereijido, and K. Matter, "Functional dissociation of paracellular permeability and transepithelial electrical resistance and disruption of the apical-basolateral intramembrane diffusion barrier by expression of a mutant tight junction membrane protein," *The Journal of cell biology,* vol. 134, pp. 1031-1049, 1996.

[13] M. A. Al-Rawhani, J. Beeley, and D. R. Cumming, "Wireless fluorescence capsule for endoscopy using single photon-based detection," *Scientific reports,* vol. 5, p. 18591, 2015.

[14] J. Faerber, G. Cummins, and M. P. Desmulliez, "Design of conformai wideband antennas for capsule endoscopy within a body tissue environment," in *Microwave Conference (EuMC), 2016 46th European*, 2016, pp. 1223-1226.

[15] P. R. Slawinski, A. Z. Taddese, K. B. Musto, K. L. Obstein, and P. Valdastri, "Autonomous Retroflexion of a Magnetic Flexible Endoscope," *IEEE Robotics and Automation Letters,* vol. 2, pp. 1352-1359, 2017.