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The number of oligosaccharide side chains on rabies virus glycoprotein (G-protein) was investigated. Analysis of glycoproteins obtained by protease diges-

2 μ g/ml. and the solution was incubated at 37°C for 1 following glycopeptides of known molecular

h. weights: thyroglobulin glycopeptide (molecular

Preparation of tryptic glycopeptides. Tryptic weight 4,100) (15), fetuin glycopeptide (molecular

digestion of rabies virus G-protein was carried out essentially as described by Cooper et al. (4). Isolated weight 4,400) (2), and ovalbumin glycopeptide (molecular weight 1,500) (17). Furthermore, raffinose

G-protein was suspended in NTE buffer (0.1 M NaCl-0.05 M Tris-hydrochloride [pH 7.5]-0.001 M and stachyose were used as standards. Ovalbumin, raffinose, and stachyose were detected by the

EDTA), 0.5 mg of bovine serum albumin was added, and the suspension was heated at 100°C for 1 min phenol-sulfuric acid assay (16), and thyroglobulin was detected by the thiobarbituric acid assay (19).

with 1/1,000 volume of a mixture of 2-hydroxy-

ethyldisulfide and 2-mercaptoethanol (50:1) and

RESULTS

then dialyzed against 1,000 volumes of 0.05 M

labeled amino acids or [^3H]glucosamine. A sin- rus grown at 33 and 37°C were exactly the

gle major viral G-protein and a smaller minor same size (data not shown). These results sug-

G-protein were labeled with [^3H]glucosamine. gest that the amount of sialic acid per glycopro-

The minor G-protein, which migrated slightly faster than the nucleocapsid protein, is a degra- tide is variable and that the G-protein in rabies virus grown at high temperature contains less

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mogeneous with respect to size. Each of the

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when tryptic glycopeptide I (Fig. 6b) was di-

[redacted] cannot exclude the possibility that protease

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