In-vitro investigations on fluorescence behavior of urinary and kidney calculi

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Laser lithotripsy – Investigations

**Propulsive movement** during treatment leads to stone chasing

Dependency of **Fragmentation** process on laser parameter

**Remaining fragments** can hardly be distinguished from surrounding tissue
Propulsion Measurement – Materials & Methods (1)

Laser system: AURIGA QI
StarMedTec
Ho:YAG (2.1 µm)

Stone phantom:
- BEGO powder
- mixture: 15:4
- edge length: 5 mm
- mass: (0.2243 ± 0.0061) g

Application fibre: 365µm bare fibre

Containment: Acrylic glass tube

Camera System: Sony RX100V
- 1000 fps
Detection of stone phantom via *colour threshold* (red value)

1

$1^{st}$ derivative of rising flanks displays **mean velocity** of stone  $v = \frac{dx}{dt}$

Recording of around 70 rising flanks per video (at repetition rate of e.g. 10 Hz)

→ **Mean of 70 velocity values per video**
Comparison of propulsive movement triggered by two laser settings on two consecutive days.

Laser parameter:
- Power: 10 W
- Energy per Pulse: 1 J/Pulse
- Repetition rate: 10 Hz
- Pulse duration: 0.3 and 1.0 ms

n = 5 per setting

- Standard error (±7.5%)
- Variation in pulse duration leads to significant different velocity values
- Day to Day variability does not differ significantly → Highly reproducible
# Propulsion Measurement – Discussion

## Pendulum

<table>
<thead>
<tr>
<th>Weakness</th>
<th>Strength</th>
<th>Weakness</th>
</tr>
</thead>
<tbody>
<tr>
<td>Only first laser pulse</td>
<td>Multiple pulses</td>
<td>Cubic stone phantom</td>
</tr>
<tr>
<td>Hardly reproducible</td>
<td>High sample size possible</td>
<td>Neglecting fluid dynamics</td>
</tr>
<tr>
<td>High Standard deviation</td>
<td>Hands-free measurement</td>
<td>Acceleration process not resolvable</td>
</tr>
</tbody>
</table>

Reproducible measurement conditions

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## Video Tracking

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Fragmentation in ureter model

Laser parameters:
- Laser system: AURIGA QI Ho:YAG
- repetition rate: 10Hz
- energy per pulse: 0.5J/pulse and 1.0J/pulse
- pulse duration: 0.25ms - 1.6ms

Stone phantom:
- BEGO powder mixture: 15:4
- edge length: 5 mm
- mass: (0.2243 ± 0.0061) g

Application fibre:
- 365µm bare fibre

Vessel:
- Acrylic Glass tube;
- Mesh size 2.3 mm

<table>
<thead>
<tr>
<th>fragments</th>
<th>dust</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.3 mm - 1 mm</td>
<td>&lt; 1 mm</td>
</tr>
</tbody>
</table>
Fragmentation in ureter model

**Experimental procedure:**

- weighing of the stone (Satorius, RC210P)
- fragmenting (≤ 2.3 mm)
  - Total application time \( t_{\text{tot}} \)
  - Time until stone breaks \( t_{\text{until break}} \)
  - Time after break \( t_{\text{after break}} \)
- sieving (mesh grid: 1 mm)
- drying (ca. 24h)
- weighing of the fragments

Mass of dust: \( m_{\text{dust}} = m_{\text{stone}} - m_{\text{fragments}} \)

Dusting part: \( D = \frac{m_{\text{dust}}}{m_{\text{stone}}} \)

Fragment part: \( F = \frac{m_{\text{fragments}}}{m_{\text{stone}}} \)
Fragmentation - Results

Variation in pulse duration or energy per pulse leads to different fragmentation times and dusting ratios.

<table>
<thead>
<tr>
<th>Time [s]</th>
<th>Duration</th>
<th>D</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.25ms</td>
<td>1.6ms</td>
<td>60.8%</td>
<td>39.2%</td>
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<tr>
<td>1.0ms</td>
<td>1.6ms</td>
<td>69.3%</td>
<td>30.7%</td>
</tr>
<tr>
<td>burst 1.6ms</td>
<td>1.6ms</td>
<td>67.5%</td>
<td>32.5%</td>
</tr>
<tr>
<td>1.2ms</td>
<td>1.6ms</td>
<td>72.9%</td>
<td>27.1%</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Time [s]</th>
<th>Duration</th>
<th>D</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.84ms</td>
<td>1.0J 10Hz</td>
<td>71.7%</td>
<td>28.3%</td>
</tr>
<tr>
<td>0.25ms</td>
<td>1.0J 10Hz</td>
<td>61.2%</td>
<td>38.3%</td>
</tr>
<tr>
<td>1.3ms</td>
<td>1.0J 10Hz</td>
<td>67.2%</td>
<td>32.8%</td>
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<td>burst 1.6ms</td>
<td>1.0J 10Hz</td>
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<td>33.1%</td>
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<table>
<thead>
<tr>
<th>Time [s]</th>
<th>Duration</th>
<th>D</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>206.4s</td>
<td>t_mean: 318.0 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 206.4 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 257.0 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 240.8 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 361.4 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 785.0 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 447.6 s</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>206.4s</td>
<td>t_mean: 616.0 s</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
Fluorescence Microscopy – Materials & Methods

- **Microscope:** Leica DM IRBE Lens 10x – 20x
- **Light source:** Osram HBO 103 Mercury Arc Lamp
- **Filter:**
  - Excitation Bandpass:
    - 400nm, 450nm und 550nm
  - Emission Longpass:
    - 470nm, 520nm und 590nm
- **Camera:** Leica DFC 300 FX
  - 1.4 MPixel Resolution
- **Spectrometer:** Ocean Optics USB 2000+
  - via 1mm multimode wave guide
Fluorescence Microscopy

$\lambda_{\text{ext}} = 400 \text{ nm}$
$\lambda_{\text{det}} > 470 \text{ nm}$

$\lambda_{\text{ext}} = 450 \text{ nm}$
$\lambda_{\text{det}} > 520 \text{ nm}$

$\lambda_{\text{ext}} = 550 \text{ nm}$
$\lambda_{\text{det}} > 590 \text{ nm}$
Fluorescence Endoscopy – Materials & Methods

- **Light-source:** D-Light, STORZ, 20133220
- **Endoscope:** STORZ, 0°, HOPKINS II
- **Long pass filter:** $\lambda_{det} < 610$ nm
- **Fiber:** 1000 $\mu$m, 0.39 NA
- **Spectrometer:** Ocean Optics, USB 2000+
- **Camera:** STORZ, telecam PDD 20212037
Fluorescence Endoscopy – In vivo images

High Intensity

Low Intensity

Fluorescence Spectra

- $\lambda_{\text{exc}} = 500 \text{ nm} - 570 \text{ nm}$
- $\lambda_{\text{det}} < 610 \text{ nm}$
Summary

<table>
<thead>
<tr>
<th>Propulsion</th>
<th>Fragmentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Method</td>
<td>developed</td>
</tr>
<tr>
<td>Evaluation</td>
<td>Mean velocity</td>
</tr>
<tr>
<td>Handling</td>
<td>No influence of experimentator</td>
</tr>
<tr>
<td>Reproducibility</td>
<td>proven</td>
</tr>
</tbody>
</table>

Two setups for investigations on dependency of laser lithotripsy on laser parameters

→ Deriving of optimal laser parameters

Fluorescence

- stones show fluorescence
- fluorescence spectra depend on $\lambda_{\text{exc}}$

→ Distinguish stones from tissue looks promising
→ Potential use for in-situ stone composition determination
Thank you for your attention.

Kiitos huomiostanne.

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