Monitoring Pancreatic α-Amylase of Postoperative Patients with Droplet-Based Microfluidics

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Introduction

• Postoperative complications after pancreatic surgery are frequent and can be life-threatening. Current clinical strategies quantify α-amylase activity intermittently, resulting in delayed treatment.
• The portable droplet-based microfluidic device is capable of real-time monitoring α-amylase activity.
• This strategy significantly improves the determination time (3 min) and detection limit (7 nmol/s·L) and reduces sample and reagent requirement (10 µL).

Methods

<table>
<thead>
<tr>
<th>Gold Standard in Clinic</th>
<th>Microplate</th>
<th>Microfluidics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Detection method</td>
<td>Colorimetric</td>
<td>Fluorometric</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>&gt; 6 U/L</td>
<td>&gt; 0.72 U/L</td>
</tr>
<tr>
<td>Range</td>
<td>4.8 U/L - 1200 U/L</td>
<td>2 U/L - 50 U/L</td>
</tr>
<tr>
<td>Detection time</td>
<td>1 h</td>
<td>20 min</td>
</tr>
<tr>
<td>Reagent requirement</td>
<td>1 kit for 1 assay (1-5 mL)</td>
<td>1 kit for 1 assay (1-5 mL)</td>
</tr>
<tr>
<td>Sample requirement</td>
<td>1-5 mL (96 replicates, one 96-well plate)</td>
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</tbody>
</table>

Reagent requirement

• Pancreatic drain liquid samples are collected and injected to the microfluidic system by switching valves.
• Reagent, amylase, and buffer are mixed as an aqueous phase through T-junctions.
• Droplets formed at cross-junction after meeting HFE oil and mineral oil (spacer).
• Amylase reacts with the reagent, resulting in cleavage products emitting fluorescence.

Detection Principle

• Different amylase concentrations result in different fluorescence intensity.
• Calibration curve determined at 3 min with LOD of 7 nmol/s·L.
• Results of the clinical and microfluidic methods have a great linear correlation in a total of 32 patient samples.

Detection

Continuous monitoring

Conclusion

• Droplet-based real-time detection of amylase offers improvement in LOD, detection time, and reagent requirements.
• All 32 samples results matched well with clinical measurements.
• Rapid response of fluorescence intensity to sample concentration fluctuations indicates the method can be implemented to continuously monitor drain α-amylase activity of patients.

Outlook

• We envision the potential utility of our technique in other clinical scenarios, e.g., detecting "anastomotic leakage in colorectal surgery" or "bile leaks in liver surgery" where our droplet-based analysis technique could be explored.

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Calibration Curve & Patient Sample Test

Continuous monitoring

PMT fluor. intensity [a.u.] 0 0.2 0.4 0.6 0.8 1.0
Sample ratio 0 0.1 0.2 0.3 0.4 0.5

PMT fluor. intensity [a.u.] 0 0.2 0.4 0.6 0.8 1.0
Sample ratio 0 100 nmol/s·L 200 nmol/s·L 300 nmol/s·L

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