Abstract ID number: 48285

Genomic Medicine Sweden

Evaluation of Microsatellite instability score from GMS 560 DNA panel

Anita Koskela von Sydow[1], Alvida Qvick[1], Ingrid Jakobsen[1], Carl Mårten Lindqvist[2], Sanja Farkas[1], Anna Green[1], Helena S. Isaksson[1], Benjamin Holmgren[3], Per Levéen[4] , Subazini Thankaswamy[1] Johan Fredriksson[2], Lina Andersson[1]and Gisela Helenius[2].
[1]University Hospital Örebro, Örebro, Sweden. [2]Örebro University, Örebro Sweden. [3] Halland Hospital, Halmstad, Sweden, [4] Region Skåne and Lund University, Lund Sweden.

Conclusion

We found the microsatellite instability (MSI) score from GMS560 DNA panel to be both diagnostically sensitive and specific (100%) for determining MSI status for colorectal cancer samples. All 33 samples, previously classified for MSI status in the clinic by other used methods, were verified using MSI score calculated by MSIsensor-pro.

Introduction

MSI is characterised by gains or losses of nucleotides in short tandem repeat sequences, microsatellites, dispersed throughout the human genome. MSI status is a molecular fingerprint for DNA mismatch repair deficiency. Clinical detection of MSI status is important for identifying inherited disease in patients with colorectal and endometrial cancer but also has a prognostic value for survival and prediction of treatment response. Lately, MSI has been used as a tumour agnostic biomarker that predicts response to immune checkpoint inhibitors. To identify MSI status clinically, PCR and immunohistochemistry have been the gold standard. On the contrary, NGS provide simultaneous accession of large number of microsatellite loci and can be combined with detection of several other biomarkers.

The national collaboration Genomic Medicine Sweden has developed a solid tumour gene panel composed of 560 cancer associated genes and a bioinformatics pipeline including calculation of MSI score. Our aim was to validate the MSI status based on MSI score from the GMS560 DNA panel against the already clinically established status.

Table 1. Number of patients (N) and MSI score for each group.

		MSI-score %			
	N	Mean	Minimum	Maximum	Std. Deviation
CRC patient MSI	21	26.4	16.5	39.9	5.70
CRC patient MSS	11	2.5	1.2	4.5	1.06
Normal samples	52	1.9	0.4	8.0	1.54
HD832 (HorizonSamples)	19	36.0	33.6	39.4	1.65
NA12878	9	1.0	0.7	1.2	0.17

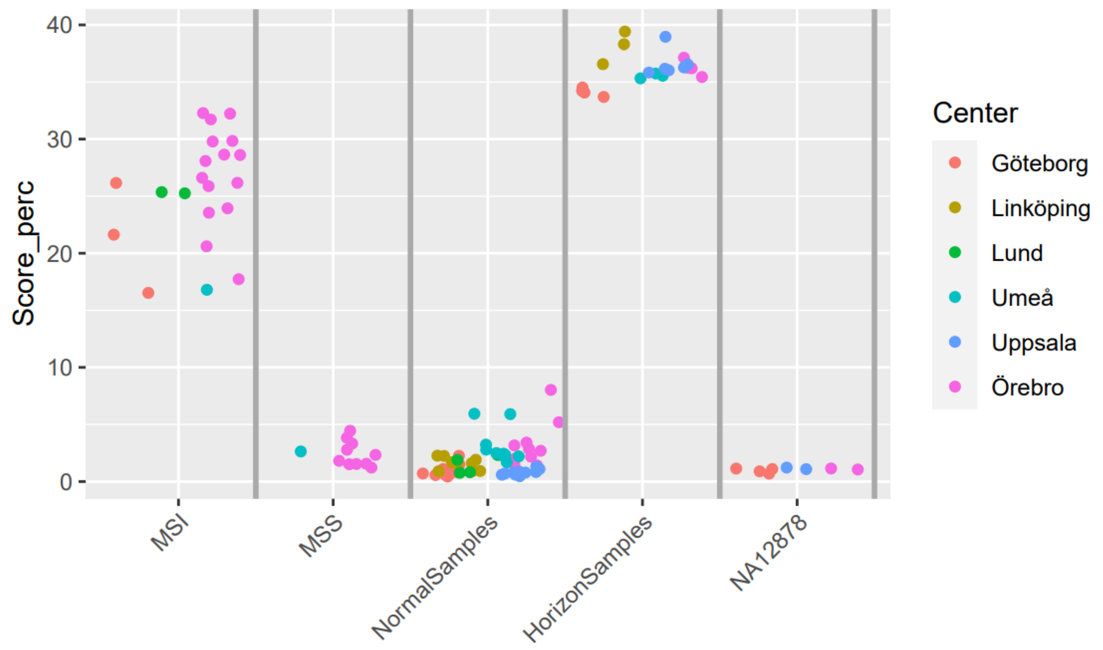

Figure 1. MSI score (as % sites) for each group, analysed at the six different centres.

Material and Methods

The cohort was composed of colorectal cancer samples classified as microsatellite instable (n=21), microsatellite stable (MSS) (n=11), normal tissue samples (52) and general controls HD832 (HorizonSamples) and NA12878. DNA (100 ng) was extracted from formalin-fixed paraffin-embedded tissue with tumour cell content >20%. GMS560 DNA panel was used for target enrichment sequencing analysis and sequenced on the Illumina NextSeq or NovaSeq platform. Allelic distributions from 5000 microsatellite markers were calculated by MSIsensor-pro to generate an instability score.

Results

The colorectal cancer samples classified as MSI had a mean MSI score of 26.4 % (range 16.5-39.9). Corresponding scores for MSS samples was 2.5 % (range 1.2-4.5) and normal samples 1.9 % (range 0.4-8.0). MSI and MSS groups showed full separation, giving assay sensitivity and specificity of 100%. A cutoff value between MSI and MSS can be set between 5-15 %, out of range of the 95% CI for both MSI and MSS samples.

Anita Koskela von Sydow
Department of Laboratory Medicine, Molecular diagnostics
Örebro University Hospital

E-mail: anita.koskela-von-sydow@regionorebrolan.se