Polysialic acid (PSA) is a linear homopolymer of α-2,8-linked sialic acid residues whose expression is developmentally regulated and modulates the adhesive property of the neural adhesion molecule, N-CAM. Recently, hamster and human cDNAs encoding polysialyltransferase (PST-1 for the hamster enzyme and PST for the human enzyme) were cloned, and by using the human cDNA it was demonstrated that the expression of PSA in N-CAM facilitates neurite outgrowth (Nakayama, J., Fukuda, M.N., Fredette, B., Ranscht, B., and Fukuda, M. (1995) Proc. Natl. Acad. Sci. U. S. A., 92, 7031–7035; Eckhardt, M.A., MüHlenhoff, M., Bethe, A., Koopman, J., Frosch, M., and Gerardy-Schahn, R. (1995) Nature 373, 715–718.) Although these studies demonstrated that PST-1 and PST synthesize PSA in cultured cells, it was not shown that they could catalyze the polycondensation of α-2,8-linked sialic acid on a glycoconjugate template containing α-2,3-linked sialic acid. Here we demonstrate that PSA formation by PST is independent from the presence of N-CAM in vivo. We then develop an in vitro assay of PSA synthesis using glycoproteins other than N-CAM as acceptors and a soluble PST as an enzyme source. The soluble PST, produced as a chimeric protein fused with protein A, was incubated with rat α1-acid glycoprotein, fetuin or human α1-acid glycoprotein as acceptors together with the donor substrate CMP-[14C]NeuNAc. Incubation of fetuin with the soluble PST, in particular, resulted in a high molecular weight protein fused with protein A, was incubated with rat α1-acid glycoprotein, fetuin or human α1-acid glycoprotein as acceptors together with the donor substrate CMP-[14C]NeuNAc. Incubation of fetuin with the soluble PST, in particular, resulted in a high molecular weight product that was susceptible to PSA-specific endonemaminidase. Polysialylated products were not formed when α-2,8-linked sialic acid was removed from the acceptor fetuin before incubation. These results establish that a single enzyme, PST, alone can catalyze both the addition of the first α-2,8-linked sialic acid to α-2,3-linked sialic acid and the polycondensation of all α-2,8-linked sialic acids, yielding PSA. 

Communication

A Human Polysialyltransferase Directs in Vitro Synthesis of Polysialic Acid*

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J un Nakayama and Minoru Fukudat

From the Glycobiology Program, Cancer Research Center, La Jolla Cancer Research Foundation, La Jolla, California 92037

Polysialic acid (PSA) is a linear homopolymer of α-2,8-linked sialic acid residues whose expression is developmentally regulated and modulates the adhesive property of the neural adhesion molecule, N-CAM. Recently, hamster and human cDNAs encoding polysialyltransferase (PST-1 for the hamster enzyme and PST for the human enzyme) were cloned, and by using the human cDNA it was demonstrated that the expression of PSA in N-CAM facilitates neurite outgrowth (Nakayama, J., Fukuda, M.N., Fredette, B., Ranscht, B., and Fukuda, M. (1995) Proc. Natl. Acad. Sci. U. S. A., 92, 7031–7035; Eckhardt, M.A., MüHlenhoff, M., Bethe, A., Koopman, J., Frosch, M., and Gerardy-Schahn, R. (1995) Nature 373, 715–718.) Although these studies demonstrated that PST-1 and PST synthesize PSA in cultured cells, it was not shown that they could catalyze the polycondensation of α-2,8-linked sialic acid on a glycoconjugate template containing α-2,3-linked sialic acid. Here we demonstrate that PSA formation by PST is independent from the presence of N-CAM in vivo. We then develop an in vitro assay of PSA synthesis using glycoproteins other than N-CAM as acceptors and a soluble PST as an enzyme source. The soluble PST, produced as a chimeric protein fused with protein A, was incubated with rat α1-acid glycoprotein, fetuin or human α1-acid glycoprotein as acceptors together with the donor substrate CMP-[14C]NeuNAc. Incubation of fetuin with the soluble PST, in particular, resulted in a high molecular weight product that was susceptible to PSA-specific endonemaminidase. Polysialylated products were not formed when α-2,8-linked sialic acid was removed from the acceptor fetuin before incubation. These results establish that a single enzyme, PST, alone can catalyze both the addition of the first α-2,8-linked sialic acid to α-2,3-linked sialic acid and the polycondensation of all α-2,8-linked sialic acids, yielding PSA. 

Polysialic acid (PSA) is a developmentally regulated glycan composed of a linear homopolymer of α-2,8-linked sialic acid residues. PSA is mainly linked to the neural cell adhesion molecule (N-CAM) and is more abundant in embryonic brain than adult brain. Presence of this large negatively charged carbohydrate modulates the adhesive property of N-CAM (1–3), and the removal of PSA from N-CAM increases the adhesive capability of N-CAM (4, 5). It was also demonstrated that the presence of PSA affects cell-cell interactions carried by other cell surface receptors (6, 7). Since PSA is mainly present in tissues undergoing synaptic rearrangement and cell migration, PSA is implicated in reducing N-CAM adhesion and thus perhaps in allowing increased neurite outgrowth and cell mobility. It was shown previously that the reducing terminus of a polysialic acid side chain is attached to α-2,3-linked sialic acid, which in turn, is linked to a galactose residue in a presumed acceptor such as N-CAM (3). It has been suggested that PSA synthesis requires two distinct α-2,8-sialyltransferases; the first “initiation” enzyme, which adds a single α-2,8-linked sialic acid to α-2,3-linked sialic acid in the acceptor and a “polymerase,” which adds multiple α-2,8-linked sialic acid residues to the NeuNAcα2→8NeuNAcα2→3Galα(R structure (8, 9). As a support for this hypothesis, the recently cloned ST8SiaI was shown to add a single α-2,8-linked sialic acid residue to a terminal α-2,3-linked sialic acid in N-glycans, but not to form PSA (10). As an alternative biosynthetic pathway, it is possible that a single enzyme catalyzes all the reactions that form α-2,8-sialic acid linkages. Recently, we and others have cloned a cDNA of polysialyltransferase (PST for human and PST-1 for hamster enzyme) that forms polysialic acid attached to N-CAM (11, 12). The amino acid sequences of PST and PST-1 are more than 97% identical. The introduction of PST or PST-1 cDNA together with N-CAM cDNA into various cell lines, such as COS-1, CHO-2A10, HeLa, and 3T3 cells, resulted in PSA expression in those cells. More recently, it has been demonstrated that STX also directs the expression of PSA in small cell lung carcinoma cell lines (13). STX was originally cloned as a sialyltransferase predominantly present in fetal brain (14), and STX and PST have 59% identity in the amino acid sequences (11, 12). These results suggest that PST and STX catalyze polycondensation of α-2,8-linked sialic acid residues. However, these results did not formally exclude the possibility that PST or STX adds the first α-2,8-linked sialic acid as an initiation enzyme, and a hitherto unknown enzyme present in those tested cells catalyzes the polycondensation of polysialic acid. Moreover, it was not certain whether or not PST or STX adds polysialic acid residues to a single α-2,8-linked sialic acid residue, which was already attached by other α-2,8-sialyltransferases such as ST8SiaII to an α-2,3-linked sialic acid residue in a precursor. To determine whether or not PST can catalyze both initiation and polycondensation, we report here the construction of a PST chimeric protein which consists of a signal peptide, protein A, and a catalytic domain of PST. This chimeric protein, released into the culture medium from COS-1 cells, was then incubated with various glycoprotein acceptors and donor substrate CMP-[14C]NeuNAc. By analyzing the products, we show that PST directly adds an α-2,8-sialic acid residue on an α-2,3-linked sialic acid in a glycoprotein template, and in addition catalyzes polycondensation of α-2,8-linked sialic acid, demonstrating that PST can add all α-2,8-linked sialic acid residues necessary for PSA formation.
Polyasialic Acid Formation by a Single Enzyme

EXPERIMENTAL PROCEDURES

Antibodies—Mouse monoclonal antibodies M6703 (15), 12E3 (16), and 735 (17) were kindly provided by Drs. Yoshio Hirohata (RIKEN, Wako, Japan), Tatsunori Seki and Yasumasa Araki (J. undento University School of Medicine, Tokyo, Japan), and Rita Gerard-Schahin (Hannover University School of Medicine, Hannover, Germany), respectively. M6703 was shown to react with at a dimer or trimer of α-2,8-linked sialic acid attached to α-2,3-linked sialic acid (15). The antibodies 12E3 and 735 were shown to react with polysialic acid containing six or more α-2,8-linked sialic acid residues (for 12E3) (18) or eight or more α-2,8-linked sialic acid residues (for 735) (19). Mouse monoclonal anti-human N-CAM antibody MY31 was obtained from Becton-Dickinson.

Transient or Stable Expression of PST—pcDNAI-PST harboring a cDNA encoding PST was cloned as described (11). COS-1 cells were transfected with pcDNAI-PST by Lipofectamine (Life Technologies, Inc.) as described (11). After 48 h, the transfected COS-1 cells were examined by immunofluorescent staining using M6703, 12E3, or 735 antibody followed by fluorescein isothiocyanate (FITC)-conjugated (Fab’), fragment of goat anti-mouse IgG antibody (for M6703 and 735) or FITC-conjugated (Fab’), fragment of goat anti-mouse IgM (for 12E3), as described previously (11).

In parallel, HeLa cells were transfected with pcDNAI-PST and pSV-neo, and the transfected cells were selected with G418 (Life Technologies, Inc.). Clonal cell lines stably expressing PSA were chosen by immunofluorescent staining with 735 antibody as described above. Both the transfected COS-1 and HeLa cells were examined for the presence of N-CAM by immunofluorescent staining with mouse anti-N-CAM antibody, followed by FITC-conjugated (Fab’), fragment of goat anti-mouse IgG antibody.

Construction of a Soluble Chimeric PST—pcDNAI-A-PST harboring cDNA encoding a fusion protein of the signal peptide of the human granulocyte colony-stimulating factor, the IgG binding domain of Staphylococcus aureus protein A, and a catalytic domain of PST was constructed as follows.

The cDNA encoding a catalytic domain of PST was prepared by polymerase chain reaction (PCR) using pcDNAI-PST (11) as a template. Upstream and downstream primers used were 5’-ggggatccgGGTGAAT-9 and 5’-ggtagctcTCAAAATGTGCTTTATTGCTTTACAC-3’ (KpnI site shown by underline), and 5’-gggtaccTCAAAATGTGCTTTATTGCTTTACAC-3’ (KpnI site shown by underline), respectively. The PCR product encompasses the sequence from nucleotide 118 (codon 40) to nucleotide 1092 (12 nucleotides after the stop codon). In parallel, pAMoA-GD3 (20) was digested with BamHI and KpnI, resulting in pAMoA containing cDNA encoding only the signal peptide and protein A. pAMoA-GD3 was kindly provided by Drs. Katsutoshi Sasaki and Tatsunori Nishi, Kyowa Hakko Kogyo Co. (Machida, Japan). The PCR product of PST was digested with BamHI and KpnI and cloned into pAMoA digested with the same restriction enzymes, yielding pAMoA-PST. From this pAMoA-PST, the cDNA encoding the fusion protein consisting of the signal peptide, the protein A and the PST catalytic domain was excised by Sall and Aspl18 digestion. The released cDNA insert was filled in by the Klenow fragment of DNA polymerase I and cloned into pcDNAI that had been digested with EcoRV, producing pcDNAI-A-PST. As a control vector, pcDNAI-A containing only the signal peptide and protein A cDNAs was similarly constructed.

After confirming the correct orientation by nucleotide sequencing, pcDNAI-A-PST and pcDNAI-A were separately transfected into COS-1 cells using Lipofectamine (11). After 62 h of transfection, the protein A-PST fusion protein secreted into the culture medium was adsorbed to IgG-Sepharose 6FF (Pharmacia Biotech Inc.) essentially as described (21). The resin was collected by centrifugation, washed nine times with 20 mM Tris-HCl, pH 7.5, containing 7.5 mM CaCl2 and 0.05% Tween 20, and finally suspended in the equal volume of a solution containing 100 mM NaCl, 90 mM sodium cacodylate buffer, pH 7.0, containing 20 mM MnCl2, 1 Triton CF-54, 2.4 nmol of CMP-[14C]NeuNaC. To this substrate solution, 50 μl of the enzyme solution prepared above was added, and the reaction mixture was incubated at 37°C for 4 h or 24 h. At the end of incubation, the reaction mixture was centrifuged. To 20 μl of this supernatant, 20 μl of the sample buffer for SDS-polyacrylamide gel electrophoresis (24) was added, and heated at 85°C for 3 min. The rest of the sample was kept at −70°C until use. The product in the sample buffer was directly subjected to SDS-polyacrylamide gel electrophoresis (7.5% acrylamide gel), and the incorporated sialic acids were visualized by fluorography. For fluorography, the gel was first fixed in 40% methanol for 30 min and then soaked in dimethyl sulfoxide (Me2SO) for 15 min twice. The treated gel was then soaked in Me2SO containing 2,5-diphenyloxazole (18.5 g in 88 ml of Me2SO) according to the procedure as described (25). Rat α,α-acid glycoprotein, fetuin, and human α,α-acid glycoprotein used as substrates were purchased from Sigma.

For linkage analysis of incorporated sialic acid, 90% of ethanol was added to the reaction mixture and the glycoproteins were recovered by centrifugation. After washing in 90% ethanol one more time, the sample was digested with NANase I (0.17 unit/ml), NANase II (5 units/ml), or NANase III (1.7 units/ml), purchased from Glyko, Inc. (Navato, CA), at 37°C for 19 h according to the protocol provided by the supplier. NANase I, II, and III specifically cleave α-2,3-linked sialic acid, α-2,3- and α-2,6-linked sialic acids, and α-2,3-, α-2,6- and α-2,8-linked sialic acids, respectively. Similarly, the reaction mixture was digested for 36 h with N-glycanase according to the manufacturer’s protocol (Genzyme, Cambridge, MA) or for 24 h with endo neuraminidase (endo-N) according to the procedures described (26). These digested materials were then subjected to SDS-polyacrylamide gel electrophoresis followed by fluorography using the same procedure as described above.

In order to determine the requirement of sialic acid residues in the acceptor glycoproteins, fetuin was digested with NANase I, NANase II, NANase III, or N-glycanase. After the incubation, ethanol (90%, the final concentration) was added to the reaction mixture and the solution was mixed well. The digested substrate was recovered by centrifugation and washed again with 90% ethanol. The recovered desialylated or de-N-glycosylated fetuin was used as acceptors for incorporation of radioactive sialic acid residues under the same conditions described above.

RESULTS AND DISCUSSION

PST Does Not Necessarily Require N-CAM for the Expression of PSA in Vivo—Previously, it has been shown that PST directs the expression of PSA when expressed together with N-CAM in COS-1 cells (11, 12). In order to determine if N-CAM is an absolute requirement for PST to form PSA, COS-1 cells were transiently transfected with PST cDNA alone in the absence of N-CAM cDNA. The results show that the transfected cells became positive for the immunostaining by M6703, 12E3, and 735 monoclonal antibodies (Fig. 1, A–C). Untransfected COS-1 cells were negative for the staining by any of these antibodies (data not shown; see also Ref. 11). M6703 antibody was shown to react with oligosialic acid with 2–3 residues of α-2,8-linked sialic acid (15), while 12E3 and 735 were shown to react with polysialic acid with 6 or more (for 12E3) residues of α-2,8-linked sialic acid and 8 or more (for 735) residues of α-2,8-linked sialic acid (18, 19), respectively. The results shown in Fig. 1 demonstrate that the transfected cells were most strongly stained by 12E3. The results thus indicate that the majority of PSA formed by PST were those containing 6 or more α-2,8-linked sialic acid residues. The absence of N-CAM was confirmed by immunofluorescent staining of COS-1 cells by anti-N-CAM antibody (Fig. 1D).

Similarly, we established HeLa cells stably expressing PSA...
in the absence of N-CAM. Immunofluorescent staining of the transfected HeLa cells confirmed that N-CAM was absent in the transfected HeLa cells (data not shown). However, the transfected HeLa cells expressed PSA, which was detected by the 735 antibody, and this expression was abolished by the treatment of endo-N, which specifically cleaves PSA (Fig. 2). These results indicate that PSA can be formed by PST in the absence of N-CAM.

PST Catalyzes in Vitro Synthesis of PSA—The above results indicate that PST can directly add PSA on carbohydrate moieties attached to proteins other than N-CAM. In order to test this hypothesis, a putative catalytic domain of PST was expressed as a protein fused with an IgG-binding domain of protein A preceded by a signal peptide sequence. The cDNA encoding this chimeric protein was cloned into pcDNA1, and the resultant vector was inserted into COS-1 cells. The fusion protein, which was secreted into the culture medium, was absorbed to IgG-Sepharose and then incubated with sialylated glycoproteins together with the donor substrate CMP-[14C]NeuNAc.

As shown in Fig. 3, the chimeric PST protein directed the formation of a broad and high molecular weight bands (mass - 100-170 kDa) when fetuin was incubated for 24 h. Although small amounts of broad and high molecular weight bands were produced from rat and human α1-acid glycoproteins, the majority of the radioactivity incorporated into α1-acid glycoproteins migrated close to the position where untreated glycoproteins migrated (mass - 48 and - 44 kDa, respectively). The products from fetuin and human α1-acid glycoprotein also contained a band with a high molecular mass in excess of 200 kDa, which did not enter into the separation gel. These bands most likely represent insoluble aggregates of sialylated glycoproteins since they were present when a large amount of the products were analyzed but absent when only a small amount of the products were analyzed, shown in Fig. 4.

In order to characterize the products formed by PST, they were digested with various enzymes and then subjected to SDS-polyacrylamide gel electrophoresis. The products were not susceptible to NANase II, which cleaves both α-2,3-linked and α-2,6-linked sialic acid (NANase II in Fig. 4). The products were, however, susceptible to NANase III, which cleaves all of α-2,3, α-2,6- and α-2,8-linked sialic acids (NANase III in Fig. 4). They were also susceptible to endo-N and N-glycanase (Fig. 4). These results establish that the broad and high molecular weight bands represent glycoproteins containing polysialic acid side chains in N-glycans. Similarly, the sialic acid residues incorporated into human α1-acid glycoprotein were removed by NANase III or N-glycanase (Fig. 4), indicating they are also attached through an α-2,8-linkage(s).

The product from fetuin after the treatment of endo-N still migrated as a larger molecule (mass - 90 kDa) than the untreated glycoprotein (Fig. 4). This is most likely due to the fact that endo-N can not cleave polysialic acid chains that are shorter than those consisting of 5 or 6 sialic acid residues (26, 27). The product after endo-N treatment most likely represents fetuin containing polysialic acid chain(s) consisting of 6 or fewer α-2,8-sialic acid residues in a side chain. Fetuin contains both N- and O-linked oligosaccharides (28, 29). Since N-glycanase treatment removed almost all of the incorporated sialic acid (Fig. 4), the majority of polysialylation took place in N-glycans.

PST Adds α-2,8-Linked Polysialic Acid on α-2,3-Linked Sialic Acid Attached to N-Glycans—In order to determine the requirement as the acceptor for PST, fetuin was treated with various neuraminidases and N-glycanase and those treated...
samples were used as acceptors. As shown in Fig. 5, once α-2,3-linked sialic acid residues were removed from fetuin, PST did not add a sialic acid residue regardless of the absence or presence of α-2,6-linked sialic acid residues (leftmost two lanes). No incorporation was detected after the removal of N-glycans (rightmost lane). These results confirmed the previous findings that PSA is attached to an α-2,3-linked sialic acid in a glycoconjugate template (3).

It is noteworthy that fetuin is the best acceptor among glycoproteins tested. Fetuin contains mainly tri-antennary N-glycans (29), while rat and human α1-acid glycoproteins mainly contain bi-antennary and tetra-antennary N-glycans (30), respectively. The present findings are consistent with the report that PSA containing N-glycans isolated from chicken and bovine fetal brains were mainly composed of tri-antennary oligosaccharides (31). Further studies are, however, required to determine whether or not PST preferentially adds sialic acid residues on tri-antennary N-glycans.

The present study established an in vitro assay system for PSA synthesis, allowing us to conclude that PST alone can add the first α-2,8-linked sialic acid to a precursor containing α-2,3-linked sialic acid and then add multiple α-2,8-linked sialic acid residues to the acceptor containing NeuNAcα2→8NeuNAcα2→3Gal→R structure, yielding PSA. It is unlikely, but possible, that the specificity of PST might be modified because a protein A is fused with the catalytic domain of PST. The results consistent with our conclusion of the results obtained in the present study, these account of the results obtained in the present study, these PST (11), ST8SiaIII (10), and GD3 synthase (20, 32). Taking

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REFERENCES
1. Edelman, G. M. (1985) Annu. Rev. Biochem. 54, 135–169
2. Rutlsuahser, U., Acheson, A., Hall, A. K., Mann, D. M., and Sunshine, J. (1988) Science 240, 53–57
3. Fine, J. (1992) J. Biol. Chem. 267, 11966–11970
4. Hoffman, S., and Edelman, G. M. (1983) Proc. Natl. Acad. Sci. U.S.A. 80, 5762–5766
5. Sadoul, R., Hirn, M., Deagostini-Bazin, H., Rougon, G., and Goridis, C. (1983) Proc. Natl. Acad. Sci. U.S.A. 80, 2555–2559
6. Bixby, J. L., Pratt, R. S., Lilien, J., and Reichardt, L. F. (1987) Proc. Natl. Acad. Sci. U.S.A. 84, 347–349
7. Kadmon, G., Kowitz, A., Altegoeg, P., and Schachner, M. (1990) J. Cell Biol. 110, 209–218
8. McCoy, R. D., Vinr, E. R., and Troy, F. A. (1985) J. Biol. Chem. 260, 12695–12699
9. Kitajima, K., Kitajima, K., Inoue, S., Inoue, Y., and Troy, F. A. (1994) J. Biol. Chem. 269, 10330–10340
10. Yoshida, Y., Kojima, N., Kurosawa, N., Hamamoto, T., and Tsuji, S. (1995) J. Biol. Chem. 270, 14628–14633
11. Nakayama, J., Fukuda, M. N., Freedte, B., Ranscht, B., and Fukuda, M. (1995) Proc. Natl. Acad. Sci. U.S.A. 92, 7031–7035
12. Eckhardt, M., Mühlenhoff, M., Bethe, A., Koopmann, J., Frosch, M., and Gerardy-Schahn, R. (1995) Nature 373, 715–718
13. Scheidegger, E. P., Sternber, L. R., Roth, J., and Lowe, J. B. (1995) J. Biol. Chem. 270, 22685–22688
14. Livingstone, B. D., and Paulson, J. C. (1993) J. Biol. Chem. 268, 11504–11507
15. Nakayama, J., Katsuyaama, T., Sugiyama, E., and Hirabayashi, Y. (1993) J. Histochem. Cytochem. 41, 1563–1572
16. Seki, T. & Arai, Y. (1991) Anat. Embryol. 184, 395–401
17. Frosch, M., Göggen, I., Boulnas, G. J., Timm, K. M., and Bitter-Suermann, D. (1985) Proc. Natl. Acad. Sci. U.S.A. 82, 1194–1198
18. Sato, C., Kitajima, K., Inoue, S., Seki, T., Troy, F. A., and Inoue, Y. (1995) J. Biol. Chem. 270, 18923–18928
19. Härinen, J., Bitter-Suermann, D., and Finne, J. (1989) Mol. Immunol. 26, 523–529
20. Sasaki, K., Kurata, K., Kojima, N., Kurosawa, N., Ohba, S., Hanai, N., Tsuji, S., and Nishi, T. (1994) J. Biol. Chem. 269, 15950–15956
21. Kukowska-Latallo, J. F., Larsen, R. D., Nair, R. P., and Lowe, J. B. (1990) Genes & Dev. 4, 1288–1303
22. Bierhuizen, M. F. A., and Fukuda, M. (1992) Proc. Natl. Acad. Sci. U.S.A. 89, 9326–9330
23. Kojima, N., Yoshida, Y., Kurosawa, N., Lee, Y.-C., and Tsuji, S. (1995) FEBS Lett. 350, 1–4
24. Laemmli, U. K. (1970) Nature 227, 680–685
25. Bonner, W. M., and Laskey, R. A. (1974) Eur. J. Biochem. 46, 83–88
26. Hallenbeck, P. C., Vinr, E. R., Yu, F., Basler, B., and Troy, F. A. (1987) J. Biol. Chem. 262, 3553–3561
27. Finne, J., and Mäkelä, P. H. (1985) J. Biol. Chem. 260, 1265–1270
28. Spiro, R. G., and Bhoyroo, V. D. (1974) J. Biol. Chem. 249, 5704–5717
29. Green, E. D., Adelt, G., Baenzinger, J. U., Wilson, S., and Van Halbeek, H. (1985) J. Biol. Chem. 260, 2269–22698
30. Yoshida, H., Matsumoto, A., Mizuochi, T., Kawasaki, T., and Kobata, A. (1994) J. Biol. Chem. 269, 8476–8484
31. Kojima, K., Kuroda, H., and Nara, K. (1995) J. Immunol. 155, 2555–2559
32. Nara, K., Watanabe, M., Maruyama, K., Kasahara, K., Nagai, Y., and Sanai, Y. (1994) Proc. Natl. Acad. Sci. U.S.A. 91, 7952–7956
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