Investigation of Human Cancers for Retrovirus by Low-Stringency Target Enrichment and High-Throughput Sequencing

Although nearly one fifth of all human cancers have an infectious aetiology, the causes for the majority of cancers remain unexplained. Despite the enormous data output from high-throughput shotgun sequencing, viral DNA in a clinical sample typically constitutes a proportion of host DNA that is too small to be detected. Sequence variation among virus genomes complicates application of sequence-specific, and highly sensitive, PCR methods. Therefore, we aimed to develop and characterize a method that permits sensitive detection of sequences despite considerable variation. We demonstrate that our low-stringency in-solution hybridization method enables detection of <100 viral copies. Furthermore, distantly related proviral sequences may be enriched by orders of magnitude, enabling discovery of hitherto unknown viral sequences by high-throughput sequencing. The sensitivity was sufficient to detect retroviral sequences in clinical samples. We used this method to conduct an investigation for novel retrovirus in samples from three cancer types. In accordance with recent studies our investigation revealed no retroviral infections in human B-cell lymphoma cells, cutaneous T-cell lymphoma or colorectal cancer biopsies. Nonetheless, our generally applicable method makes sensitive detection possible and permits sequencing of distantly related sequences from complex material.

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