

Oral presentation

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## Orally Delivered, Plant-Produced Tat Protein Primes Mice For a Challenge DNA Vaccine Expressing Tat

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The Tat protein has been recently explored as a prospective vaccine candidate against HIV-1 with broad, subtype non-specific action. A truncated version of Tat(ΔTat) with the basic loop, involved in immunosuppression, removed has been previously demonstrated as efficacious as the full-size Tat protein. We produced both full-size Tat and truncated ΔTat in plants, including one edible species – spinach, thus simultaneously addressing problems of an inexpensive Tat production and a direct delivery through the mucosal route. We tested this oral delivery route in a mouse model. *Tat* and Δ*Tat* genes were assembled from a set of synthetic overlapping oligonucleotides, and subsequently cloned into a plant virus-based expression vector. Codon optimization allows production of up to 300–500 mg of Tat or ΔTat antigen per 1 g of leaf tissue in spinach. Spinach plants inoculated with the Tat-producing constructs were collected and fed to mice 7–14 days post inoculation with or without mucosal adjuvants. Mice were fed with the Tat-producing or control vector-inoculated spinach. After 3 voluntary feedings, 1 week apart, 1 g per mice, no differences were detected in the growth rate or behavior of the animals fed with these two types of spinach. None of the animals developed measurable Tat antibodies. Challenge DNA vaccination with a homologous Tat-expressing construct was performed using a gene gun. Following DNA vaccination, however, mice previously receiving oral Tat with cholera toxin as an adjuvant, developed higher antibody titers to Tat than did the controls, with the titers peaking at four weeks post-vaccination. Thus, our data suggested that oral Tat primed for the development of Tat antibodies when mice were challenge-vaccinated with plasmid DNA for expression of Tat.