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Non-invasive vesicoureteral reflux detection: Heating risk studies for a new device

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Abstract

OBJECTIVE—A novel non-invasive device is being developed to warm bladder urine and to measure kidney temperature to detect vesicoureteral reflux (VUR).

MATERIALS AND METHODS—Microwave antennas focus energy within the bladder. Phantom experiments measured the results. The heating protocol was optimized in an *in vivo* porcine model. The heating protocol safety performed once, twice and three times consecutively was investigated in three pigs followed by pathologic examinations.

RESULTS—Computer simulations showed a dual concentric conductor square slot antenna the best. Phantom studies revealed this antenna easily heated a bladder phantom without over heating intervening layers. *In vivo* a bladder heating protocol of 3 minutes with 30 W each to 2 adjacent antennas 45sec on 15sec off followed by 15 minutes of 15sec on and 45sec off was sufficient. When pigs were heated once, twice and three times with this heating protocol, pathologic examination of all tissues in the heated area showed no thermal changes. More intensive heating in the animal may have resulted in damage to muscle fibers in the anterior abdominal wall.

CONCLUSION—Selective warming of bladder urine was successfully demonstrated in phantom and animals. Localized heating for this novel VUR device requires low power levels and should be safe for humans.

Keywords

non-invasive vesicoureteral reflux detection; heating risk study

INTRODUCTION

Vesicoureteral reflux (VUR) is a known risk factor for kidney damage in children [1]. In spite of VUR's known association with kidney scarring, clinicians are reluctant to evaluate patients with current imaging because it requires an unpleasant urethral catheter and potentially harmful ionizing radiation. A new non-invasive vesicoureteral reflux detection device has been conceived to comfortably and preferentially warm bladder urine to 40–44°C with microwave antennas located on the skin surface. Kidney temperatures can then be measured before, during and after bladder warming with a microwave receive antenna placed on the skin surface and connected to an ultrasensitive microwave radiometer [2],

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When reflux occurs, warm urine from the bladder will return to the kidney and any temperature rise noted in the kidney will document VUR. The grade of the reflux should correlate to the temperature change detected by the radiometer within the kidney. This device would avoid both urethral catheterization and ionizing radiation, which would be important steps forward for VUR detection in children. The most important first step regarding this new approach is to determine the heating risk to patients. This manuscript describes an investigation of the proposed bladder heating, including applicator design, testing in phantom models, and measurements from in vivo animal experiments that assess the heating risks.

MATERIALS AND METHODS

Applicator Design

Blinded CT scans of 3–5 year olds were used during the device design phase to create a simplified model of the kidneys and bladder for EM simulations. 3D image segmentation software, Avizo® (Visualization Sciences Group), was used to reconstruct and evaluate the size and location of bladder and kidneys. Once this was done, electromagnetic (EM) computer simulations of bladder urine warming were carried out using HFSS software (Ansys Inc.) to develop dual concentric conductor (DCC) antenna arrays that could deposit microwave energy in the bladder urine (120–210 mL) and minimize energy absorption in the skin and intervening tissues. The DCC is a slot antenna with dual concentric square metal patches separated by a narrow slot that is center fed along each side for coherent wave propagation inside the slot. The DCC antenna used for hyperthermia treatment of superficial tissue disease extending 10–15 mm from tissue surface reported in [3, 4] was redesigned for deep heating inside the bladder.

Bladder Warming in Phantoms

Performance of the microwave heating antenna array fabricated on 1.524 mm thick printed circuit board material (R03850 Rogers Corp, Arizona, USA) was evaluated in phantom models with tissue equivalent electromagnetic properties. Fig 1 illustrates the layered phantom model that includes: i) a cubic frame covered with thin Mylar (0.125 mm) film and filled with saline (75 mL; 2% salt in deionized water by weight) to mimic a urinary bladder located 1.5 cm deep below skin, ii) a 5 mm thick fat tissue-equivalent layer, and iii) a 12×12×12 cm acrylic container with Mylar window on one side for coupling an external microwave antenna into the interior which is otherwise filled with muscle tissue-equivalent liquid phantom at the heating frequency of 915 MHz. According to analysis of the CT scans, a 7 mm thick layer of solid muscle tissue was placed adjacent to the Mylar window and in contact with the circulating liquid muscle media which was maintained at 36.5°C to mimic core temperature as shown in Fig. 1. The 5 mm thick layer of fat tissue-equivalent solid phantom was mounted on the outside of the Mylar window, adjacent to a 7 mm thick bag of deionized water which was circulated at 20°C during heating to simulate mild (room temperature) cooling of the skin. Recipes for urine and phantom tissues were optimized to match literature values based on measurements of electrical conductivity and dielectric constant made with a dielectric probe connected to a network analyzer (Model 85070D Agilent Corp, Santa Rosa CA) [5, 6]. Plastic fiberoptic temperature probes (Luxtron 3100, Lumasense Technologies, Santa Clara CA) were placed in each layer to accurately measure phantom heating.

Due to smaller urine volume (75 mL) in the experimental setup, a modified version of the single square Dual Concentric Conductor [7, 8] microwave antenna (5 cm wide) was used for phantom studies to deposit microwave power inside the 4.22 cm wide urine phantom at depth. Thus, power was applied to a single element DCC test antenna that was directed into

the multilayer waterbolus-fat-muscle load and into the urine filled bladder (75 ml). Temperatures were recorded with fiberoptic temperature sensors located at the: a) interface between waterbolus and fat, b) interface between fat and muscle layer, and c) at 1 cm increments along a thermal mapping catheter inside the “bladder” cube. Microwave power (30Watts) was delivered at 915 MHz to raise the urine temperature to about 4–6°C above baseline (36.5°C).

In-vivo Bladder Warming in Pigs

Following successful in vitro studies 15–17kg female pigs considered to be a good model for 3–5 year old children underwent anesthesia in an animal committee approved protocol to study bladder heating in vivo.

Tissue temperatures were monitored at many locations in the various pigs. Probes were placed subcutaneously in the fat layer 2–3 mm below skin under the antennas, in abdominal wall muscle 5–6 mm below the skin, and crisscrossing through the bladder and into posterior lateral tissues deep in the abdomen. Fig. 2a shows a typical placement of the interstitial thermal mapping catheters with fiberoptic temperature probes. In this case, catheter P1 was inserted laterally through the bladder into beyond about 4–5 cm deep at the tip; probe P2 was stationary at the tip of the Foley inserted inside the bladder; probe P3 was inserted from the perineum superiorly ending inside the bladder about 2–3 cm deep below skin; and probe P4 was inserted superficially in fat layer about 2–3 mm deep under the 2 DCC antenna array. Interstitial tissue temperatures were measured by pulling the probes in 1 cm incremental steps across the insertion length of the catheters with 5 s dwell time at each position using a computer controlled mapping device. A schematic representation of the catheter placement relative to the bladder is shown in Fig. 2b–c. Ultrasound measurements confirmed the depth and position of the implanted catheters inside the pig bladder which was first drained and then filled with 180 mL of urine phantom pre-warmed to 36.5°C prior to turning microwave power on.

Due to the larger bladder volume (180 mL), bladder warming trials in pigs were carried out using an 2×1 DCC antenna array optimized for selective deep heating [8].

Temperatures were recorded at 13 positions along each catheter to document temperatures inside, above, below and beside the bladder during heating. The heating antenna was coupled to the skin through a 7–10mm thick deionized water bolus slowly circulating room temperature water (20°C) during bladder warming. Bladder warming experiments were performed using 915 MHz microwave source and power amplifiers (American Microwave Technologies Inc, Anaheim CA) delivering 30Watts to each DCC antenna with varying on/off cycles.

Heating experiments were evaluated on one preliminary pig (protocol determination animal) to establish the optimal bladder warming protocol that produced appropriate heating of the stagnant urine while allowing the perfused skin and intervening fat/muscle tissues to cool during the time intervals with microwave power off. The heating protocol determined on the preliminary study was then tested repeatedly on three additional pigs that were examined for post-treatment pathologic changes within the bladder and abdomen. The first pig had the warming protocol performed for 20 minutes at intra-bladder temperatures of 40–44°C. The second pig had the warming protocol for 20 minutes at temperature followed by an equalization period of 20 minutes and then a second identical 20 minute warming protocol. The third pig had the warming protocol repeated three times with the same 20 minute rest period between warming cycles. At the end of the warming studies, the animals were euthanized; the lower abdominal wall, urinary bladder, internal genitalia, and rectum were excised en bloc and placed in 10% neutral buffered formalin; and sections from the

abdominal wall, urinary bladder, and uterus and vagina were processed into paraffin and stained with hematoxylin and eosin for histologic examination.

RESULTS

Applicator Design

CT scans of children aged 3–5 years revealed a 1cm average abdominal wall thickness to the anterior surface of the bladder, and a 3cm average distance to the center of the bladder. The average depth of the kidney was measured as 3.5cm to the renal pelvis from the skin. EM computer simulations of the 915 MHz DCC antenna indicated that multiple warming antennas minimized skin and intervening tissue warming and maximized the warming of urine in the bladder [7, 8]. A DCC antenna array with two 3×3 cm square elements operating incoherently at 915 MHz was found to be the best for bladder warming through a thin layer (7–10mm) of deionized waterbolus at room temperature [9].

Bladder Warming in Phantoms

Phantom experiments using a single DCC antenna clearly demonstrate the ability to heat the 75 mL saline filled bladder model surrounded by temperature controlled muscle phantom (36.5 °C) beneath a 5 mm fat layer and through 7 mm room temperature waterbolus at 20°C. Fig. 3 shows the temperatures measured inside the layered bladder model with 75 mL urine that was externally warmed by a square single element DCC antenna. The temperature rose about 4°C at the bolus-fat layer interface and less than 1.5°C in the muscle overlying the bladder urine. Even with continuous power, temperature of the surrounding liquid muscle phantom remained within 2.5°C of the initial muscle temperature (36.5°C) for more than 20 minutes of heating, and the temperature rose less than 1°C at the antenna-tissue interface.

In-vivo Bladder Warming in Pigs

A heating protocol was determined with optimum time sequencing to warm the pig bladder with interspersed power off time periods for cooling of overlying perfused tissues and passive (radiometric) temperature monitoring. An appropriate protocol was experimentally determined to be 30 Watts to both antennas with 75% duty cycle (45s on, 15s off) for the first 3 minutes (22.5 Watts average) changing to 25% duty cycle (7.5 Watts average) when bladder reached the goal maximum temperature of 44°C. This is because more energy is required during the initial warm up phase and less energy is required for maintenance/radiometric monitoring phase typically 15–20 minutes. Small adjustments of the power level (± 5 W) were applied during the maintenance phase to ensure a stable bladder temperature, leaving the fixed duty cycle unchanged. The bladder warming algorithm was tested on three other pigs under anesthesia for the heating cycles mentioned before. Fig. 4 shows the tissue temperatures recorded by the interstitial temperature sensors for the second pathology pig that was exposed to two bladder warming cycles with 20 minutes break. Note the temperatures ranged from 40–44°C inside the two bladder probes for the 20 minutes heat cycles. Pathologic examination of the heating protocol determination pig showed focal marked vascular congestion and hemorrhage in the sub-epithelial tissues of the bladder (Fig 5A). Of the remaining 3 pigs only local trauma likely from the temperature probe placement (Fig 5B) was seen and no other thermal changes were found (Fig 5C–D).

Large areas of hemorrhage were observed over the anterior aspect of the bladder in the protocol determination animal, but no abnormalities were observed on gross examination of the resectioned tissue in the three study animals. Histologic examination of the urinary bladder revealed marked congestion, edema, and acute subepithelial hemorrhage in the protocol determination animal. Mild patchy edema and a small focus of epithelial erosion with focal acute hemorrhage was seen in one of the three study animals probably from

temperature probe placement, but no histologic abnormality was encountered in the other two. Histologic examination of the anterior abdominal wall musculature in the protocol determination animal revealed swollen and degenerating fibers with marked interstitial edema in the protocol determination animal, but no skeletal muscle injury was seen in any of the three study animals (Fig. 6). The lesions observed in the urinary bladders of the protocol determination animal and one of the three test animals were most likely due to local trauma, though a contribution from thermal injury cannot be completely excluded. The lesion observed in the anterior abdominal wall skeletal muscle in the protocol determination animal but not any of the test animals, may be due to thermal injury, but the myofiber damage would likely be reversible.

DISCUSSION

Hyperthermia has been used for many years, primarily as an adjuvant to radiation and/or chemotherapy to improve cancer outcomes. There is extensive clinical experience with microwave heating of superficial tissues extending about 3 cm from the skin surface. Typical hyperthermia treatments consist of raising the target tissue temperature from a baseline of 35–37°C to 40–45°C within about 5–10 minutes of power on (i.e. about 0.5–1°C/min heating rate) and maintained within this range for approximately 1 hour [10–13]. It should be noted that there are numerous reversible changes in biological tissue in this range of thermal dose which involves temperatures of 40–45°C for treatment times of about 60 minutes [14]. The new device for non-invasive vesicoureteral reflux detection aims to use this same microwave heating technology at a lower thermal dose level of 40–44°C for 20 minutes, producing comfortable bladder warming. It is assumed that the bladder contents will rapidly homogenize in temperature due to natural convection within the bladder, which will minimize temperature gradients arising during external bladder warming and provide stable warm urine ready for the reflux event.

Phantom heating experiments with single DCC microwave antenna at 915 MHz demonstrated the feasibility of localized heating of 40–44°C in a 75 mL urinary bladder model with low temperatures in overlying fat and muscle ($\leq 38^\circ\text{C}$), as shown in Fig 3. This is indicative of low patient discomfort and minimal complications. The phantom studies provided sufficient data to move on to confirmation of these results in *in vivo* porcine bladder experiments. A 2x1 DCC antenna array was used for the pig bladder warming experiments due to the larger bladder volume (180 mL urine surrogate) typical for older patients (3–5 year old). A pulsed power heating protocol with 30 Watts alternately to each antenna and 75% duty cycle for the first 3–4 minutes followed by 25% duty cycle for the next 20 minutes was determined based on heating cycles delivered to several protocol determination pigs. The high electrical conductivity of urine contained inside the bladder leads to preferential power absorption when irradiated using a microwave hyperthermia applicator [5, 15]. At our advantage, the lack of blood perfusion and the presence of a thermal barrier inside the bladder result in heat retention by the urine. Thus, the urine cools significantly slower when power is turned off for short durations than the surrounding perfused muscle and subcutaneous tissues. In contrast, temperature in the surrounding muscle and skin loses temperature quickly when microwave power is turned off primarily due to cooling from blood perfusion in these normal tissues [16] as well as cooling from the room temperature waterbolus on the skin [17]. Thus, the differential heating of urine was enhanced by the power modulation scheme, by allowing surface tissues to cool intermittently during periods of power off while the bladder contents continued to rise due to heating alternately from one antenna or the other which both couple the same stagnant urine reservoir.

The predetermined heating protocol was administered to three pigs destined for pathologic examination and the bladder (urine) temperature was raised to 40–44°C within the first 3–4 minutes with low skin/muscle tissues as seen in Fig 4. The time delay between second and third repeated heat exposures was determined for these three pigs such that the urine and abdominal tissues cooled down to their respective base temperatures that were recorded prior to the first heat cycle. Each 20 minutes heat cycle used almost similar power levels with few Watts variation for the 6 heat exposures in 3 pigs and slight variations in temperature probe placement, and initial core temperatures. Pathology results of the three pigs demonstrated safe heating of the bladder, urine and upper pelvis for intra-bladder temperatures of 40–44°C without significant tissue damage.

We were pleased to find that the power necessary to warm even large (180 mL) bladder to 40–44°C could be delivered from the simple low profile printed circuit board type DCC antennas with a modulation (75% on/off cycle followed by 25% on/off cycle) that provided sparing of surface tissues. By minimizing the time averaged power per antenna, the skin and overlying structures were spared any thermal damage as evidenced in the pathology examination of the skin, subcutaneous fatty tissues, abdominal musculature, anterior and posterior bladder wall, peritoneum, ovaries, and vagina.

CONCLUSIONS

In-vivo porcine experiments established an appropriate heating protocol to gently warm urine inside the bladder as a first step in radiometric detection of VUR. A safe and effective heating mode was established using 25% on/off cycle modulated power alternately to two adjacent antennas coupled to the bladder urine through overlying tissue regions. Microwave warming of urine within the bladder to 40–44°C was accomplished safely in three pigs with no histopathology lesion associated with heating the bladder on this protocol even after three consecutive 20 minute warming cycles. No potential risks associated with heating bladder as a stimulus for non-invasive radiometric diagnosis of VUR were evidenced. From previous experience in hyperthermia therapy, we anticipate this low power bladder warming will be comfortable for the patient. Seldom in the past have bladders been warmed for diagnostic purpose, and to our knowledge this is the first non-invasive urinary bladder warming study. This work demonstrates that this new device is safe for human testing.

References

1. Snow, BW.; Stauffer, PR.; Arunachalam, K.; Maccarini, PF.; De Luca, V.; Klemetsen, Ø., et al. Non-invasive vesicoureteral reflux detection: A New Device. Presented at the American Academy of Pediatrics National Conference and Exhibition; San Francisco, CA. 2010.
2. Arunachalam K, Maccarini PF, De Luca V, Tognolatti P, Bardati F, Snow BW, et al. Detection of vesicoureteral reflux using microwave radiometry: System characterization with tissue phantoms. *IEEE Trans Biomed Eng.* 2011 Jan 20. [Epub ahead of print].
3. Rosetto F, Diederich CJ, Stauffer PR. Thermal and SAR characterization of multielement dual concentric conductor microwave applicators for hyperthermia, a theoretical investigation. *Med Phys.* 2000; 27(4):745–753. [PubMed: 10798697]
4. Stauffer PR, Rosetto F, Leoncini M, Gentili GB. Radiation patterns of dual concentric conductor microstrip antennas for superficial hyperthermia. *IEEE Trans Biomed Eng.* 1998; 45(5):605–613. [PubMed: 9581059]
5. Arunachalam K, Maccarini PF, De Luca V, Bardati F, Snow BW, Stauffer PR. Modeling of the detectability of vesicoureteral reflux in children using microwave radiometry. *Phys Med Biol.* 2010; 55(18):5417–5435. [PubMed: 20736499]
6. Gabriel S, Lau RW, Gabriel C. The dielectric properties of biological tissues: III Parametric models for the dielectric spectrum of tissues. *Phys Med Biol.* 1996; 41(11):2271–2293. [PubMed: 8938026]

7. Maccarini, PF.; Arunachalam, K.; Juang, T.; De Luca, V.; Rangarao, S.; Neumann, D., et al. Shaping and resizing of multifed slot radiators used in conformal microwave antenna arrays for hyperthermia treatment of large superficial diseases. *IEEE Proc of Int Conf Electromagnetism in Advanced Applications*; Turin, Italy: IEEE Press; 2009. p. 746-749.
8. Maccarini, PF.; Paulides, M.; Stauffer, PR. The effect of excitation phase on SAR distribution and penetration depth of square slot and parallel slot antennas for superficial hyperthermia. Presented at the Society Thermal Medicine 2010 Annual Meeting; Clearwater, Florida. 2010.
9. Stauffer, P.; Maccarini, P.; Arunachalam, K.; Salah, S.; De Luca, V.; Boico, A.; Klemetsen, O.; Birkelund, Y.; Jacobsen, S.; Bardati, F.; Tognolatti, P.; Snow, B. Microwave Radiometry for Non-Invasive Detection of Vesicoureteral Reflux (VUR) Following Bladder Warming. Ryan, T., editor. Bellingham WA: Proc of SPIE: SPIE Press; 2011. p. 7901OV-1-11
10. Stauffer PR. Evolving technology for thermal therapy of cancer. *Int J Hyperthermia*. 2005; 21:731–744. [PubMed: 16338856]
11. Stauffer, PR.; Diederich, CJ.; Pouliot, J. Thermal Therapy for Cancer. In: Thomadsen, B.; Rivard, M.; Butler, W., editors. *Brachytherapy Physics*. 2nd Ed.. joint AAPM/ABS Summer School; 2005. p. 901-932.
12. Viglianti, BL.; Dewhirst, MW.; Stauffer, PR.; Repasky, E.; Jones, E.; Vujaskovic, Z. Hyperthermia. In: Kufe, E.; Bast, R.; Hait, WN., editors. *Cancer Medicine*. Hamilton: B.C. Decker; 2010.
13. Sneed, PK.; Stauffer, PR.; Li, G.; Sun, X.; Myerson, R. Hyperthermia. In: Phillips, TL.; Hoppe, R.; Roach, M., editors. *Textbook of Radiation Oncology*. 3rd ed.. Elsevier Saunders Co.; 2010. Hyperthermia
14. Dewhirst MW, Vujaskovic Z, Jones E, Thrall D. Re-setting the biologic rationale for thermal therapy. *Int J Hyperthermia*. 2005; 21:779–790. [PubMed: 16338861]
15. Despretz D, Camart JC, Michel C, Fabre JJ, Prevost B, Sozanski JP. Microwave prostatic hyperthermia: Combination-theoretical study interest of urethral and rectal applicators combination. Theoretical and animal experimental results. *IEEE Trans Microw Theory Tech*. 1996; 44:1762–1768.
16. Van de Kamer JB, Van Wieringen N, De Leeuw AA, Lagendijk JJ. The significance of accurate dielectric tissue data for hyperthermia treatment planning. *Int J Hyperthermia*. 2001; 17:123–142. [PubMed: 11252357]
17. Arunachalam K, Maccarini PF, Craciunescu IO, Schlorff JL, Stauffer PR. Thermal characteristics of thermobrachytherapy surface applicators (TBSA) for treating chestwall recurrence. *Phys Med Biol*. 2010; 55(7):1949–1969. [PubMed: 20224154]

Fig. 1.
Top view of experimental setup to investigate bladder heating at depth in layered bladder tissue model (the illustration is not drawn to scale).

Fig. 2.

(a) Placement of interstitial catheters for thermal mapping of fiberoptic temperature probes during bladder warming in pig 1; schematic illustration of (b) anterior and (c) lateral catheter placement for thermal mapping.

Fig. 3. Phantom temperatures measured during heating experiment of the 75mL bladder surrounded by temperature controlled muscle phantom at 36.5°C. Antenna located 1.5 cm from the 0° bladder front surface. The temperature at the bolus phantom interface remained below 25°C during the experiment.

Fig. 4.

Tissue temperatures acquired during bladder heating for the second pig exposed to two 20 minutes warming cycles. Temperature data is simplified to display peak temperature in urine measured by the two probes (P1, P3) crossing inside the bladder, compared to temperatures measured in nearby urethra inside the Foley catheter which remained close to core temperature.

Fig. 5. Histological examination of the urinary bladder revealed marked congestion, edema, and acute sub-epithelial hemorrhage in the protocol determination animal (A), but only focal edema and a small focus of epithelial erosion with focal acute hemorrhage (very likely due to the local trauma of temperature probe placement) in one of the three study animal (B) but no abnormality in the other two (C and D). All sections stained with hematoxylin and eosin and photographed at an original magnification of 100X.

Fig. 6. Histological examination of the anterior abdominal wall musculature in the protocol determination animal revealed swollen and degenerating fibers with marked interstitial edema in the protocol determination animal (A), but no skeletal muscle injury in any of the three study animals. All sections stained with hematoxylin and eosin and photographed at an original magnification of 200 $\times$.