Glycosylation deficiency at either one of the two glycan attachment sites of cellular prion protein preserves susceptibility to bovine spongiform encephalopathy and scrapie infections.

The conversion into abnormally folded prion protein (PrP) plays a key role in prion diseases. PrP(C) carries two N-linked glycan chains at amino acid residues 180 and 196 (mouse). Previous in vitro data indicated that the conversion process may not require glycosylation of PrP. However, it is conceivable that these glycans function as intermolecular binding sites during the de novo infection of cells on susceptible organisms and/or play a role for the interaction of both PrP isoforms. Such receptor-like properties could contribute to the formation of specific prion strains. However, in earlier studies, mutations at the glycosylation sites of PrP led to intracellular trafficking abnormalities, which made it impossible to generate PrP glycosylation-deficient mice that were susceptible to bovine spongiform encephalopathy (BSE) or scrapie. We have now tested more than 25 different mutations at both consensus sites and found one nonglycosylated (T182N/T198A) and two monoglycosylated (T182N and T198A) mutants that rather retained authentic cellular trafficking properties. In vitro all three mutants were converted into PrP(res). PrP mutant T182N/T198A also provoked a strong dominant-negative inhibition on the endogenous wild type PrP conversion reaction. By using the two monoglycosylated mutants, we
generated transgenic mice overexpressing PrP(C) in their brains at levels of 2-4 times that of nontransgenic mice. Most interestingly, such mice proved readily susceptible to a challenge with either scrapie (Chandler and Me7) or with BSE. Incubation times were comparable or in some instances even significantly shorter than those of nontransgenic mice. These data indicate that diglycosylation of PrP(C) is not mandatory for prion infection in vivo.