Fast and easy construction method of ryanodine receptor mutants aiming at genetic screening of malignant hyperthermia diagnosis

Hideto Oyamada[1,2], Masahide Nakano[1,3], Takahiro Hayashi[1], Yusuke Ubukata[1,4], Takashi Makino[1], Masaaki Tanaka[1,5], Takuya Kikuchi[1,6], Katsuji Oguchi[1,2], Yuji Kiuchi[1,2]

[1]Dept Pharmcol, Sch Med, SHOWA Univ, [2]Pharmacol Res Centr, SHOWA Univ, [3]Dept Surg, Thyroid Centr, SHOWA Univ, Northern Yokohama Hosp, [4]Dept Anesthesiol, Ebara Hosp, [5]Dept Anesthesiol, Heisei Yokohama Hosp, [6]Kikuchi Eye Clinic

Malignant hyperthermia (MH) is a potentially fatal pharmacogenetic disorder that manifests clinically as a hypermetabolic with skeletal muscle rigidity when MH-susceptible (MHS) individual is exposed to commonly used volatile anesthetics. At present, definitive diagnosis of MH before anesthesia requires the detection of the functional abnormality using biopsied muscle samples, a painful test for patients and requiring skillful diagnosticians. Recent reports showed that the frequency of MH episodes has increased though the MH crises are very rare case. The exact percentage of MHS is difficult to determine, but the prevalence of MH can be estimated up to 1:2750 due to the autosomal -dominant inheritance in humans. The noninvasive diagnosis such as the genetic screening of the MHS is asked for, because safer anesthesia is performed. Approximately 50 % of known cases of MH are caused by mutations in the gene locus of the ryanodine receptor type 1 (RyR1, calcium release channel) and the numbers of RyR1 mutation sites reported in the patients have been increased up to about 200, but only 48 have been formally shown to be causative; the remainder await confirmatory studies. So we have employed the fast and easy making procedures for these mutants to be expressed in stable cultured cells to be discussed in this paper.