Novel urinary peptide markers for detection of colorectal cancer – results of a pilot study

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Introduction

- Colorectal cancer (CRC) is common and survival is dependent on stage at diagnosis.
- The involvement of proteases in growth and progression of colorectal tumours is well recognised.
- One approach to peptide biomarker detection is Capillary Electrophoresis-Mass Spectrometry (CE-MS).

Method

- Patients with CRC (n=12), colonic adenomas (n=6) and normal controls (n=6) were recruited from UHCW and analysed by Mosaïques Diagnostics GmbH, Hannover, Germany.
- Disease specific peptide marker models were generated using the support vector machine (SVM)-based MosaCluster software.
- Sensitivity and specificity were estimated by calculation of the number of correctly classified samples. The area under curve (AUC) for the receiver operating characteristic (ROC) plot was measured for overall accuracy.

Results

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<th>Male (%)</th>
<th>Median age</th>
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<tbody>
<tr>
<td>CRC</td>
<td>8/12 (66.6)</td>
<td>70</td>
</tr>
<tr>
<td>Adenoma</td>
<td>4/6 (66.6)</td>
<td>75</td>
</tr>
<tr>
<td>Control</td>
<td>3/6 (69.3)</td>
<td>67</td>
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Table 1. Patient characteristics

- Prior work on plasma identified 143 peptide fragments, used for cross reference.
- 52 sequenced urinary peptide markers derived from 19 proteins were identified (figure 2+table 2). The most prominent proteins are collagen alpha-1(I) and collagen alpha-1(III)
- 26 candidate markers from blood and urine were identified to form an SVM-based classification model to calculate sensitivity and specificity data for separating cancer from control (adenoma+normal) (figure 3).
- Sensitivity 100%, specificity 92% (95% CI: 0.84–1), AUC: 0.99 on the training data after total cross-validation.

Aim

This pilot study evaluates the utility of urinary proteome analysis in the diagnosis of CRC using peptide marker patterns analysed by CE-MS.

Validation is required using an independent set of case and control samples to test accuracy.

Table 2. Proteins from CRC peptide marker candidates in urine and number of peptide markers for each protein precursor.

Figure 2. Identification chain from urinary peptides to proteins

Figure 3. Receiver operating characteristic (ROC) curve for measuring the accuracy of the classification model.

Conclusion

- Urinary peptide markers for CRC can be differentiated from control samples with a high degree of accuracy using 26 urinary markers detected via CE-MS in this pilot study.
- Linkage of the identified marker peptides to the pathology of CRC is currently being investigated using immunoassay techniques.