Early-stage Dielectric Characterisation of Renal Cell Carcinoma for Positive Surgical Margin Detection

Alessandra La Gioia[1], Adnan Elahi[1], Anna Bottiglieri[1], Niko Ištuk[1], Catherine Dowling[2], Frank D'Arcy[2], Martin O'Halloran[1] and Emily Porter[1]

[1] Translational Medical Device Laboratory, National University of Ireland Galway, Galway, Ireland, a.lagioia1@nuigalway.ie

[2] Urology Department, University Hospital Galway, Galway, Ireland

***Abstract*—Partial nephrectomy is preferred to total nephrectomy for clinically localised renal cell carcinoma. In order to minimise the risk of local tumour recurrence after partial nephrectomy, ideally, the surgical margins of the excised sample should be negative for the disease. Currently, the risk of positive margins during partial nephrectomy is minimised with the use of intraoperative ultrasound. In this study, dielectric spectroscopy is proposed for the detection of positive margins during partial nephrectomy. Specifically, the feasibility of using an open-ended coaxial probe operating at microwave frequencies is evaluated for *in vivo* differentiation between positive and negative surgical margins. Due to the lack of dielectric properties of renal cancerous tissue in the literature, early stage *ex vivo* dielectric measurements were conducted on five human renal samples immediately after excision. A wide range of dielectric measurement results were obtained due to the heterogeneity of renal samples and the different longitudinal location of the cancerous tissue across the samples. This outcome suggests the need to refine the protocol for dielectric characterisation of renal cell carcinoma and highlights the limitations of a coaxial probe at detecting renal tumour margins.**

***Index Terms*—renal cell carcinoma, surgical margin detection, dielectric spectroscopy, open-ended coaxial probe.**

I. Introduction

The incidence of renal cell carcinoma (RCC) has been increasing over the past several decades [1]. RCC is often diagnosed at an early stage and then is suitable for partial nephrectomy, which is the accepted surgical treatment for localised RCC [1]. Partial nephrectomy can be either conducted by open surgery or laparoscopically. In both cases, a layer of healthy tissue is also removed in order to avoid positive surgical margins. Wide negative margins have traditionally been preferred to thin negative margins to maximise the efficacy of the surgery [2]. However, recent studies have demonstrated that the size of the surgical margin in partial nephrectomy does not affect the risk of local tumour recurrence [2], [3]. In particular, it was observed that any surgical margin, even with a width of less than 1 mm, can be adequate as long as the entire cancerous region is removed [2]. Smaller surgical margins have the advantage of decreasing morbidity and post-operation injuries [3].

Currently, the margin assessment is performed with the use of intraoperative ultrasound, and the risk of positive margins can be reduced with adequate hilar clamping, embolization or with cold-scissor parenchymal transection [4]–[6].

In this study, the applicability of dielectric spectroscopy for *in vivo* differentiation of surgical margins has been investigated. There are several techniques that can be used for measuring the dielectric properties of biological tissues, such as tetrapolar impedance, cavity perturbation, transmission line, and open-ended coaxial probe techniques [7]. However, the open-ended coaxial probe has been selected for renal cell carcinoma detection in this study, since this technique is simple, broadband, non-destructive, requires minimal sample handling, and allows for both *ex vivo* and *in vivo* measurements [7]. Specifically, this study evaluated the feasibility of using the Keysight slim form probe [8] for tumour margin detection. The choice of the slim form is due to the small size of the probe, which has a diameter of only 2.2 mm, and thus a small sensing volume [9], [10]. In fact, such a sensing volume has the potential to easily detect the tumour interface and thereby minimise the width of negative margins.

In order to dielectrically detect the position of the RCC interface with healthy tissue, it is necessary to know the dielectric properties of the cancerous tissue with respect to the dielectric properties of the healthy tissue. However, existing data is limited, with only one study reporting the dielectric properties of renal cancerous tissue up to 0.9 GHz. Therefore, in this study, *ex vivo* dielectric measurements were conducted to characterise human renal cancerous and healthy tissue regions in the microwave range between 0.5 and 8.5 GHz. Due to clinical reasons, during the dielectric measurement, it was not possible to section the sample to better localise the cancerous region. However, the identification of the cancerous region was supported by the surgeon during the measurement and by histology after the measurement. Details about the measured samples and the dielectric measurement protocol are reported in the following section before discussing the outcome of the study.

II. Methodology

A. Characteristics of the measured samples

This study was designed to minimise influence on normal clinical procedures and the treatment pathway of patients. The study proposal was approved ethically and patients whose tissue samples are included in the study gave informed consent.

TABLE I
PATIENT INFORMATION AND SAMPLE HISTOLOGICAL CHARACTERISTICS

Patient #	*Age*	*Cancer type*	*Histological observations*
Patient 1	52	Cystic papillary RCC type 1	Cyst with max diameter of 42 mm and sparse lesions located beneath the capsule
Patient 2	82	Clear cell RCC grade 2	Centrally located tumour of max diameter 82 mm surrounded by fat and pushing to the capsule
Patient 3	55	Clear cell RCC grade 2	Tumour with max diameter of 20 mm surrounded by thin layer of fat pushing to the capsule
Patient 4	58	Clear cell RCC grade 2	Tumour with max diameter of 21 mm surrounded by fibrous capsule and neocapsule
Patient 5	57	Clear cell RCC grade 2	Tumour with max diameter of 38 mm presenting necrotic regions close to the capsule

Five renal samples were excised from five patients. Four nephrectomies were radical (i.e., the full kidney was removed), and one was partial. The patients were all male, with four aged between 52 and 58, and one with an age of 82. Four samples showed clear cell RCC, the most common renal cell cancer [11], and one sample presented cystic papillary RCC, the second most common renal cell cancer [11]. The patient information and the RCC histological details for each sample are reported in Table I.

As is clear from Table I, each sample presented different histological characteristics. The heterogeneity of the samples is better illustrated in Fig. 1, where pictures of the samples from patient 1 and patient 5 are reported. On each sample, the RCC location is marked with a black contour. Details on the dielectric measurement procedure adopted to characterise the five renal samples is reported in the following subsection.

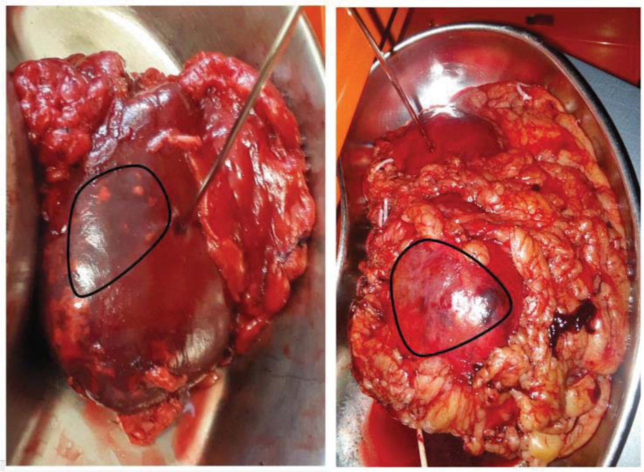

Fig. 1. On the left, kidney sample from patient 1; on the right, kidney sample from patient 5. In both pictures, the probe is in contact with healthy tissue. However, the cancerous area is marked with a black contour.

B. Dielectric measurement protocol

In this study, dielectric measurements were performed using the Keysight slim form probe connected to the Agilent E5063A network analyser. Before each set of measurements, the system was calibrated using the three-load standard procedure.

The uncertainty of the measurement system was characterised after calibration by performing repeated measurements of 0.1 M NaCl (saline), one of the most commonly used reference liquids [12]. Data from multiple 0.1 M NaCl measurements (performed after different system calibrations) was used to quantify the total combined uncertainty (TCU) of the measurement system for both the relative permittivity ϵ_r, and the conductivity, σ. The TCU was calculated as reported in [12]. The TCU was found to be 2.1% for relative permittivity and 4.2% for conductivity [9].

Based on the knowledge of the system performance, the quality of the calibration was validated after each calibration instance by measuring the dielectric properties of the 0.1 M NaCl solution. The measured dielectric signal was then compared to the molarity- and temperature-dependent Cole-Cole model in [13], which was implemented from measurements conducted, at different temperatures, on saline solutions with different molarities. Thus, the average percent difference between the model of 0.1 M NaCl at the measurement temperature and the dielectric data measured from 0.1 M NaCl was calculated to obtain the measurement accuracy and verify that the accuracy was within the TCU. Specifically, the accuracy of saline measurements resulted to be within the TCU throughout each calibration instance, thus demonstrating that the dielectric measurements were not affected by significant errors. All measurements were acquired as soon as possible after calibrating the system in order to reduce noise due to VNA drift. Measurements were recorded with 101 frequency points on a linear scale over the microwave frequency range of 0.5–8.5 GHz. This bandwidth was selected by considering the operating frequency range of the probe and the VNA. The temperature of the calibration and validation liquids were recorded for each calibration and validation.

Furthermore, each measurement was performed by bringing the sample towards the probe using a lift table in order to avoid system perturbations that could introduce errors in the measured data. For each renal tissue sample, the time of excision and the measurement time were recorded. Experiments were conducted in the operating room (a temperature controlled environment) as soon as the sample was excised. Whenever it was not possible to perform the measurement immediately after the excision for logistical reasons, the sample was stored in a closed container to prevent tissue dehydration. Nonetheless, the measurements were always completed within 50 minutes of the excision. Multiple measurements (from three up to five measurements) were taken across both healthy and cancerous regions of the samples, which were indicated by the surgeon. For each tissue measurement, an appropriate probe-tissue contact was ensured by visual inspection and by applying a steady pressure as suggested in [7]. Specifically, repeated measurements were taken at the same point after the application of a low probe-tissue pressure. If the dielectric traces obtained from the repeated measurements were not consistent (due to air gaps), the probe-contact pressure was gradually increased until measurements at the same location were repeatable. Between measurements taken from different regions, the probe tip was cleaned with alcohol wipes to avoid contamination. Also, the temperature of each region was recorded with an infrared thermometer, and excess blood on the surface was removed with cotton swabs.

In order to facilitate the data interpretation, in Table II, the measurement time from excision and the temperature range recorded during each set of measurements are reported for each sample. Specifically, in Table II, the measurement time from excision is reported as a range to cover the first and last measurements, and the temperature range indicates a drop in temperature during the measurement.

TABLE II
MEASUREMENT TIME FROM EXCISION AND TEMPERATURE FOR EACH MEASURED SAMPLE

Sample #	*Time from excision [min]*	*Temperature [°C]*
Sample 1	30–50	23.5–24.1
Sample 2	18–37	25.8–26.8
Sample 3	19–40	26.0–28.1
Sample 4	20–38	26.5–28.0
Sample 5	5–20	28.1–33.0

Lastly, the measured data across all samples was analysed and reported in terms of mean and standard deviation. Also, two pole Cole-Cole models were fitted to the dielectric traces using the minimum squared error algorithm. The dielectric traces are reported and discussed in the following section.

III. RESULTS AND DISCUSSION

In Fig. 2, the average Cole-Cole models obtained across all samples are illustrated. The parameters of each average Cole-Cole model are summarised in Table III. The average fractional error between the measured data and the fitted model in all cases was less than 0.7%.

In both relative permittivity and conductivity plots of Fig. 2, the blue dash-dotted trace and the blue dotted traces are obtained from the mean and standard deviation calculated across the dielectric signals from the healthy regions of all samples. Specifically, the blue dash-dotted line refers to the mean dielectric values, and the blue dotted lines indicate two standard deviations from the mean values.

In Fig. 2, all the solid traces are average dielectric values obtained across the cancerous regions of each of the five samples. In the plots, healthy tissue dielectric properties from [14], and both healthy and cancerous tissue dielectric properties from [15], are also reported. The renal healthy tissue dielectric properties are consistent with the data from Gabriel *et al.* [14]. Specifically, the average relative permittivity and conductivity are approximately 1.9% and 5.5% lower than the reference data. Furthermore, the dielectric values from the healthy region were obtained by positioning the probe on the capsule. In fact, the acquired dielectric properties match the dielectric properties obtained from measurements on porcine kidney capsule in [16].

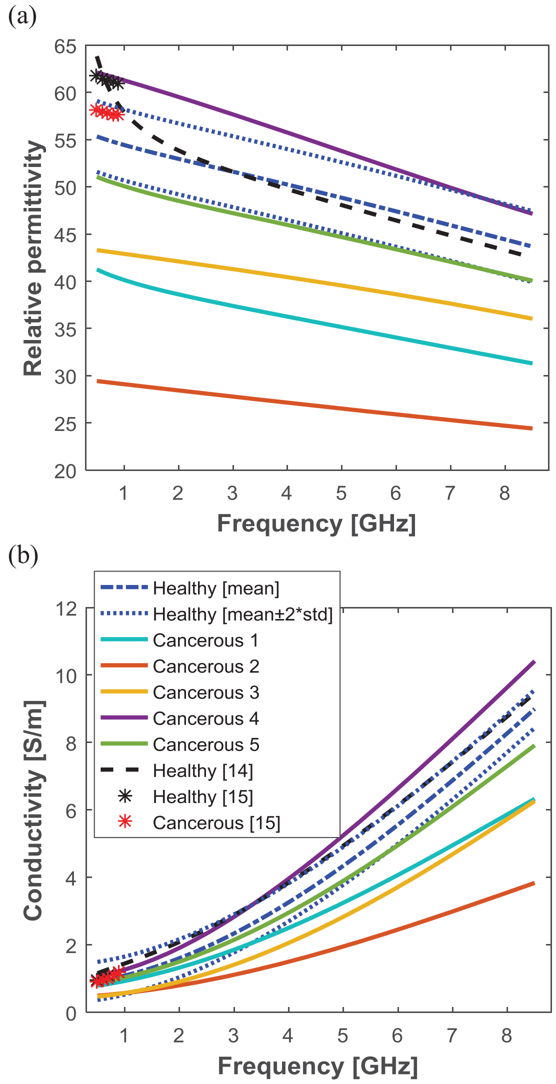

Fig. 2. (a) Relative permittivity and (b) conductivity traces obtained by fitting a two pole Cole-Cole model into the data measured from the five excised samples. In both plots, the renal healthy tissue mean and standard deviation traces were obtained from the dielectric traces from all samples. Also, the average dielectric properties obtained across the cancerous region of each sample are reported. For a comparison with data from the literature, healthy tissue dielectric properties from [14], and both healthy and cancerous tissue dielectric properties from [15], are plotted.

On the other hand, none of the dielectric traces in Fig. 2 are in agreement with the dielectric data from [15]. From both relative permittivity and conductivity values plots, it is clear that the dielectric properties of the cancerous regions are not consistent across samples and span a wide range of values.

The large difference in terms of dielectric properties across the cancerous tissue region is due to the renal sample heterogeneity and to the small sensing volume of the probe. Specifically, as for healthy tissue regions, the cancerous tissue measurements were performed by positioning the probe on the capsule. Considering that, as specified in Table I, all cancerous regions were located underneath the capsule and, in some cases (as for sample 2 and sample 3), they were also surrounded by a layer of fat, it is likely that the acquired signal is a combination of the dielectric properties of all tissues occupying the sensing volume of the probe. Furthermore, previous studies, which demonstrated that the dielectric properties acquired with the slim form probe from layered structures are mostly influenced by the tissues within 0.2 mm from the probe tip [9], suggest that the dielectric contribution of the cancerous tissue to the acquired signal can be small compared to the dielectric properties of the tissues above (i.e., capsule, fat).

Thus, for a more accurate dielectric analysis of renal cancerous samples, the findings of this study suggest a refinement of the dielectric measurement protocol by sectioning the excised renal sample to ensure that the probe is positioned directly on the cancerous region and no other tissue affects the acquired dielectric signal.

TABLE III
TWO POLE COLE-COLE MODEL PARAMETERS FOR THE DIELECTRIC TRACES REPORTED IN FIG. 2

Parameters	*Healthy*	*Cancer 1*	*Cancer 2*	*Cancer 3*	*Cancer 4*	*Cancer 5*
ϵ_∞	1.38	2.70	2.53	9.30	2.51	2.40
$\Delta\epsilon_1$	23.70	19.80	13.66	30.17	30.22	23.94
τ_1 [ps]	6.47	9.70	5.64	8.23	8.63	6.40
α_1	0.44	1.66e-4	0.21	1.37e-5	0.16	0.52
$\Delta\epsilon_2$	32.06	22.38	13.66	4.32	30.21	27.38
τ_2 [ps]	8.69	9.52	5.65	3.97	8.57	8.99
α_2	2.78	0.57	0.21	0.29	0.15	9.47e-5
σ_s [S/m]	0.86	0.71	0.47	0.43	0.99	0.80

In addition, this study highlights the limitations of using the slim form probe to detect renal positive surgical margins. In fact, the margin detection with the open-ended coaxial probe technique is feasible if a consistent and significant dielectric contrast is found between renal healthy and cancerous tissue, and if these tissues are included within the sensing volume of the probe.

To this extent, further rigorous dielectric studies are needed to accurately characterise renal cancerous tissues with the aim of using dielectric spectroscopy for positive margin detection during partial nephrectomies. Only with a significant and consistent dielectric contrast between renal healthy and cancerous tissue this method of detection could be useful. Based on such a contrast, the feasibility of other probes with larger sensing volume than the slim form probe, such as larger open-ended coaxial probes operating at lower frequencies (than microwave ones) or tetrapolar probes, can be evaluated for in depth surgical margin detection. However, due to the heterogeneity of renal samples and the impossibility of inserting the probe into the sample in *in vivo* procedures, techniques to compensate for the dielectric contribution of the tissues surrounding the cancerous tissue need to be developed for an accurate differentiation between healthy and cancerous tissue.

Lastly, by considering that a larger probe results in a less precise tumour margin detection, the sensing volume of the sensor needs to be large enough to permit tumour detection yet small enough to avoid unnecessarily large negative margins, which are likely to increase the morbidity.

IV. CONCLUSION

In this study, renal cancerous samples have been dielectrically characterised with the Keysight slim form probe with the aim of evaluating the feasibility of detecting positive surgical margins with the slim form probe.

The high variation in dielectric properties obtained across five human renal cancerous samples underscore the challenges in using the slim form probe to detect renal surgical margins. For future dielectric studies involving renal cancerous tissue, the authors suggest to section the excised renal sample in order to accurately discriminate the cancerous tissue from other tissue types and characterise the cancerous tissue with respect to the healthy tissue.

Lastly, in case a significant and consistent dielectric contrast is found between renal healthy and cancerous tissue in future dielectric studies, larger dielectric probes operating at lower frequencies (than microwave frequencies) still represent potential sensors for the detection of renal positive surgical margins.

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