

SAMT ANALOG INACTIVATION OF HIV-1 NUCLEOCAPSID NCP7: *IN VITRO* MECHANISM AND *IN VIVO* EFFICACY

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Molecules that inactivate the HIV-1 nucleocapsid protein (NCp7) are currently being evaluated as new antiviral drugs. Among them, derivatives based on a 2-mercaptobenzamide thioester template (SAMTs) have been developed to specifically eject coordinated zinc from NCp7 and are being explored for use as topical microbicides. The SAMT compounds function *via* acyl transfer from the thioester to a lysine residue in the protein by means of the sulfur of a zinc-coordinating cysteine residue. Using a panel of active and inactive analogs, we have also explored the basis for the antiviral effects of the SAMT compounds in infected cells. These studies have demonstrated that the compounds specifically induce processing defects of the gag polyprotein which result in accumulation of unprocessed, aggregated gag protein. These processing defects likely give rise to the potent antiviral activity observed for the SAMT compounds. We have demonstrated that the SAMT compounds prevent cell-to-cell transmission of HIV, inhibit dissemination of virus from cervical explants, and confer protection from infection to Rhesus macaques. We have also established that the SAMTs retain activity in gel formulation and are safe for vaginal use in rabbit vaginal irritation studies. We are initiating formulation studies to explore means for drug delivery. Preliminary studies with silicone rings have found that the SAMTs are safely released from these rings *in vivo*. Finally, mass spectrometry experiments have revealed a panel of potential metabolites of the SAMT compounds that could be useful to understand the metabolic fate of the compounds *in vivo*. Together, these experiments represent the first demonstrations that a small molecule targeting the retroviral NCp7 can be safely used as a topical microbicide to prevent infection. The highly specific interactions that are observed between SAMTs and NCp7 highlight the potential power offered by compounds directed towards a target that is resistant to viral mutation.