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School of Pharmacy and Pharmaceutical Sciences
Isfahan University of Medical Sciences

Construction of a plasmid containing D 297G mutation of V2 vasopressin receptor

Y. Sebti[1,*], H. Mir Mohammad Sadeghi[1], S. Sardari[2]

[1]Department of Pharmaceutical Biotechnology and Isfahan Pharmaceutical Sciences Research Center, School of Pharmacy and Pharmaceutical Sciences, Isfahan University of Medical Sciences, Isfahan, I.R. Iran
[2]Medical Biotechnology Department, Pasteur Institute of Iran, Tehran, I.R. Iran

Background and Aims: Based on the previous studies it seems that D 297G mutation of V2 Vasopressin Receptor involved in receptor binding domain. Therefor construction a plasmid containing the mutated V2 receptor cDNA seems to be the necessary first step in this project.
Methods: The specific primers were designed. Nested Polymerase Chain Reaction (PCR) was used for synthesizing the mutant V2 receptor inserts. After sequencing, the PCR product having the desired mutation was ligated to a suitable vector (pcDNA3). Transformations were carried out by 2 methods; using a commercial kit method and heat shock method. After cloning the orientation of the insert were concerned.
Results: In the first round of PCR, two pieces of 1203 and 361bp DNAs were produced. After extraction of these two above segments, they used as a template for nested PCR. The final PCR product was sequenced and the desired mutation was approved. Plasmids containing suitable insert are under investigation.
Conclusions: Construction of the mutated V2 receptor cDNA was done successfully.

Keywords: V2 Vasopressin receptor; Mutation; Binding