Accurate early-stage colorectal cancer detection through analysis of cell-free circulating tumor DNA (ctDNA) methylation patterns.

Authors:
James M. Kinross, Pol Canal-Noguer, Marko Chersicola, Primož Knap, Marko Bitenc, Alexandre Perera-Lluna, Michael H. A. Roehrl, Kristi Kruusmaa; Section of Biosurgery & Surgical Technology, Department of Surgery & Cancer, Imperial College London, London, United Kingdom; Universal Diagnostics S.L., Sevilla, Spain; Geneplanet D.O.O., Ljubljana, Slovenia; Universitat Politècnica de Catalunya, Barcelona, Spain; Memorial Sloan Kettering Cancer Center, Department of Pathology, Human Oncology and Pathogenesis Program, New York, NY

Abstract Disclosures

Research Funding:
Universal Diagnostics S.L

Background:
Colorectal cancer (CRC) screening programs suffer from poor uptake and biomarkers have limited diagnostic accuracy. The measurement of the methylation status of tumor-derived cell-free DNA in plasma may address these challenges. We used a targeted methylation panel, tumor-derived signal deduction and machine learning algorithm to refine a blood test for the detection of early-stage CRC.

Methods:
This was a prospective, international multicenter observational cohort study. Plasma samples were collected either prior to a scheduled colonoscopy as part of standard colorectal cancer screening or prior to colonic surgery for primary CRC. Differentially methylated regions (DMRs) were initially selected by analyzing CRC and control tissue samples with whole genome bisulfite sequencing. A targeted sequencing assay was designed to capture these DMRs in plasma ctDNA. Individual sequencing reads were evaluated

for cancer-specific methylation signal and scores calculated for each DMR in a sample. A panel of methylation scores originating from 203 DMRs was used in a prediction model building and validated in a test cohort of patients.

Results:

Calculated scores were used to train a machine learning model on 68 ctDNA samples from 18 early stage (I-II) and 16 late-stage (III-IV) CRC patients and 34 age, BMI, gender and country of origin matched neoplasia-free controls (median age 63 [50-74], mean BMI 27 [19.5-37], female 50%, Spanish and Ukrainian population, distal cancers 50%). This model was then applied to an independent set of subjects from Spanish, Ukraine and Germany, including 36 stage I-IV cancer patients (median age 61.5 [55-82], BMI 28 [16-39], female 47%, 42% of the tumors were distal) and 159 age and sex matched controls. 87 of the control patients had a negative colonoscopy finding (cNEG), 19 had hyperplastic polyps (HP), 37 had small non-advanced adenomas (NAA) and 16 were diagnosed with other benign gastrointestinal diseas( )). The model correctly classified 92% (33/36) of CRC patients. Sensitivity per cancer stage ranged from 83% (5/6) for stage I, 92% (11/12) for stage II, 92% (12/13) for stage III to 100% (5/5) for stage IV. Specificity of the model was 97% (154/159), with 100% (37/37) NAA, 94% (15/16) GID, 95% (18/19) HP and 97% cNEG patients correctly identified. Lesion location, gender, BMI, age and country of origin were not significantly correlated to prediction outcome.

Conclusions:

Methylation sequencing data analyzed using read-wise scoring approach combined with a machine-learning algorithm is highly diagnostic for early-stage (I-II) CRCs (89% sensitivity at 97% specificity). This method could serve as the basis for a highly accurate and minimally invasive blood-based CRC screening test with significant implications for the clinical utility of ctDNA in early-stage cancer detection.