Supplementary Materials: Early Assessment of Colorectal Cancer by Quantifying Circulating Tumor Cells in Peripheral Blood: ECT2 in Diagnosis of Colorectal Cancer

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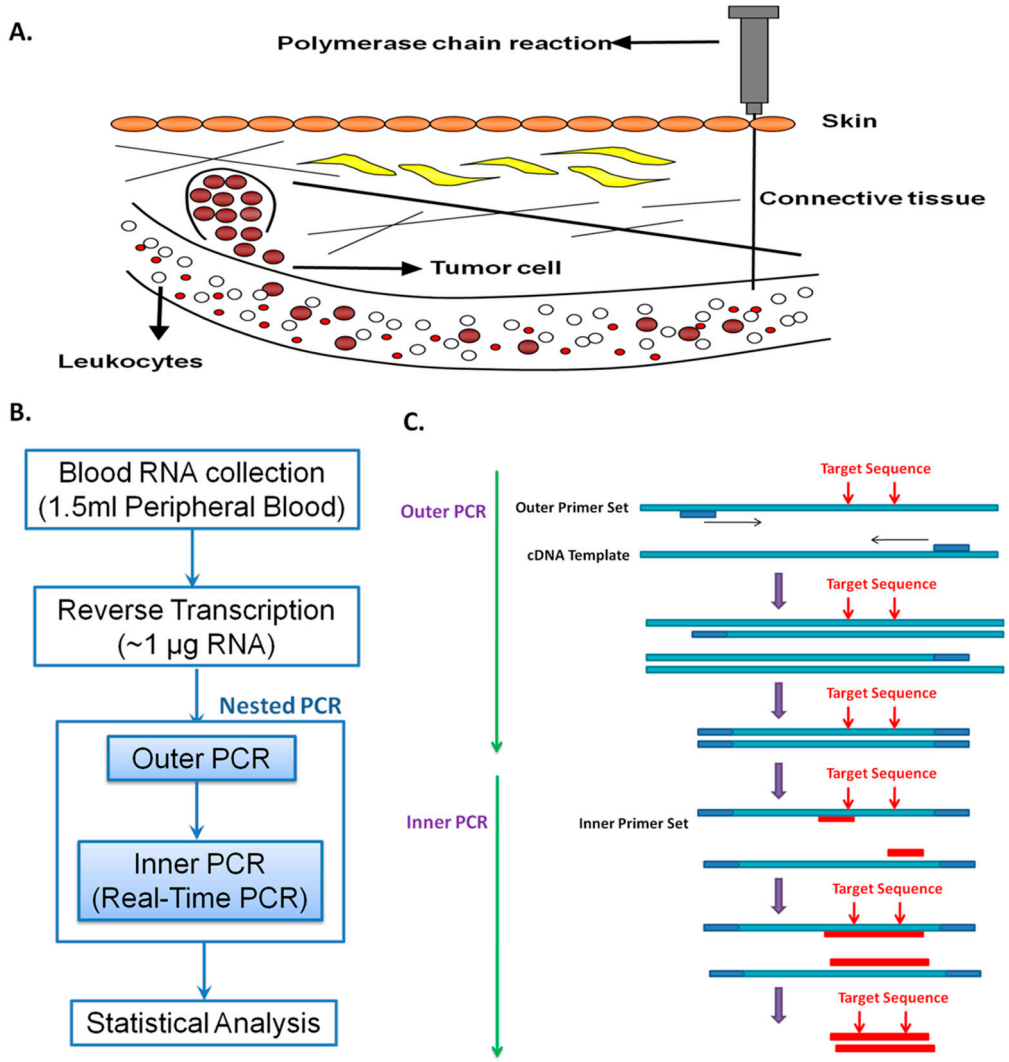

Figure S1. Candidate marker genes examined for detection of circulating tumor cells by nested real-time polymerase chain reaction (PCR). (**A**) Tumor cells undergoing the epithelial to mesenchymal transition (EMT) acquire motile propensities and enter the blood. A 1.5 mL sample of peripheral blood from colorectal cancer patients was directly taken for candidate marker gene determination. (**B**) RNA was extracted from cells isolated from the peripheral blood. After reverse transcription of RNA, expression of candidate marker genes was quantified by nested real-time PCR. (**C**) Nested real-time PCR diagram. First, a target sequence is amplified by an outer primer set with a few rounds. The PCR products from the first PCR reaction are subjected to real-time PCR using a hydrolysable fluorescent probe for detection. Two rounds of PCR as well as sequence-based probe detection ensure the sensitivity and specificity of this method.