学位論文抄録

Effects of cytotoxic T lymphocyte escape-conferring HIV-1 Nef variants on viral receptor down regulation and susceptibility to superinfection

（細胞傷害性T細胞から逃避するHIV-1 Nef変異株におけるウイルス受容体の発現低下と重複感染に関する研究）

ムイマンズィ フィリップ マカヤンスィ

Mwimanzi, Philip Makayanswi

熊本大学大学院医学教育部博士課程
病態制御学専攻エイズ先端研究者育成コース

指導教員

上野 貴将 准教授
熊本大学大学院医学教育部博士課程医学専攻エイズ学Ⅴ

Abstract of the Thesis

Background and purpose: Nef is an accessory gene product encoded by primate lentiviruses HIV-1, HIV-2 and SIV and is a key factor for viral pathogenesis in vivo. HIV-1 Nef is known to down-regulate functionally important molecules, including viral entry receptor CD4 and CCR5 and CXCR4, from the surface of HIV-infected cells. Some of these Nef activities are mediated by the well-conserved proline-rich region of Nef, and this region is highly targeted by cytotoxic T lymphocytes (CTLs). In the present study, I asked whether Nef variants selected under CTL-mediated selective pressure in vivo may constrain these important Nef activities.

Methods: I analyzed autologous *nef* sequences isolated from a cohort of total 235 subjects in Japan, I tried to look for mutations that were over-represented in subjects having specific HLA class I alleles. I analyzed cytotoxic activity of HIV-1 specific CTL clones toward cells expressing Nef fusion proteins having various mutations. I also analyzed Nef activity in viral receptor and co-receptor down regulation and tested for these effects on susceptibility to superinfection.

Results: The analysis of autologous *nef* sequences revealed that the subjects showing amino acids variations, Arg75Thr (75T) and Tyr85Phe (85F), located within the proline-rich region were significantly over-represented by those having *HLA-B*3501*. HLA-B*3501 restricted CTL clones did not kill target cells expressing 75T and 85F or the combination of the two – 75T/85F (TF) variant, indicating that these variants conferred escape from HLA-B*3501-restricted CTLs. The 75T and double mutant 75T/85F (TF) variant Nef selectively impaired CCR5, but not CXCR4, down-regulation activity from the cell surface; whereas the 85F variant Nef alone affected neither CCR5 nor CXCR4 down-regulation activity. These CTL escape mutations either alone or in combination had no effect on cell surface expression of CD4. Moreover, the cells expressing the 75T or TF variant Nef significantly impaired protection from superinfection by CCR5-tropic, but not CXCR4-tropic, viruses.

Conclusion: I showed that some naturally arising amino acid variations in the well-conserved proline-rich region of Nef are associated with escape from HLA-B*3501-restricted CTLs. One of these variants, Arg75Thr selectively impaired CCR5, but not CXCR4, down-regulation activity by Nef and decreased protection capacity against superinfection by CCR5-tropic HIV-1 but not CXCR4-tropic viruses. My findings highlight the importance of certain Nef-specific CTLs in modulation of viral co-receptor down-regulation activity and protection from HIV-1 superinfection, providing us with an additional insight in vaccine design.