MECHANISMS AND APPLICATIONS OF BOILING HISTOTRIPSY FOR MECHANICAL TISSUE ABLATION IN HIFU

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OUTLINE

- High intensity focused ultrasound (HIFU): main principles and current challenges
- Histotripsy – HIFU-based mechanical ablation
  - Cavitation and boiling histotripsy: physical mechanisms
  - Advantages over thermal ablation
  - Instrumentation
- Clinical applications and preclinical studies of boiling histotripsy
  - Ablation of solid cancers: liver, kidney, pancreas, prostate
  - Evidence for enhanced anti-tumor immune response
  - Release of blood based biomarkers
  - Liquefaction of large hematomas for fine needle aspiration
HIGH INTENSITY FOCUSED ULTRASOUND (HIFU) THERAPY

MRI and MR thermometry
Based on proton resonance frequency shift

FDA-approved:
- uterine fibroids
- bone tumors
- essential tremor

CE mark in Europe:
- breast tumors
- kidney and liver tumors
- pancreas tumors

Challenges: costly, lengthy procedure
ULTRASOUND-GUIDED HIFU ABLATION

FEP-BY-02 (Yuande Biomedical Engineering Limited Corporation)

Other devices:
- Haifu (China)
- Alpinion (Korea)
- Theraclion (France)
- Ablatherm (France)
- Sonablate (USA)

Challenges: limited feedback on completeness of ablation

CHALLENGES IN HIFU THERMAL ABLATION

- Near-field heating of the intervening tissues: ribs, skin, muscle, fat
- Heat sink effect in well vascularized targets: incompleteness of ablation (ultrasound-guided)
- Cost, lengthy procedure (MR-guided)
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HIFU FOR NON-INVASIVE TISSUE EROSION – “HISTOTRIPSY”

Mechanical damage to tissue without significant thermal effects
University of Michigan, since 2004

- Fluid-filled void in tissue
- Cavitation bubble cloud
- Ultrasound guidance

Maxwell et al., Acoustics today 2012
**ALTERNATIVE HISTOTRIPSY REGIME: BOILING HISTOTRIPSY**

*Based on nonlinear propagation effects in HIFU*

University of Washington, since 2009

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### Linear Propagation

- $P^+ = P^-$
- Heating: $T \sim P^2$

### Nonlinear Propagation

- $P^+ >> P^-$
- Heating: $T \sim (A_S)^3$

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**Shock wave heating**

- 100°C can be reached in several milliseconds!
- Vapor bubble!

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**Examples in ex vivo bovine liver**

- 5 mm
- 1 MHz
- 2 MHz
- 100 um

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**Boiling Histotripsy**

- 60-100 MPa
- 10-16 MPa
- 1-20 ms HIFU pulse
- 0.1-1 s pause
- boiling

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**ALTERNATIVE HISTOTRIPSY REGIME: BOILING HISTOTRIPSY**

University of Washington, since 2009
In vivo porcine kidney

Hyperechoic region corresponds to vapor bubbles

Post-treatment

Differential Ablation Threshold and Connective Tissue Structure sparing

Volumetric BH lesions in ex-vivo bovine liver (produced by Phillips Sonalleve)
With contents, Contents removed

Treatment time: 30 min
Lesion volume: 6 cc

Histology: Masson’s trichrome stain

All larger caliber vessels (~1 mm) and some of smaller (~100 μm) vessels preserved

Volumetric BH lesions in-vivo porcine liver: lobules and ducts preserved
TEM IMAGING OF BOILING HISTOTRIPSY LESIONS

Lesion contents

Transition zone from fully intact to completely destroyed tissue ~20 μm

Lesion border

MECHANISM OF TISSUE FRACTIONATION BY BOILING HISTOTRIPSY

How does a mm-sized bubble fractionate tissue down to subcellular level?

1. Shock-wave super-focusing and heating within ms to 100°C
2. Interaction of shocks with vapor cavity: acoustic micro-fountain and sub-surface cavitation

Why is there no measurable thermal effect?

Super-heated area is much smaller (~100 um) compared to the vapor bubble (~ 1 mm) and the resulting cavity (~2-5 mm)

EX-VIVO TISSUE ATOMIZATION BY HIFU

Focusing HIFU on the free tissue-air interface


ADVANTAGES OVER THERMAL HIFU

- Less potential for damaging intervening tissues (ribs, skin, muscle)
- Ablation not affected by heat-sink effect
- Real-time B-mode ultrasound imaging of the bubbles (hyperecho) and outcome (hypoecho)
- Sparing of connective tissue structures (ducts, vessels, fascial planes), tissue selectivity
- Tissue lysate resorbed without fibrosis
- Tumor lysate may provide benefits in terms of enhanced antitumor immune response
BOILING HISTOTRIPSY INSTRUMENTATION

- Pulse duration: 1-20 milliseconds
- Duty factor: 0.5-2%
- Transducer F#: 0.75 (90° sector) – 1.4 (40° sector)
- Frequency: 1-3 MHz
- Peak input power: 300 Watts – 4000 Watts

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In vivo histotripsy of pig liver and kidney

**Goals:**
- Evaluate feasibility of transcutaneous and transcostal ablation
- Evaluate boiling histotripsy "dose" – number of pulses per spot – for different tissues

Treatment feasible in 11 targeted kidneys
- Power threshold 600-1000W (2-fold vs ex-vivo)
  - 50% rib coverage feasible (additional 2-fold increase)
- No hematuria, no prefocal heating/bruising
- Differential sensitivities: cortex > medulla > collecting system > capsule
- Precise targeting despite no motion tracking
- Ablation rate up to 27 cc/hour

Results: kidney
Differential sensitivities of renal tissues

- Cortex
- Medulla
- Collecting System
- Capsule

Human renal tumor tissue more sensitive than benign renal tissues (separate ex vivo study)

Results: liver

- Lesion shape more irregular due to respiratory motion and lobular structure
- Lesion visualization more challenging
- At higher output powers, collateral damage to fat layer

Challenge: aberration and defocusing by soft tissues, primarily fat
Aberration correction strategies needed
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HISTOTRIPSY-INDUCED IMMUNE RESPONSE

Hypothesis: mechanical effects of HIFU cause the response, which is CD8+ T-cell mediated

Histotripsy alone is unlikely to trigger a clinically significant anti-tumor immune response


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ANIMAL MODEL: EKER RAT

Naturally occurring syndromic RCC model\(^1\)

*\(Tsc2\)* tumor suppressor gene

Autosomal dominant: 100\% penetrance by 1 yr

Similar biology to human clear cell\(^2,3\)

- Upregulated HIF-2\(^\alpha\)
- Overexpression of VEGF
- Increased MTOR signaling

Bilateral tumors, rare metastases

25-50\% of the tumor liquefied by boiling histotripsy

Rats survived for 1, 2, 7, 14 and 56 days


**EVOLUTION OF THE TREATED AREA**

B-mode ultrasound

<table>
<thead>
<tr>
<th>Eker rat</th>
<th>Wild-type rat</th>
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<tbody>
<tr>
<td>Day 7</td>
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<td>Day 14</td>
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<td>Day 56</td>
<td>Day 56</td>
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Hypoechoic cavity peaks on day 7

- disappears by day 14

- correlates with contraction grossly

Kidney appears healed by day 56
POST-TREATMENT INFLAMMATION

Significant inflammation at 48 hrs

Immunohistochemistry: CD8+ T-cells (stained brown) increased in treated tumor and contralateral kidney, but not in sham-treated control

Tumor Infiltrating Leukocytes (48 h)

Immunohistochemistry: CD8+ T-cells (stained brown) increased in treated tumor and contralateral kidney, but not in sham-treated control

T=tumor, BH=liquefied lesion, C=benign kidney cortex
Plasma Cytokine Profile (up to 48 hrs)

**Pro-inflammatory**

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<th>Pre</th>
<th>0.25</th>
<th>1</th>
<th>4</th>
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<tr>
<td><strong>Plasma TNF-α</strong></td>
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**Anti-inflammatory**

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**Proliferation marker Ki67 (48 h)**

Boiling histotripsy ablation of the renal tumor does not appear to stimulate tumor proliferation

**Treated kidney**

**Distant tumor**

**Sham control**

T=tumor, BH=liquefied lesion, C=benign kidney cortex
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BOILING HISTOTRIPSY AS NON-INVASIVE LIQUID BIOPSY TOOL

Blood-based cancer biomarker – a cancer-specific molecule secreted by the tumor into the patient’s circulation

Problem: concentration of nucleic biomarkers (miRNA, mRNA, DNA) with high predictive value is often low, at the limit of detection

Syngeneic rat prostate cancer model (MatLyLu)

Boiling histotripsy

Chevillet, Khokhlova et al. Radiology 2016
BH-STIMULATED TUMOR miRNA RELEASE

**Tumor-associated miRNAs**

**Broadly expressed control**

**Outcome**

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Motivation

**Hematoma** – a collection of blood outside of blood vessels

**Causes:** sharp and blunt trauma, surgical procedures, muscle sprain and overuse in sports

**Health effects of large hematomas:** PAIN, risk of infection, risk of compartment syndrome

**Clinical management:** Rest, Immobilization, Compression, Elevation (RICE)

Indwelling catheters (ineffective)

Surgical evacuation (e.g. fasciotomy)

**Goal:** use histotripsy methods to rapidly (15-20 minutes) liquefy large (~20-50 cc’s) hematoma volumes for fine needle aspiration

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**BH liquefaction of hematomas in vitro**

**Fine needle aspiration**

Liquefaction rate up to 1.3 cc/minute
Boiling histotripsy is a non-invasive, non-thermal HIFU-based ablation method
- Can be implemented with existing clinical HIFU systems
- Precise treatment, sharp lesion borders
- Real-time and post treatment ultrasound guidance
- Differential threshold for damage depending on tissue type
- Liquefied tissue reabsorbs quickly without fibrosis
- Stimulates anti-tumor immune response
ACKNOWLEDGEMENTS