عنوان مقاله:

The identification of single strand DNA aptamers which specifically bind to platelets using cell-SELEX technique

محل انتشار:

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نویسندگان: فاطمه عالمی - Division of Biotechnology, Faculty of Veterinary Medicine, Ferdowsi University of Mashhad, Mashhad, .Iran

محتبي سنكيان . Immunology Research Center, Faculty of Medicine, Mashhad University of Medical Sciences .Mashhad, Iran

عليرضا حق يرست - Division of Biotechnology, Faculty of Veterinary Medicine, Ferdowsi University of Mashhad, .Mashhad, Iran

محمدرضا باسامي . Division of Biotechnology, Faculty of Veterinary Medicine, Ferdowsi University of Mashhad, .Mashhad, Iran & Research Institute of Biotechnology, Ferdowsi University of Mashhad, Mashhad, Iran

غلامرضا هاشمي تبار - Department of Pathobiology, Faculty of Veterinary Medicine, Ferdowsi University of Mashhad .Mashhad, Iran & Research Institute of Biotechnology, Ferdowsi University of Mashhad, Mashhad, Iran

خلاصه مقاله:

Aptamers are oligonucleotides that can be easily synthesized and bind to their targets with high affinity and specificity. Several aptamers specific to soluble factors of coagulation cascade have been produced, however, aptamers specific to platelet cell membrane molecules have not been reported yet. We aimed to discover DNA aptamers that specifically bind to human platelets. The cell-SELEX method was used for aptamer discovery. Synthetic V9 nucleotides length single-strand oligonucleotides were used as a library. Ultra-pure platelets were prepared using differential centrifugation steps and magnetic-bead-assisted removal of contaminating cells. The FITC-labeled forward primer was used for amplification of the selected oligonucleotides by PCR, and Lambda exonuclease was used for digestion of the lagging strand. After IY rounds of cell-SELEX, selected oligos were amplified and cloned to pTG19-T vector, transfected into E. coli (TOPI) and sequenced. Sequences of aptamers from Yoo individual positive colonies were aligned and seven clusters were identified. Representative aptamers were amplified and their affinity, specificity, and digestibility of their targets were evaluated. Interferences of the aptamers to two platelet function tests were also investigated. Affinity (KD) of the representative aptamers were between 1.9 and ٣F. nM. Trypsin exposure of the platelets completely abolished the binding of the Y aptamers to the targets. The binding of the four aptamers fully protected their target molecules from digestion. No one of the aptamers changed the parameters of the platelet function tests. Seven aptamers specific to platelets were identified and characterized. These aptamers may have potentially diverse applications in the diagnosis or treatment of platelet disorders.

كلمات كليدي:

Cell-SELEX, Platelet, DNA aptamer, Platelet-specific aptamer

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