1 **Title page**

2

3 **Novel Balloon Surface Scanning Device for**
4 **Intraoperative Breast Endomicroscopy**

5

6

7 Siyang Zuo, Michael Hughes, and Guang-Zhong Yang

8 Hamlyn Centre for Robotic Surgery, Imperial College London, London

9 SW7 2AZ, U.K.

10 Tel: [+44 \(0\) 20 7594 0814](tel:+442075940814)

11 Fax: +44 (0)20 7594 8260

12 E-mail: siyang.zuo@gmail.com

13

14

15

16

17

18

19

20

21

22

23

24

25

26 **Abstract**

27 Recent advances in fluorescence confocal endomicroscopy have
28 allowed real-time identification of residual tumour cells on the walls of
29 the cavity left by breast conserving surgery. However, it is difficult to
30 systematically survey the surgical site because of the small imaging
31 field-of-view of these probes, compounded by tissue deformation and
32 inconsistent probe-tissue contact when operated manually. Therefore,
33 a new robotized scanning device is required for controlled, large area
34 scanning and mosaicing. This paper presents a robotic scanning probe
35 with an inflatable balloon, providing stable cavity scanning over
36 undulating surfaces. It has a compact design, with an outer diameter of
37 4 mm and a working channel of 2.2 mm, suitable for a leached flexible
38 fibre bundle endomicroscope probe. With the probe inserted, the tip
39 positioning accuracy measured to be 0.26 mm for bending and
40 0.17 mm for rotational motions. Large area scanning was achieved (25-
41 35 mm²) and the experimental results demonstrate the potential clinical
42 value of the device for intraoperative cavity tumour margin evaluation.

43 *Keywords: Breast conserving surgery, confocal endomicroscopy,*
44 *image mosaicing, mechanical design, surgical robot*

45 **INTRODUCTION**

46 Breast cancer is the second most common cancer in the world and,
47 by far the most frequent cancer among women with an estimated 1.67
48 million new cancer cases diagnosed in 2012 (11.9% of all cancers) [1].
49 Breast conserving surgery (BCS) is a procedure for treating smaller
50 cancers by removing the tumour and a part of normal tissue while
51 sparing the remainder of the breast [2]. However, complete tumour
52 removal is currently confirmed post-operatively by histology, which often
53 results in patient being required to return to theatre for further surgery
54 [3]. A re-operation has several disadvantages, including risks of
55 infections, poor cosmesis, and higher costs. [4].

56 Photoacoustic microscopy using focused lasers with ultrasound
57 transducers as receivers have been shown to have the ability to create
58 sub-micron resolution images of cells and tissue [5]. Furthermore,
59 fluorescence confocal endomicroscopy[6]has been developed to image
60 and diagnose cancer *in situ* [7-11], particularly in the gastro-intestinal
61 tract. It can obtain optically sectioned, high-resolution images of tissue
62 *in situ*, which has previously been stained using a fluorescent agent
63 such fluorescein or acriflavine. It involves use of a miniaturized
64 microscope probe, inserted through the instrument channel of an
65 endoscope or similar device, and placed in direct contact with the tissue.
66 Given the difficulties of miniaturizing the device for the high-speed laser
67 scanning needed for confocal imaging, a common approach is to relay
68 light to and from the tissue using a fibre imaging bundle [12-14]. This
69 allows the entire optical system to be positioned outside the patient.

70 More recently, it has been suggested that endomicroscopy could be
71 adapted to intraoperative imaging of breast tissue [15-16]. For this
72 application, the probe would be inserted into the cavity created by
73 removal of the tumour and used to search for residual tumour cells on
74 the cavity wall. Bench-top confocal endomicroscopy has already been
75 considered for analysis of breast tissues, and been shown to allow key
76 morphological structures of normal and cancerous breast tissues to be
77 identified and differentiated [17]. By offering a way to identify the
78 margins of the tumour *in situ*, endomicroscopy could therefore reduce the
79 currently high rates of reoperations [18], as well as reducing high cancer
80 recurrence rates due to incomplete excision.

81 However, endomicroscopy has several limitations that have made
82 breast imaging challenging. Most importantly, the limited number of fibre
83 cores in the fibre bundle means that only a small field-of-view can be
84 obtained (typically less than 0.5 mm for high resolution probes). Video
85 mosaicing techniques can be used to mitigate this problem [19-21], but
86 scanning over large areas is time-consuming and requires high dexterity.
87 Mechanical scanning of the probe has therefore been proposed for
88 laparoscopic applications, including a scanning device using hydraulic

89 micro-balloons [22], and a conic screw-like structure [23]. However, the
90 area imaged by the miniaturised versions of these designs is typically
91 less than 3 mm².

92 Many existing robotic devices, including the da Vinci robot (Intuitive
93 Surgical), utilize wire-driven mechanisms to transfer actuation power
94 from actuators to actuated parts in the tip of the forceps [24-26]. While
95 the wire driven mechanism allows miniaturization of multi Degree of
96 Freedom (DOF) structures, problems of friction, wearing and extension
97 are practical difficulties to consider. Alternatively, some manipulators
98 are driven by linkage mechanisms [27-29]. They provide a high stiffness,
99 durability, and strong force. However, the reported linkage driven
100 mechanisms often result in an increase in the number of parts.
101 Furthermore, it is particularly difficult to reduce the backlash when there
102 is a large number of linkages.

103 A preliminary prototype of a breast scanning device, using a rigid
104 concentric tubes scanning mechanism, has previously been reported
105 [30]. However, the outer diameter of the device was 6 mm, which is
106 large for this application. The device was demonstrated using a fibre
107 bundle probe taken from a Cellvizioendomicroscope (Mauna Kea
108 Technologies), which had limited flexibility, restricting the scan
109 area. There were also difficulties with tissue deformation as the probe
110 was scanned across the tissue.

111 In this study, we propose a new device which uses a gear-guided
112 linkage scanning mechanism to enable safe, efficient and accurate
113 large area scanning of the breast cavity. This scanning device would be
114 inserted through the incision created during BCS, and deployed against
115 the cavity walls, thus allowing high quality image mosaics to be created
116 (Fig. 1). The scanning is performed inside an inflatable balloon which
117 could help to stabilize the tissue by pressing the balloon on it. A custom-
118 built probe, using a leached fibre bundle and a micro-lens, allows the
119 full range of motion of the device to be used and provides sufficient
120 working distance to allow imaging through the balloon. The mechanical
121 characteristics of the device are analyzed, and imaging results from a

122 breast phantom and ex vivo human breast tissue are presented,
123 showing that the device can achieve large area endomicroscopy
124 imaging and mosaicing.

125

126 **FIGURE 1.**Concept of the breast scanning system with an inflated balloon
127 during breast conserving surgery for breast cancer.

128 **MATERIALS AND METHODS**

129 *Scanning Mechanism*

130 To achieve both a small outer diameter and a large inner diameter for
131 the device, the design concept needed to be simple and with a small
132 number of parts. The proposed device has the ability to scan over an
133 arbitrary trajectory by combining bending and rotational motion of the
134 tip. The scanning structure, illustrated in Fig. 2, is based on an epicyclic
135 motion between the tip and base frames. The tip frame and base frame
136 are in contact with spur gears and are connected by the middle frame.
137 Bending motion is driven by linear movement of the linkage, and
138 rotational motion is provided by rotation of the base frame. This
139 provides 0 to +90 degrees bending on one axis, and 360 degrees of
140 rotation on the second axis. The tip frame and base frame are guided
141 by spur gears, which enable smooth bending motion without slippage
142 (tip bending repeatability of 0.6 degrees as shown in RESULTS
143 section)between the frames.

FIGURE 2.The configuration of the scanning mechanism, transforming linear motion of the linkages and rotational motion of the base frame to a 2D scanning motion of the tip.

Miniature Scanning Device

The scanning prototype is 4 mm in diameter, incorporating a central hollow channel (2.2 mm in diameter), through which the endomicroscope probe can be deployed, as shown in Fig. 3(a). The device has a length of 95 mm, while the bending tip has a length of 25 mm. The scanning structure was fabricated from stainless steel. A passive linear structure is located at the distal end (Fig. 3(b)), consisting of a slider, a spring and a slide guiding tube. The endomicroscope probe is fixed on the linear structure, helping to ensure consistent contact between the tip and the tissue (Fig. 3(b)). The endomicroscope probe can easily be separated from this structure for cleaning. The workspace of the scanning device's tip is a large hemisphere, as shown in Fig. 3 (e).

FIGURE 3 A prototype of the scanning device, showing (a) the image of scanner, (b) distal end with distal object, (c) Balloon with guide tube, (d) distal end with balloon inflation, (e) The workspace of the scanning device's tip with motorized bending and rotating motions. The workspace of the tip is a large hemi-sphere. (f) Details of the drive unit to drive 2-DOFs scanning mechanism.

The base frame is driven by a high-resolution brushless DC-servomotor equipped with a gear head and Hall effect sensor (1226 E 012 B K1855, Faulhaber SA, Germany) (Fig. 3(f)). The unit driving the linear motion of the linkage consists of a lead screw, a brushless DC-servomotor (0620 C 012 B K1855, Faulhaber SA, Germany), a linear position sensor (EVA-W7XR04B34, Panasonic, Japan), a linkage guide and a bearing slide block (Fig. 3(f)). The linkage rod is fixed to the bearing structure which rotates with the base frame, thus providing

176 simultaneous linear and rotational motions. The drive unit is enclosed
177 within in a cylindrical cover with outer diameter less than 30 mm, and
178 the total weight of the scanning device is 83 g, thus making it suitable
179 for handheld use. For cleaning and sterilization purposes, the scanning
180 distal side can easily be separated from the driver unit. A standard
181 computer calculates rotational and linear displacements of the base
182 frame and linkage from inputted target values.

183 *Tip Scanning Balloon*

184 A balloon inflation system was developed to provide a smooth scan
185 surface and minimize tissue deformation. The balloon is a commercial
186 medical balloon (Model: 30000000BC, material: Urethane Very Low
187 Durometer, Vention Medical, USA) with an inflated diameter of 30-50
188 mm and an inflated wall thickness of 15- 20 μm (Fig. 3(c-d)). The balloon
189 is fixed on a guide tube, and the scanning device is inserted into the
190 guide tube (5 mm in outer diameter) during scanning (Fig. 3(c-d)). An
191 O-ring is used to seal the cap between guide tube and scanning
192 device, and there is an inner spacer inserted into the working channel
193 to prevent an air leakage. The scanning device is connected to a micro
194 diaphragm air pump (8018Gt, Namiki Precision Jewel Co., Ltd, Japan),
195 and the inner pressure of the balloon was detected by a pressure
196 sensor (MLH150PSB01A, Honeywell). It was experimentally
197 determined that stable scanning was obtained when maintaining an
198 inner pressure of 140 kPa.

199 *Kinematics, Control algorithm and Trajectory*

200 We developed a custom user interface (Labview, National
201 Instruments) to control the prototype from a standard PC. Once the
202 scan parameters are entered, the device can scan the target surface
203 automatically.

204 The relationship between the bending angle, θ , and linear linkage
205 displacement, y , is given by (1), where the lengths a , b , and c are
206 shown in Fig. 4 (a).

$$y = a + b \left(\cos \frac{\theta}{2} - 1 \right) + c \sin \frac{\theta}{2} - \sqrt{a^2 - \left\{ b \sin \frac{\theta}{2} + c(1 - \cos \frac{\theta}{2}) \right\}^2}$$

$$= f(\theta, a, b, c) \quad (1)$$

207

208 The inverse function $f^{-1}(\theta, a, b, c)$ of (3) can be computed as follows:

$$a^2 - \left\{ b \sin \frac{\theta}{2} + c(1 - \cos \frac{\theta}{2}) \right\}^2 = \left\{ a + b \left(\cos \frac{\theta}{2} - 1 \right) + c \sin \frac{\theta}{2} - y \right\}^2$$

$$\{b^2 + c^2 + (y - a)b\} \cos \frac{\theta}{2} + (y - a)c \sin \frac{\theta}{2}$$

$$= b^2 + c^2 + (b - a)y + \frac{1}{2}y^2 - ba \quad (2)$$

209 When,

$$A(y) = b^2 + c^2 + (y - a)b$$

$$B(y) = (y - a)c$$

$$C(y) = b^2 + c^2 + (b - a)y + \frac{1}{2}y^2 - ba$$

210 Then equation (2) can be converted to:

$$A(y) \cos \frac{\theta}{2} = C(y) - B(y) \sin \frac{\theta}{2}$$

$$\{A(y)^2 + B(y)^2\} \sin^2 \frac{\theta}{2} - 2B(y)C(y) \sin \frac{\theta}{2} + \{C(y)^2 - A(y)^2\} = 0 \quad (3)$$

211 When $A(y) = 0$; $y = \frac{1}{b}(ab - b^2 - c^2)$,

$$\theta = 2 \arcsin \left\{ \frac{C(y)}{B(y)} \right\} \quad (4)$$

212 When $A(y) \neq 0$,

$$\sin \frac{\theta}{2} = \frac{B(y)C(y) \pm \sqrt{B(y)^2 C(y)^2 - (A(y)^2 + B(y)^2)(C(y)^2 - A(y)^2)}}{A(y)^2 + B(y)^2} \quad (5)$$

213 Combining (4) and (5), the complete expression for the bending angle

214 θ can be given as follows:

$$\theta = \begin{cases} 2 \arcsin \left\{ \frac{B(y)C(y) - \sqrt{B(y)^2 C(y)^2 - (A(y)^2 + B(y)^2)(C(y)^2 - A(y)^2)}}{A(y)^2 + B(y)^2} \right\} & (y < \frac{1}{b}(ab - b^2 - c^2)) \\ 2 \arcsin \left\{ \frac{C(y)}{B(y)} \right\} & (y = \frac{1}{b}(ab - b^2 - c^2)) \\ 2 \arcsin \left\{ \frac{B(y)C(y) + \sqrt{B(y)^2 C(y)^2 - (A(y)^2 + B(y)^2)(C(y)^2 - A(y)^2)}}{A(y)^2 + B(y)^2} \right\} & (y > \frac{1}{b}(ab - b^2 - c^2)) \end{cases}$$

215 The relationship between linear linkage displacement, y , and bending
 216 angle, θ , is illustrated in Fig. 4 (b). For our prototype, where $a = 2.9$
 217 mm, $b = 1.14$ mm, $c = 1.3$ mm, and $0 < \theta < 90$ degrees, y is given by:

$$y = \frac{W_{m1} \cdot t \cdot \Delta p}{\mu_1 \cdot 60} \quad (6)$$

218 where μ_1 is the gear reduction ratio of Motor 1 (which controls
 219 bending), t is time, Δp is the pitch of the lead screw M3 (0.5mm), and
 220 W_{m1} is the frequency of rotation of Motor 1. The relationship between
 221 time, t , frequency of rotation of Motor 1, W_{m1} , and bending angle, θ , is
 222 given in Fig. 4 (c).

223 The setup for scanning a raster pattern over a portion of a spherical
 224 surface is presented in Fig. 4 (d). The length of the raster, L_t ,
 225 achievable in a given time, t , is related to the angle of rotation for each
 226 loop, β_t , and the initial bending angle, θ_{start} , and the range of the
 227 bending angle, $\Delta\theta_t$, by (7):

$$L_t = \beta_t \cdot r \cdot \sin(\theta_{start} + \Delta\theta_t) \quad (7)$$

$$\beta_t = \frac{W_{m2} \cdot \pi}{\mu_2 \cdot 30} \cdot t \quad (8)$$

228 where μ_2 is the gear reduction ratio of Motor 2 which controls rotation, r
 229 is the radius of the sphere, and W_{m2} is the frequency of rotation of
 230 Motor 2.

231 For cases where the lines of the raster don't overlap, the
 232 approximate mosaic area is given by (9), being simply the product of
 233 length of the raster, L_t , and the diameter of the field-of-view of
 234 probe, Δh .

$$S_t = L_t \cdot \Delta h \quad (9)$$

235 The relationship between mosaic area, time and angular rotation for
 236 each loop is illustrated in Fig. 4 (e). Note that the initial bending angle
 237 is 45 degrees, the initial rotational angle is 0 degrees, the frequency of
 238 rotation of Motor 2 is 1000 rpm, and the frequency of rotation of Motor
 239 1 (used for bending) is 3000 rpm.

240 The control diagram of the 1-DOF bending motion of the tip is shown
 241 in Fig. 4 (f). In Fig. 4 (f), θ_i is the angle of Motor 1 driving the linkage,

242 $P(\theta_t)$ is the target linear displacement of the linkage, Y_t and Y_f are the
 243 target and current linear displacements of the linkage respectively, ΔY
 244 is the differential linear displacement of the linkage, and $S(\Delta Y)$ is the
 245 mapping function from displacement ΔY to output voltage, and V and θ_f
 246 are the output voltage and 1-DOF bending angle. The control process
 247 of the 1-DOF rotational motion is similar, but with rotational rather than
 248 linear displacement and using a Hall sensor to for closed loop control.

249
 250 **FIGURE 4. (a) Concept of 1 DOF bending mechanism. (b) Relationship between**
 251 **linkage displacement and bending angle. (c) Relationship between time and**
 252 **bending angle. (d) Co-ordinates for spherical raster. (e) The mosaic area that**
 253 **the device can scan over in a given time for values of angle of rotation, β_t , (f)**
 254 **Control diagram for bending.**

255 *Visualisation and Mosaicing*

256 The endomicroscope imaging probe was built in-house, using a
 257 leached fibre bundle containing 17,000 cores, each essentially
 258 providing a 'pixel' in the final image. As suggested previously [29],
 259 using a leached fibre bundle, which is much more flexible than the
 260 fused bundles typically used in endomicroscopy, enabled the use of full

range of motion of the scanner. However, as discussed previously [29], the leached bundles have a large core-core spacing of $8\text{ }\mu\text{m}$, which limits resolution. Therefore, we coupled a 2X, 0.8 NA magnification micro-lens (GT-MO-080-0415-488, GRINTECH GmbH, Germany) to the distal end of the bundle, enhancing the resolution by a factor of 2 (Fig. 5(b)). The micro-lens and bundle are connected by epoxying inside a laser scattered concentric tube, which is fixed to the slider of the scanner by screws. The lens also increases the working distance from 0 to approximately $80\text{ }\mu\text{m}$ in air, which is important for allowing good quality confocal imaging through the approximately $20\text{ }\mu\text{m}$ thickness of the balloon.

The probe is coupled to a fluorescence confocal endomicroscope of an in-house design and based around a commercial laser scanning confocal system (CLS, Thorlabs USA). It operates at 488 nm wavelength and collects fluorescence emission above 500 nm by a photo-multiplier tube via a pinhole and a fluorescence filter set. A custom interface to an endomicroscope probe was built using a tube lens, a scan lens and a microscope objective, arranged so that a raster pattern was scanned over the distal end of a probe at 20 frames per second. Videos were recorded using the laser scanning unit's software and processed off-line. Each raw image was 800×800 pixels.

In the first stage of processing, the pattern of the fibre cores was removed and fluorescence background from the fibre bundle was removed using an approach similar to that reported in [18]. In brief, a background image was acquired with the probe not in contact with the tissue, and a calibration image was formed by averaging over 200 image frames while the probe was scanning. From the calibration image, the (x,y) pixel co-ordinate of the centre of each fibre core was found and the intensity of each core in the background ($I_{B,i}$) and calibration ($I_{C,i}$) images was extracted. A Delaunay triangulation was then formed over the core positions, allowing each pixel on a reconstruction grid to be associated with an enclosing triangle of three fibre cores (k,l,m) , and its position recorded in barycentric co-

ordinates with respect to the vertices of the triangle $(b_{i,k}, b_{i,l}, b_{i,m})$.

For each image in the video, the core intensities $(I_{raw,i})$ were then extracted by taking the pixel values at each previously calculated core location and then corrected for core-core variations and background signal using:

$$I_i = \frac{I_{raw,i} - I_{B,i}}{I_{C,i}}$$

To reconstruct each pixel on the reconstruction grid, a linear triangular interpolation was made between the corrected intensity values of the three surrounding cores, such that

$$I_{final,i} = I_k b_{i,k} + I_l b_{i,l} + I_m b_{i,m}$$

Finally, each image was cropped to a circle with a diameter of 480 pixels (equivalent to 330 microns on the tissue surface), and resized to 300 x 300 pixels by bicubic interpolation.

FIGURE 5.(a) Optical layout of laser scanning confocal endomicroscopy system, (b) Three quarter section view of distal end.

For mosaicing, the pair-wise normalised cross correlation was calculated between a template extracted from each image and the previous image. The position of the peak of the cross correlation was taken to be the shift between the two images. To create the mosaic,

each image was inserted into the mosaic with the correct shift relative to the previous image using distance-weighted alpha blending to smooth the transition between frames. Correction of distortions due to probe tracking the balloon surface may be achieved using a more sophisticated mosaicing algorithm which does not assume rigid transformations [20]. These algorithms may not be suitable for real time image reconstruction [31], but could be used for retrospective analysis. However, development of such algorithms is not the focus of this paper.

RESULTS

Mechanical Performance Evaluation

We evaluated bending and rotating characteristics of the 2-DOF scanning mechanism. These measurements were made with the imaging probe inserted into the channel. Repeatability measurements were performed over five trials, measuring actual bending and rotational angles against the target angles. For assessment of the bending performance, measurements were made in two parts: (a) bending from 0 degrees to 90 degrees, and (b) returning to 0 degrees. Backlash compensation was made by inverse rotation of Motor 1 when changing the bending direction. The bending angles were measured by a high-resolution digital camera set up over the tip. The accuracy of the camera image was checked by capturing a regular grid image (pitch 0.05 mm) before the experiments. No distortion was visible and, thus, the error caused by distortion was assumed to be negligible. The measured hysteresis curve is shown in Fig. 6(a), and the relationship between the target angle and bending repeatability error is given in Fig. 6(b). The measured values of bending range, bending repeatability error (standard deviation), and tip positioning accuracy are presented in Table I.

We also tested in detail the performance of the rotational motion. The scanning device was first rotated from 0 to 360 degrees (c), and

345 then returned to 0 degrees (d) (Fig. 6(c - d) and Table II). Moreover, we
 346 tested the following capability of rotating motions between 0 and 360
 347 degrees (Fig. 6(e)). The frequency of rotation of Motor 2 was 1000 rpm.
 348

349 **FIGURE6.Results of 2-DOFs scanning characteristics test, showing the**
 350 **relationship between target angles and actual bending and rotational angles.**
 351 **(a) 1-DOF target bending angle against measurement angle, (b) relationship**
 352 **between target bending angle and bending repeatability error, (c) 1-DOF target**
 353 **rotational angle against measurement angle, (d) relationship between target**
 354 **rotational angle and rotational repeatability error, (e) following capability of**
 355 **rotational motions between 0 and 360 degrees.**
 356

357

358

359 **TABLE 1.Result of repeatability measurement in each bending**
360 **mechanism.**

Measurement item	(a)	(b)
Bending range (°)	0.26 to 90.10	90.10 to -0.692
Repeatability (°)	0.60	0.48
Tip positioning accuracy (mm)	0.26	0.2

361

362 **TABLE 2. Result of repeatability measurement in rotating**
363 **mechanism.**

Measurement item	(c)	(d)
One circle rotational range (°)	0 to 359.96	359.96 to -0.36
Repeatability (°)	0.39	0.40
Tip positioning accuracy (mm)	0.17	0.17

364

365

366 **TABLE 3.Generated power and torque at tip.**

DOF	Direction (°)	Power [N]	Torque [Nmm]
Bending	0 to 90	0.6	15
Rotating (Bending 45°)	-360 to 360	1.6	40

367

368 *Breast Phantom Experiment*

369 For general performance evaluation, we simulated use of the system
370 during breast conserving surgery using a silicon breast phantom as
371 shown in Fig. 7(a). The breast phantom contained a realistic cavity as it
372 had previously been used for surgical training for this procedure. To
373 allow microscopic imaging, the cavity was lined with tissue

374 paperstained with acriflavine. The scanning device was attached to a
 375 passive arm during experiment.

376
 377 **FIGURE 7. Breast phantom experiment results showing (a) the experimental set**
 378 **up: the cavity was lined with tissue paperstained with acriflavine. The scanning**
 379 **device was attached to a passive arm during experiment, (b) scanning position:**
 380 **four parts of the hemisphere (90 degrees apart) were targeted; the spacing**
 381 **between each line was set at 0.25 mm, and the angle of rotation for each loop at**
 382 **0.1 turn which is 36 degrees, and (c-f) raster mosaics derived.**

383
 384 We performed raster scans using the rotational motion as the fast
 385 axis and the bending motion as the slow axis. The starting position and
 386 scanning area are shown in Fig. 7 (b). We scanned four parts of the
 387 hemisphere (90 degrees apart); the spacing between each line was set
 388 at 0.25 mm, and the angle of rotation for each loop at 0.1 turn which is
 389 36 degrees (Fig. 7 (b)). Fig. 7(c-f) shows the four mosaics which cover
 390 an area of approximately 25.0 mm², 25.9 mm², 15.5 mm² and 17.8

391 mm². In Fig. 7(c-f), the intended automatic scan areas are 33.1 mm²,
392 36.9 mm², 19.2 mm², and 21.3 mm² respectively. Thus, the ratio of the
393 covered area to the intended area are 0.75(in Fig. 7 (c)), 0.7 (in Fig. 7
394 (d)), 0.8 (in Fig. 7 (e)), and 0.84 (in Fig. 7 (f)) respectively.

395 The results show that the probe could maintain almost constant
396 tissue contact when scanning over large area on different parts of
397 hemisphere surface. There was distortion in the mosaics, primarily due
398 to deformation of the phantom during scanning. This deformation affects
399 repeatability, and prevents scanning over an exactly defined area, but
400 we were able to extract mosaics covering significant areas. This
401 demonstrates that, in principle, large area imaging inside a realistic
402 breast cavity phantom is possible. This result is also important because
403 it demonstrates that the mechanism, including the inflated balloon,
404 allows reshaping and scanning of irregular surfaces, as would be
405 encountered in practice.

406 *Ex vivo Human Breast Tissue Experiments*

407 The above demonstrated a capability to image within a breast-cavity,
408 but the imaging phantom was not representative of breast tissue. We
409 therefore performed imaging studies using freshly excised, acriflavine-
410 stained human breast tissues in order to confirm that large mosaics
411 could be formed. The subject gave prior written informed consent and
412 Human Tissue Authority licence and ethics approval was obtained
413 through Imperial College Tissue Bank (No. CX803886). As only a small
414 portion of tissue was available (approx. 20 x 10 mm), we placed this
415 inside a hemispherical plastic shell (radius 30 mm) to provide an
416 approximate simulation of the curvature of the breast cavity (Fig. 8(a)).
417 We performed raster scans, with the spacing between raster lines set at
418 0.25mm(Fig. 8(b)).The scanning device was hold by passive arm during
419 experiment.

420 Examples of mosaics extracted from these experiments are shown in
421 (Fig. 8(c-e)). Fig 8(c-d) are for angle range of 36 degrees, while Fig 8(e)
422 is for an angle range of 45 degrees. These images demonstrate that the

key morphological features of normal breast tissue can be identified. Most prominent are the adipocytes, the large polygonal-shaped fat cells, while areas of fibrous tissue can also clearly be seen. When compared with the our previous results of scanning using a Gastroflex UHD Probe (Mauna Kea Technolugues) [29], this provides indicates that the use of a highly flexible leached bundle probe, together with a distal micro-lens, could provide adequate image quality.

FIGURE8. Human breast tissue mosaic images, (a) Setup, (b) Scanning position, (c– e) Raster mosaic results of human breast tissue.

433 Fig. 8(c) shows a mosaic that covers an area of approximately 21.5
434 mm². The ratio of the covered area to the intended area (34.1 mm²) is
435 0.63. Fig. 8 (d) shows a second scan, where the raster has an area
436 approximately 19.8 mm². The extended area is 27.7 mm², resulting in
437 an area ratio of 0.71. We observed that scan here is deformed and
438 sheared; the main cause is likely to be movement of the specimen
439 during scanning.

440 Finally, Fig. 8(e) is a mosaic covering an area 34.8 mm² and the ratio of
441 the covered area with respect to the intended area (38.69 mm²) is
442 0.9. The very large image mosaics that could be generated allow for a
443 much more global appreciation of the tissue morphology than can be
444 achieved through individual images or small, manual mosaics.

445 **DISCUSSION**

446 In this paper, we have developed a miniature robot scanning probe
447 for in vivo application of endomicroscopy for breast cancer surgery. By
448 using the scanning device, we achieved mosaics approaching 35 mm²
449 in size. Previous studies such as [22] were targeted towards generation
450 a continuous mosaic of less than 3 mm², and so this device represents
451 a significant increase in area coverage. Key novelties in this work
452 include the incorporate of an inflatable balloon and design of an
453 endomicroscope with sufficient flexibility to image anywhere on the
454 hemisphere and with a working distance sufficient to image through the
455 balloon.

456 It has been shown that the scanning device can achieve a
457 repeatability of 0.6 degrees (bending motion) and 0.40 degrees
458 (rotational motion). This corresponds to positional accuracies of 0.26
459 mm and 0.17 mm at the tip, respectively. This is comparable to the
460 field-of-view of the endomicroscope probe (0.33 mm) and far smaller
461 than any likely tissue deformation effects. Therefore in practice it may
462 be necessary to over-sample to ensure complete tissue coverage. It is
463 important to achieve speedy scanning, thus, we selected the maximum
464 acceptable scanning velocity which could obtain consistent imaging

465 mosaic by experiments. For example, it took almost 50 seconds to
466 reach the targeted scanning area 30 mm^2 as shown in Fig. 4 (e). At
467 present, the waiting time for other methods of examining tumor margins
468 such as frozen section can be as long as 40 minutes. Intraoperative
469 specimen x-rays may take as long as 5-10 minutes but it has low
470 accuracy rates. The ability to obtain good imaging quality within a
471 minute is certainly well for in vivo.

472 We used the device to scan raster patterns on a spherical surface,
473 with the rotational motion used for the fast axis and the bending motion
474 used for the slow axis. The achievable bending range was slightly over
475 90 degrees due to overrun of the linkage, but that doesn't affect the
476 practical performance of the device. The bending and rotating forces
477 were measured to be 0.6 N and 1.6 N respectively. This was evidently
478 sufficient in order to carry out a scan inside the balloon.

479 The extension and deformation of the spur gear's engagement parts
480 could be minimized by increasing the thickness of the spur gear.
481 However, the backlash was small because the spur gears are used for
482 guiding the bending motion instead of driving it. Moreover, backlash
483 often occurs when changing the rotational direction of the gears; when
484 forming the slow axis of a raster the motion was in a single direction
485 only. Nevertheless, the quality of the connecting parts of the linkage
486 could be improved in future. The positioning inaccuracies could be
487 reduced by increasing the precision and stiffness of the gear and
488 linkage. Because the inner air pressure is 140kPa which is a low
489 pressure, the tissues will not be damaged even there is an air leakage.
490 Using an aqueous to inflate the balloon is another option which has
491 potential to provide better refractive index matching minimizing internal
492 reflections.

493 Experiments with the realistic breasts phantom showed that, in
494 principle, the probe can maintain contact with irregular surfaces. This is
495 helped greatly by the inflatable balloon that tends to reshape the
496 surface. The balloon could be moved due to reaction force exerted by
497 tissue and probe. A passive linear structure is used to solve this
498 issue. The passive linear structure with a working distance 3 mm,

499 meaning that the hemispherical radius can vary by 3 mm at the tip
500 which could cover a certain level of distance. We show that, thanks to
501 the contact modulation effect of the passive linear structure and the
502 reshaping effect of the balloon, the probe can maintain contact over
503 large area during phantom experiment. We observed failures to image
504 some parts of the phantom. It is therefore unlikely that an entire
505 hemisphere could be imaged in a single shot, and the device would
506 need to be positioned to as to optimize contact for a particular region of
507 the cavity. Further work will be needed to explore the optimal workflow
508 in this regard. It may also be necessary to optimize the size of the
509 balloon and the length of the bending tip for different sized cavities. A
510 range of differently sized balloons could be used to suit different
511 cavities. For initial work, we targeted a 5 cm diameter cavity as; in
512 general, a smaller cavity provides more challenges in terms of
513 miniaturization and robustness.

514 In the *ex vivo* human tissue evaluation, the device was shown to be
515 able to scan the tissue stably. These results demonstrate that when
516 contact is maintained, the motion of the probe and the image quality
517 are suitable for generating large area mosaics from breasts tissue.
518 Except one very successful mosaics (Fig. 8(e)), the rate of area
519 covered by the mosaic to the intended area in *ex vivo* experiments is
520 smaller (0.63 and 0.71 in Fig. 8(c-d)) than those with the breast
521 phantom mosaics performed in Fig 7 (average ratio is 0.77). There are
522 two reasons. The first is that the breast phantom with tissue paper is
523 less sticking and deformation than human breast tissue. The second
524 reason is the stabilizing effect of the balloon. The inflation of the
525 balloon causes the tissue to deform and take on the shape of the
526 inflated balloon that provides for stabilization of the tissue. There will be
527 tissue distortion caused by the inflated balloon and scanning probe. To
528 an extent this problem is unavoidable, as any contact based imaging
529 method will result in tissue deformation if the probe is driven across the
530 tissue. Scanning the internal cavity surface directly with only the probe
531 could lead to large tissue deformation and losing tissue contact since
532 the excised tissue cavity is highly irregular. On the other hand, the

533 distortion of the cavity surface by inflated balloon works as a “positive
534 deformation” which could stabilize the irregular tissue cavity and limit
535 uncontrollable tissue deformation. Further work will be needed to
536 determine the effect of this on image interpretation. In future, this could
537 include an automated balloon shaping mechanism using pressure
538 control to allow a precise fit to the size of the cavity. Closed loop control
539 of the device, using visual servoing based on position estimates from
540 the image registration, is likely to be necessary in to progress further.

541 Our miniature balloon scanning device is a smart, cost-effective
542 platform to generate accurate mosaics in real-time to aid intraoperative
543 decision making. A particular benefit is the simple mechanical design,
544 which will make commercial deployment feasible.

545 **CONCLUSION**

546 In this paper, we have presented and characterised a miniature
547 scanning device for microscopic imaging of the walls of the cavity
548 created during breast conserving surgery. Its novel features include a
549 small diameter, a custom and highly flexible endomicroscopy probe to
550 allow use of the full range of motion, an inflatable balloon to provide a
551 more regular surface for imaging, and its ability to scan over a large
552 surface area. We demonstrated the possibility of reshaping and
553 scanning over irregular surfaces using a realistic breast cavity
554 phantom, and the ability to form mosaics over breast morphology using
555 *ex vivo* human tissue. The resulting mosaics show that the 2-DOF
556 scanning approach, when combined with the inflatable balloon
557 mechanism, allows imaging at high resolution over a large surface
558 area. Despite the use of a balloon, deformation of the tissue remains a
559 challenge and further research to tackle this problem will be needed if
560 repeatability is to be improved. Nevertheless, these results
561 demonstrate the potential clinical value of the device to improve the
562 prospects for intraoperative cavity margin evaluation, which in future
563 could lead to improved outcomes for breast cancer patients.

564 **ACKNOWLEDGMENTS**

565 The authors would like to thank Dr. Daniel R Leff and Vyas Khushi for
566 providing the breast tissue and discussion for ex vivo experiments, and
567 to Petros Giataganas for discussions with mechanical design. This
568 work was supported by EPSRC grant EP/IO27769/1: SMART
569 Endomicroscopy.

570 **CONFLICT OF INTEREST**

571 None.

572

573 **REFERENCES**

- 574 [1] Ferlay, J. et al. Cancer incidence and mortality worldwide: sources,
575 methods and major patterns in GLOBOCAN 2012. *Int. J. Cancer*
576 136: E359–E386, 2015.
- 577 [2] Schwartz, G.F., U. Veronesi, K.B. Clough, et al. Consensus
578 conference on breast conservation, *J. Am. Coll. Surg.* 203: 198-207,
579 2006.
- 580 [3] Singletary, S.E. Surgical margins in patients with early-stage breast
581 cancer treated with breast conservation therapy, *Am. J. Surg.* 184:
582 383-393, 2002.
- 583 [4] Kreike, B., A. A. Hart, T. van de Velde, J. Borger, H. Peterse, E.
584 Rutgers, H. Bartelink, and M. J. van de Vijver. Continuing risk of
585 ipsilateral breast relapse after breast-conserving therapy at long-
586 term follow-up, *International journal of radiation oncology, biology,*
587 *physics* 71: 1014-1021, 2008.
- 588 [5] Xu M., L.V.Wang. Photoacoustic imaging in biomedicine. *Rev*
589 *SciInstrum* 77:041101, 2006
- 590 [6] Jabbour, J. M., M. A. Saldua, J. N. Bixler. And K.C. Maitland.
591 Confocal Endomicroscopy: Instrumentation and Medical
592 Applications, *Ann. Biomed. Eng.* 40:378–397, 2011.
- 593 [7] Pohl, H, T. Rosch, M. Vieth, M. Koch, V. Becker, M. Anders, A.C.
594 Khalifa, and A. Meining. Miniprobe confocal laser microscopy for
595 the detection of invisible neoplasia in patients with Barrett's
596 oesophagus. *Gut* 57:1648–1653, 2008.
- 597 [8] Newton, R.C., S.V. Kemp, G.Z. Yang, A. Darzi, M.N. Sheppard, and
598 P.L. Shah. Tracheobronchial Amyloidosis and Confocal
599 Endomicroscopy. *Respiration* 82(2):209–211, 2011.
- 600 [9] Newton, R.C., S. Kemp, Z. Zoumot, G.Z. Yang, A. Darzi, and P.L.
601 Shah. An unusual case of haemoptysis. *Thorax* 65:309, 353, 2010.

- 602 [10] Newton, R.C., S.V. Kemp, G.Z. Yang, D. Ellson, A. Darzi, and P.
603 Shah. Imaging parenchymal lung diseases with confocal
604 endomicroscopy. *Respiratory Medicine* 106(1):127–137, 2012.
- 605 [11] Newton, R.C., S.V. Kemp, P. Shah, D. Elson, A. Darzi, K.
606 Shibuya, S. Mulgrew, and G.Z. Yang. Progress toward optical
607 biopsy:bringing the microscope to the patient. *Lung* 189:111–119,
608 2011.
- 609 [12] Gmitro, A. F., and D. Aziz. Confocal microscopy through a fiber-
610 optic imaging bundle, *Optics Letters*, 18: 565-567, 1993.
- 611 [13] Le Goualher, G., A. Perchant, M. Genet, C. Cave, B. Viellerobe,
612 F. Berier, B. Abrat, and N. Ayache. Towards optical biopsies with an
613 integrated fibered confocal fluorescence microscope. Part II. In:
614 Proceedings of the 7th International Conference on Medical Image
615 Computing and Computer-Assisted Intervention, Saint-Malo, France,
616 pp. 761–768, 2004.
- 617 [14] Laemmel, E., M. Genet, G. Le Goualher, Perchant A, J.F. Le
618 Gargasson, and E. Vicaut, Fibered confocal fluorescence
619 microscopy (Cell-viZio™) facilitates extended imaging in the field of
620 microcirculation, *Journal of Vascular Research* 41(5): 400-411,
621 2004.
- 622 [15] Abeytunge, S., Y. Li, B. Larson, G. Peterson, E. Seltzer, R.
623 Toledo-Crow, et al. Confocal microscopy with strip mosaicing for
624 rapid imaging over large areas of excised tissue. *Journal of*
625 *biomedical optics* 18: 061227, 2013.
- 626 [16] Chang, T. P., D. R. Leff, S. Shousha, D. J. Hadjiminis, R.
627 Ramakrishnan, M. Gudi, R. Al-Mufti, M. R. Hughes, A. Darzi, and G.
628 Z. Yang. Imaging of breast cancer morphology using probe-based
629 confocal laser endomicroscopy: Towards a novel imaging tool for
630 real-time intra-operative cavity scanning, *Eur J Surg Oncol.* 39(11):
631 S80, 2013.
- 632 [17] Tilli, M. T., M. C. Cabrera, A. R. Parrish, K.M. Torre, M.K. Sidawy,
633 A.L. Gallagher, E. Makariou, S.A. Polin, M.C. Liu, and P.A. Furth.
634 Real-time imaging and characterization of human breast tissue by
635 reflectance confocal microscopy, *Journal of biomedical optics.* 12:
636 051901, 2007.
- 637 [18] Jeevan, R., D. Cromwell, M. Trivella, G. Lawrence, O. Kearins, J.
638 Pereira, C. Sheppard, C.M. Caddy, J.H. van der Meulen.
639 Reoperation rates after breast conserving surgery for breast cancer
640 among women in England: retrospective study of hospital episode
641 statistics,” *BMJ: British Medical Journal*, 345, 2012.
- 642 [19] Vercauteren, T., A. Perchant, X. Pennec, and N. Ayache.
643 Mosaicing of Confocal Microscopic In Vivo Soft Tissue Video
644 Sequences. In: *Medical Image Computing and Computer-Assisted*
645 *Intervention—MICCAI 2005*, pp. 753–760, 2005.
- 646 [20] Vercauteren, T., A. Perchant, G. Malandain, X. Pennec, and N.
647 Ayache. Robust mosaicing with correction of motion distortions and
648 tissue deformation for in vivo fibered microscopy. *Medical Image*
649 *Analysis* 10(5): 673–692, 2006.
- 650 [21] Mahé, J., T. Vercauteren, B. Rosa, and J. Dauguet. A Viterbi
651 approach to topology inference for large scale

652 endomicroscopyvideo mosaicing, In Medical Image Computing and
653 Computer-Assisted Intervention–MICCAI 2013, pp. 404-411, 2005.

654 [22] Rosa, B., B. Herman, J. Szewczyk, B. Gayet, and G. Morel.
655 Laparoscopic optical biopsies: in vivo robotized mosaicing with
656 probe-based confocal endomicroscopy, in Proc. of IROS'2011, San
657 Francisco, California, pp. 25-30, 2011.

658 [23] Erden, M. S., B. Rosa, N. Boularot, B. Gayet, G. Morel, and J.
659 Szewczyk. Conic-Spiraleur: A Miniature Distal Scanner for Confocal
660 Microlaparoscope, IEEE/ASME transactions on mechatronics 19(6):
661 1786-1798, 2014

662 [24] Dario, P., M. Carrozza, C. Marcacci, M. Attanasio, B. Magnani,
663 O. Tonet, and G. Megali. A Novel Mechatronic Tool for Computer-
664 Assisted Arthroscopy," IEEE Trans. Inform. Technol. Biomed. 4(1):
665 15–28, 2000.

666 [25] Zuo, S., K. Iijima, T. Tokumiya, and K. Masamune. Variable
667 stiffness outer sheath with "Dragon skin" structure and negative
668 pneumatic shape-locking", International Journal of Computer
669 Assisted Radiology and Surgery9(5): 857-865, 2014

670 [26] Shang, J., D. Noonan, C. Payne, J. Clark, M. Sodergren, A.
671 Darzi, and G.Z. Yang. An articulated universal joint based
672 flexibleaccess robot for minimally invasive surgery, Proc. IEEE Int.
673 Conf. Robotics Automation, pp. 1147–1152, 2011.

674 [27] Peirs, J., D. Reynaerts, H. Van Brussel. A miniature manipulator
675 for integration in a self-propelling endoscope, Sensors and
676 Actuators A.: 343–349, 2001

677 [28] Yamashita, H., D. Kim, N. Hata, T. Dohi. Multi-slider linkage
678 mechanism for endoscopic forceps manipulator, Proc. IEEE/RSJ Int.
679 Conf. Intell. Robots Syst., pp. 2577-2582, 2003.

680 [29] Yamashita, H., K. Matsumiya, K. Masamune, H. Liao, T. Chiba,
681 T. Dohi. Miniature bending manipulator for fetoscopic intrauterine
682 laser therapy in twin-to-twin transfusion syndrome. Surgical
683 Endoscopy 22(2): 430–435,2007

684 [30] Zuo, S., M. Hughes, C. Seneci, T.P. Chang, G.Z. Yang. Towards
685 Intraoperative Breast Endomicroscopy with a Novel Surface
686 Scanning Device, *IEEE Trans. Biomed. Eng.*,
687 doi:10.1109/TBME.2015.2455597, 2015

688 [31] Vercauteren, T., A. Meining, F. Lacombe, A. Perchant, "Real
689 time autonomous video image registration for endomicroscopy:
690 fighting the compromises," In *Biomedical Optics (BiOS) 2008*, pp.
691 68610C-68610C, 2008.