Identification of MAGE A1-A10 mRNA from testicular tissue using the common primer for RT-PCR

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Abstract

One of the cancer that expressed the MAGE A mRNA is lung cancer. The specimen for diagnose of lung cancer are usually just a few of tissue from core biopsy, forcep biopsy, Fine needle aspiration biopsy, or bronchoalveolar lavage. The melanoma antigen (MAGE) gene is the only gene that is expressed in cancer cells and in testicular tissue. The MAGE A gene is consist of 12 variants called MAGE A1, A2, A3, A4, A5, A6, A7 (pseudo gene), A8, A9, A10, A11, and A12. The MAGE A mRNA can be detected using the Reverse Transcription-Polymerase Chain Reaction (RT-PCR), but it is required a common primer for detecting the most of MAGE A variants. The objective of this study was to develop the common primer for detecting the MAGE A1-A10 mRNA. The specimen used in this research was testicular tissue taken from patients who had received the orchidectomy therapy at Dr Soetomo Hospital, Surabaya, Indonesia, in 2017. Detection of MAGE A1-A10 mRNA was performed by RT-PCR technique using the common primer for MAGE A1-A10. MAGE A1, A2, A3, A4, A5, A6, A8, A9, and A10 variants mRNA were detected using primer set of GMF10/GMR10 for the first round and primer set of GMF10/GMR12 for the second round. Primer set of MMRP1/MMRP2 was used as a comparison for MAGE A1-A6 mRNA identification. The mRNA from testicular tissue was extracted, followed by RT-PCR. The PCR products were analysed by 2% agarose gel electrophoresis. Result showed that the PCR products of primer pair GMF10/GMR10 ranges from 823-919 base pair (bp) whereas products of GMF10/GMR12 were 461-557bp in size depending on the variants of MAGE A1-A10. The next optimization test was RNA total dilution test. A total of 133.4 ng/ml of RNA was used, and was diluted with a ratio of 1:10, 1:100, 1:1000. All of the MAGE A1-A10 mRNA can be detected together in the same tube PCR. This common primer can be used as a biomolecular tool for identification of all variants of MAGE A1-A10 mRNA in cancer cells.

Keywords: MAGE A1-A10 mRNA; common primer; testicular tissue; cancer testis antigen; RT-PCR

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