

Z6 16 Expression and purification of Enteroviral proteases, 2Apro and 3Cpro

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Enteroviral proteases, 2Apro and 3Cpro cleave host cellular proteins that require cap-dependent translation. This process is accompanied by the cleavage of the translation initiation factor eIF4G, a component of the cap-binding complex eIF4. This cleavage is mediated by the viral proteases, 2Apro and 3Cpro. In this experiment, enteroviral proteases, 2Apro and 3Cpro were cloned and expressed in E. coli expression system. The viral cDNA fragment containing the proteases coding region was cloned into the pET29 (a) vector and expressed in the E. coli strain, BL21 (DE3). Expressed poly-histidine-tagged proteins were then purified by a Nickel-affinity column and the enzymatic activity of the purified protein was measured by a fluorescent assay. The purified enteroviral proteins have cysteine protease activities and their optimal temperature and pH were determined. The proteases characterized in this study enable us to develop in vitro protease inhibitor assay system, so that the system would be used as a mass screening strategy of anti-viral agents, in vitro.

Z6 17 HNA-1a and HNA-1b Frequencies among Koreans

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The human neutrophil antigens (HNA)-1a and -1b are polymorphic forms of the Fc γ RIIIb which plays a major role in immune neutropenias and pulmonary transfusion reactions. FCGR3B revealed that all 5 nucleotide substitutions which are associated with HNA-1a and HNA-1b polymorphism are located on exon 3 coding for the membrane distal extracellular domain. We analyzed HNA-1a and HNA-1b polymorphism of FCGR3B using PCR-sequence specific primer in 280 unrelated Koreans. The allele frequencies FCGR3B*1 and FCGR3B*2 were 0.61 and 0.39, respectively. No deviation from expectation according to Hardy-Weinberg equilibrium found. The FCGR3B*1 allele frequency is more frequent than FCGR3*2 and these frequencies were similar to the reported frequencies in Japanese and Chinese populations. The FCGR3B*1 allele frequency among Oriental population is twice as high as that of Caucasians.