A-11: Entrapment and inhibition of human immunodeficiency virus proteinase by α_2-macroglobulin and structural changes in the inhibitor

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Occurrence of antibodies against Human immunodeficiency virus (HIV) proteinase in the plasma of acquired immunodeficiency syndromes (AIDS) patients has been reported suggesting a possibility of the presence of HIV proteinase in their sera. Further, mutation in the proteinase coding region of the HIV genome have been shown to result in production of non-infective virions and thus HIV proteinase has been targeted as a possible therapeutic intervention point in the treatment of AIDS. Therefore it seems to be of fundamental importance and interest to study the effect of α_2-macroglobulin(α_2M), a major proteinase inhibitor in plasma, on the proteolytic activity of HIV proteinase.

The activity of HIV proteinase towards the Moloney murine sarcoma virus-derived gag protein (a high molecular mass substrate) was found to be inhibited by α_2M at pH 5.5-7.4. On the other hand, the activity towards B chain of oxidized insulin (a low molecular mass substrate) was scarcely inhibited. Thus, the active site of HIV proteinase in the α_2M-HIV proteinase complex is inaccessible to the MSV substrate, although accessible to the B chain of oxidized insulin, indicating the entrapment of HIV proteinase by α_2M.

The complex of α_2M and HIV proteinase was isolated by gel filtration and the enzyme shown to be significantly protected by the complex formation from autoinactivation under nonreducing conditions. The stoichiometry of the complex formation was found to be 2:1(enzyme : α_2M, mol/mol). Under these conditions, HIV proteinase cleaved α_2M at the Phe^{684}-Tyr^{685}bond in the bait region sequence, resulting in a drastic conformational change in the inhibitor as revealed by electron microscopy, polyacrylamide gel electrophoresis.

These results demonstrate the covalent entrapment and concomitant inhibition of HIV proteinase by α_2M.