Identification of five novel circular RNAs deriving from BCL2L12 apoptosis-related gene in colon cancer cell lines

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Circular RNAs (circRNAs) result from back-splicing of pre-mRNA. Recent advances in high-throughput sequencing uncovered the widespread expression of circRNAs in cancer cells, arousing scientific interest. However, current knowledge regarding circRNAs deriving from human apoptosis-related genes is poor. BCL2L12 is a member of the BCL2 family, which – despite not bearing a typical anti-apoptotic protein structure – has been reported to exert an anti-apoptotic activity. Moreover, *BCL2L12* is widely expressed in colon cancer. The purpose of this study was to identify novel *BCL2L12* circRNAs in colon cancer cells. Total RNA extracts from seven colon cancer cell lines (Caco-2, HCT 116, HT-29, COLO 205, SW 620, DLD-1, and RKO) were reverse-transcribed, using random hexamer primers. First-round PCR was conducted using divergent primers, to amplify *BCL2L12* cDNAs having derived from circRNAs. Nested PCR was then performed, followed by Sanger sequencing, to determine the sequence of each circRNA. Subsequently, bioinformatical tools were used to predict the interactions of these circRNAs with microRNAs (miRNAs) and RNA-binding proteins (RBPs), and to predict internal ribosome entry sites (IRES) and open reading frames (ORFs). Thus, we discovered five circRNAs, all consisting of both complete and truncated exons of *BCL2L12* gene. Expression analysis revealed differential expression patterns of these circRNAs in the seven colon cancer cell lines. Next, in silico analysis revealed that these circRNAs are likely to interact with miRNAs and RBPs; for instance, circ-BCL2L12-1 is predicted to bind miR-1915-5p, while circ-BCL2L12-1 and circ-BCL2L12-2 are predicted to sequester splicing factors such as MNBL1, SRFS1, SRFS2, SRFS3, and SRFS5. Moreover, circ-BCL2L12-2 is predicted to possess a functional ORF, but no IRES. In conclusion, this study uncovered, for the first time, the complete sequence of circRNAs deriving from *BCL2L12* and suggested some putative interactions of these molecules.