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Down-regulation of the expression of β1,4-galactosyltransferase V promotes integrin β1 maturation

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Abstract

In previous study, we have shown that β1,4-galactosyltransferase V (GalT V) functions as a positive growth regulator in glioma. Here, we reported that down-regulation of the expression of GalT V in SHG44 cells by transfection with antisense cDNA specifically up-regulated the expression of cell surface integrin β1 without the change of its mRNA, and with integrin β1 125 kDa mature form increased and 105 kDa precursor form decreased. It is well known that the *N*-glycans of integrins modulate the location and functions of integrins. The SHG44 cells transfected with antisense cDNA of GalT V demonstrated decreased Golgi localization of integrin β1, strengthened the interaction between integrin α5 and β1 subunit, and enhanced the adhesion ability to fibronectin and the level of focal adhesion kinase phosphorylation. Our results suggested that the down-regulation of the expression of GalT V could promote the expression of cell surface integrin β1 and subsequently inhibit glioma malignant phenotype.

Keywords: β1,4-Galactosyltransferase V; Integrin β1; Protein maturation; Adhesion; FAK

N-Acetylglucosaminyl transferase V (GnT V) and β1,4-galactosyltransferase V (GalT V) cooperatively synthesized the Galβ1 → 4GlcNAcβ1 → 6Man structure. The increased expression of Galβ1 → 4GlcNAcβ1 → 6Man outer chain on *N*-glycan is a common feature for malignant transformation compared with normal phenotype cells [1,2]. The aberrant cell surface glycosylation patterns lead to alterations in cell–cell and cell–matrix interactions by affecting the function of adhesion molecules such as E-cadherin, integrins, and CD44 [3–5]. When alterations in the glycosylation patterns of the glioma associated integrin, α3β1, were introduced by transfection of the α2,6-sialyltransferase gene into a malignant glioma cell line, inhibition of invasion was observed in vitro [6].

Many factors including GnT V can affect the maturation of integrin β1 and its transport to the cell surface [7,8]. Changes in the expression of cell surface integrins may be important in cancers, impacting on various aspects of cancer metastasis [9]. Integrin β1 is a transmembrane glycoprotein. After synthesized as an 87 kDa polypeptide, it undergoes glycosylation in the endoplasmic reticulum (ER), and then in the Golgi apparatus. The incompletely glycosylated form of integrin β1 has a mass of 105 kDa and is referred to as p105. The completely glycosylated form of integrin β1 has a mass of 125 kDa (p125) [10]. p105 is not found at the cell surface, thereby unable to play a part in cell adhesion or cell signaling [10,11].

GalT V is one of the β1,4-galactosyltransferase (β1,4-GalTs) family members [12]. The gene expression levels of GalT V were strongly correlated with those of GnT V whose activity was shown to increase by malignant transformation [13]. It has been reported that apparent GalT V activities in MTAg, the most malignant transformation

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of NIH3T3, do not change significantly but the gene expression patterns of β1,4-GalTs change drastically, markedly increasing the mRNA level of GalT V [14]. Our previous study showed that increased GalT V transcript which appeared in the process of astrocytoma progressed with the highest level in grade IV [15]. This suggests that the malignant degree of astrocytoma may be correlated with the expression of GalT V and GalT V may affect the malignant phenotype [16,17]. The present study shows that down-regulation of the expression of GalT V could increase the expression of cell surface integrin β1 and subsequently enhance cell adhesion ability to FN and the tyrosine phosphorylation level of FAK in SHG44 cells.

Experimental procedures

Materials. Astrocytoma cell lines SHG were obtained from the affiliated Huashan Hospital of Fudan University. Anti-mouse IgG, anti-rabbit IgG, FN, Dulbecco's modified Eagle's medium (DMEM), and crystal violet were all from Sigma. HRP, rhodamine or FITC-conjugated goat anti-mouse secondary antibody were from Santa Cruz Biotechnology. Monoclonal antibody against integrin β1 was obtained from BD Pharmingen. Endoglycosidase H (Endo H) was purchased from Roche Applied Science. Transfection Reagent Sofast™ was purchased from Sunma Biotechnology (Xiamen, China). The pcDNA3.0 vector was from Invitrogen. The plasmids sense cDNA of GalT V (GalT V-S) and antisense cDNA of GalT V (GalT V-AS) have been previously described [16].

Cell culture and transfection. The SHG44 cells stably transfected with GalT V-AS and pcDNA3.0 vector plasmids were named as GalT V-AS/SHG44 and vector/SHG44, respectively. Cells were routinely cultured in DMEM. Transient transfections were performed with Sofast™ reagent according to manufacturer's instruction.

Detection of integrin on cell surface with fluorescence activated cell spectra. Fluorescence activated cell spectra (FACS) has been described previously [7]. Quantitative data were expressed at the mean fluorescence intensity in three independent and reproducible experiments.

Detection of the mRNA expression of integrins with real-time RT-PCR. Quantitative real-time RT-PCR experiments were performed as described previously [7]. The primers used for integrin β1 forward: 5′-AACTTGATCCCTAAGTCAGCAGTAG-3′, reverse: 5′-ATCAGCAGTAATGCAAGGCC-3′; for integrin α5 forward: 5′-ACCAAGGCCCCAGCTCCATTAG-3′, reverse: 5′-GCCTCACACTGCAGGCTAAATG-3′; for β-actin forward: 5′-GATATCGCCGCGCTCGTCGTCGAC-3′, reverse: 5′-CAGGAAGGAAGGCTGGAAGAGTGC-3′.

Co-immunoprecipitation and Western blot analysis. Co-immunoprecipitation of integrin α5 and β1 subunit experiments and Western blot experiments were performed as described previously [7]. Immunocomplexes were subjected to Western blot with anti-integrin β1 or α5 antibody.

Endoglycosidase digestion. Cells extracts were subjected to Endo H digestion as described previously [18]. Then the samples were subjected to immunoblot analysis with anti-integrin β1 antibody.

Subcellular localization of β1 integrin in SHG44 cells. Cells were fixed with 4% formaldehyde and then permeabilized with 0.5% Triton X-100. After blocked with 1% BSA, cells were stained with anti-β1 integrin mouse monoclonal antibody, followed by incubation with a mixture of rhodamine-conjugated anti-mouse IgG. Then the cells were stained with C6-NBD, a special dye labeling Golgi apparatus, according to manufacturer's instruction. After washing with PBS, the coverslips were photographed by using fluorescence microscopy.

Cell adhesion assays. Cell adhesion experiments were performed as described previously [19]. The data were expressed as means of triplicate wells ± SD.

Immunoprecipitation of focal adhesion kinase. Immunoprecipitation of focal adhesion kinase (FAK) experiment was performed as described previously [7]. Immunocomplexes were subjected to Western blot with anti-PY20 or anti-FAK antibody.

Results

Down-regulation of GalT V promotes the expression of cell surface integrin β1

The expression of integrin β1 was compared by immunoblot analysis in HeLa cells after transient transfection with GalT V-S, GalT V-AS or pcDNA3.0 vectors. We found that the expression of p125 and p105 both increased in the cells transfection with GalT V-AS (Fig. 1A), which showed enhanced expression of cell surface integrin β1 by FACS analysis (Fig. 1B).

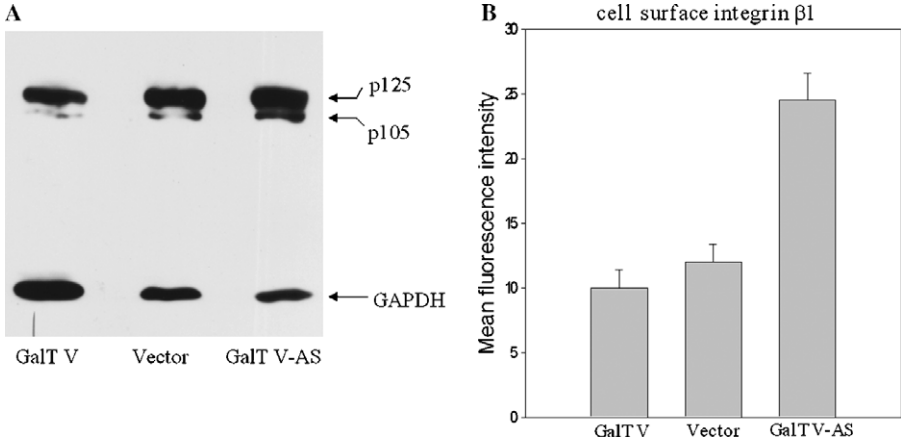

Fig. 1. Down-regulation of the expression of GalT V promotes the expression of cell surface integrin β1 subunit. (A) Equal amounts of cellular protein form HeLa cells transiently transfected with GalT V, GalT V-AS or pcDNA3.0 vectors were subjected to immunoblot analysis with anti-integrin β1 and anti-GAPDH antibodies. (B) HeLa cells transiently transfection with GalT V, GalT V-AS or pcDNA3.0 vectors were subjected to analysis with FACS of cell surface integrin β1. Each bar represents the mean (±SD) of triplicate determinations.

Analysis of the expression on cell surface and transcription of integrin α5 and β1 in GalT V-AS/SHG44 and vector/SHG44 cells

We further investigated the expression of integrin β1 in GalT V-AS/SHG44 and vector/SHG44 by means of real-time RT-PCR. As shown in Fig. 2A, the mRNA expression level of integrin β1 was nearly identical between the GalT V-AS/SHG44 cells and vector/SHG44 cells. However, the GalT V-AS/SHG44 cells demonstrated increased the cell surface integrin β1 than vector/SHG44 cells by means of FACS analysis (Fig. 2B). In order to elucidate the mechanism of the altered expression of integrin subunits, we further investigated the mRNA expression level of integrin α5. Though the mRNA expression level of integrin α5 subunit was increased to 4.81 times in GalT V-AS/SHG44 cells as compared to the control (Fig. 2C), the expression of cell surface integrin α5 was unchanged by FACS analysis (Fig. 2D). These results suggested that GalT V-AS could specifically up-regulate the expression of cell surface β1 integrin.

Increased p125 and decreased p105 of β1 integrin in GalT V-AS/SHG44 cells

We performed Western blot to compare the protein level of integrin β1 in GalT V-AS/SHG44 cells with those in vector/SHG44 cells. As shown in Fig. 3A, there were no significant differences in the total amount of integrin β1 in GalT V-AS/SHG44 and vector/SHG44 cells; however, a subtle alteration was noted in the ratio of precursor to mature integrin β1 isoform. Compared with vector/SHG44, GalT V-AS/SHG44 cells demonstrated increased p125 and significantly decreased p105 of integrin β1. Endo H could dissociate N-linked mannose-rich glycans which become Endo H-resistant after modification by glycosyltransferase in the Golgi apparatus. To test the validity of Endo H, we then treated cell extracts from SHG44 with Endo H or vehicle. As expected, p105 was eliminated by Endo H and replaced with a new band of about 85 kDa (Fig. 3B). This molecular mass was consistent with the expected migration of the unglycosylated core peptide of integrin β1 [20]. And p125 was not modified by Endo H. In order to confirm that

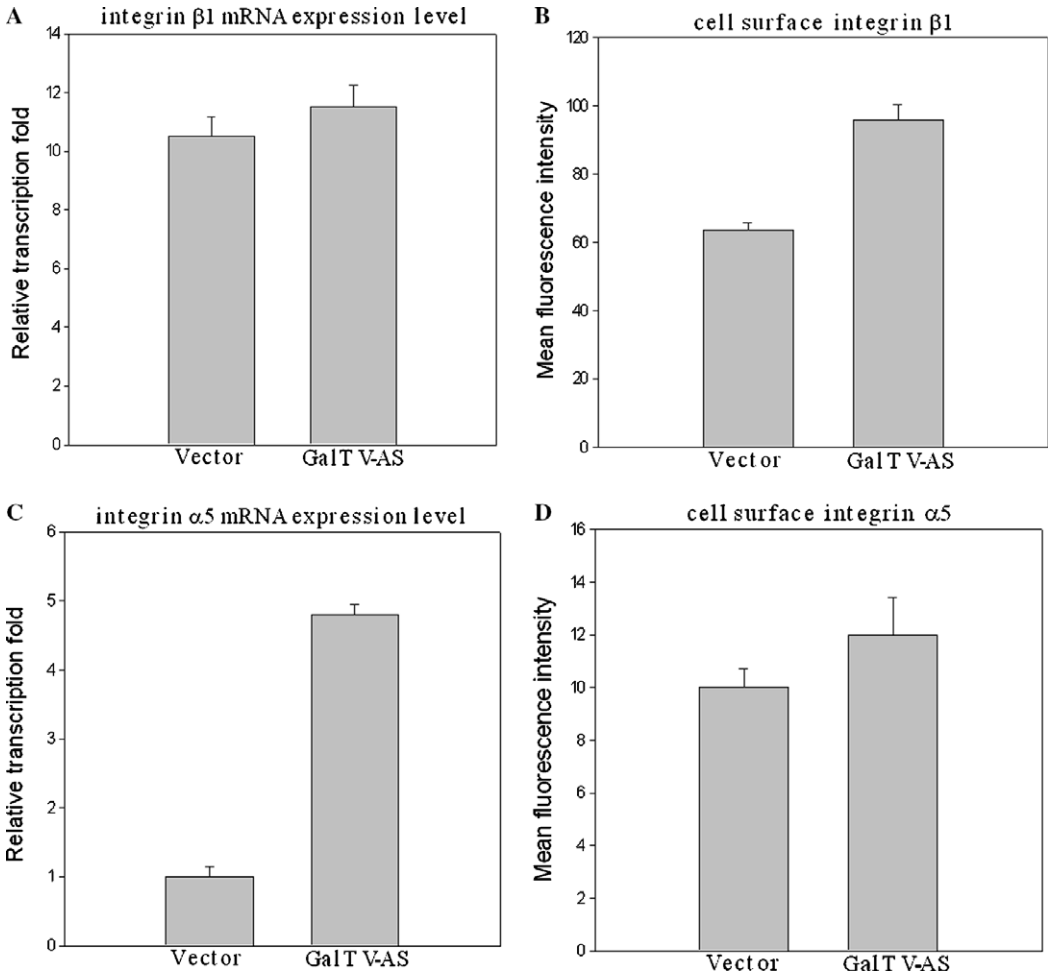

Fig. 2. Analysis of the cell surface expression and transcription of integrin β1 and α5 subunit in GalT V-AS/SHG44 and vector/SHG44 cells. (A) Real-time RT-PCR analysis of the transcription of integrin β1 in GalT V-AS/SHG44 and vector/SHG44 cells. (B) GalT V-AS/SHG44 and vector/SHG44 cells were subjected to analysis with FACS of cell surface integrin β1 subunit. (C) Real-time RT-PCR analysis of the transcription of integrin α5 in GalT V-AS/SHG44 and vector/SHG44 cells. (D) GalT V-AS/SHG44 and vector/SHG44 cells were subjected to analysis with FACS of cell surface integrin α5. Each bar represents the mean (±SD) of triplicate determinations.

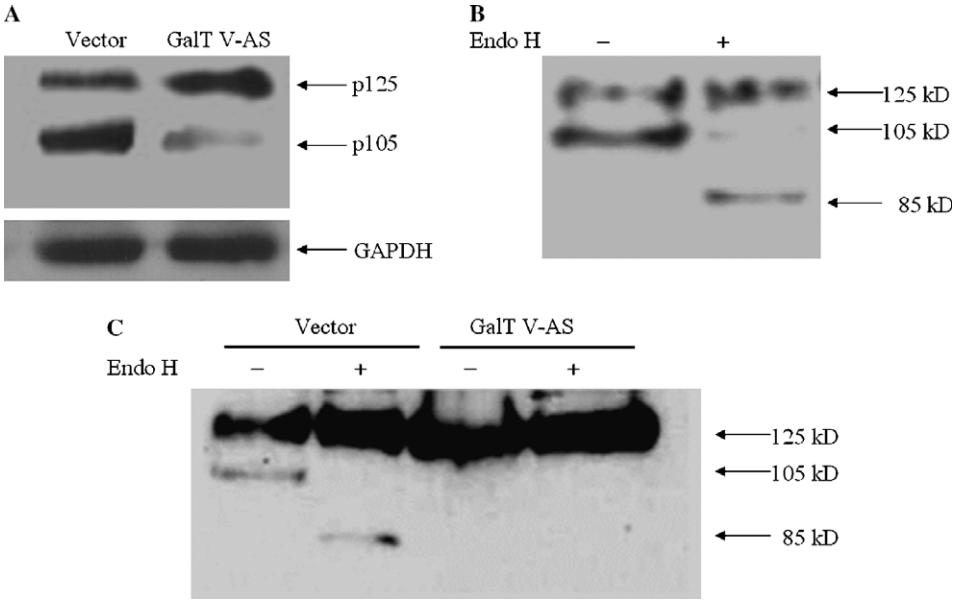

Fig. 3. Down-regulation of the expression of GalT V increases p125 and decreases p105. (A) Equal amounts of cellular protein from GalT V-AS/SHG44 and vector/SHG44 cells were subjected to immunoblot analysis with anti-integrin β1 (upper panel) or anti-GAPDH (lower panel) antibodies. Both a 105 kDa precursor form and a 125 kDa mature form were noted. (B) Equal amounts of cellular protein from whole cell extracts of SHG44 cells were treated with Endo H (+) or vehicle (−) for 24 h at 37 °C. The samples were subjected to immunoblot analysis with anti-integrin β1 antibody. (C) Equal amounts of cellular protein from SHG44 cells transiently transfection with GalT V-AS (+) or pcDNA3.0 vectors (−) were treated with Endo H (+) or vehicle (−) for 24 h at 37 °C. The samples were subjected to immunoblot analysis with anti-integrin β1 antibody.

down-regulation of the expression of GalT V could promote integrin β1 maturation, we transiently transfected SHG44 cells with GalT V-AS or pcDNA3.0 vectors followed by treatment with Endo H. The vector transient transfection was shown to two bands (Fig. 3C, lane 1) and an 85 kDa new band followed digestion by Endo H (Fig. 3C, lane 3). Compared with the control (Fig. 3C, lanes 1 and 3), p105 was extremely decreased (Fig. 3C, lane 2), and the new band about 85 kDa was absent (Fig. 3C, lane 4) in GalT V-AS transient transfection.

Integrin β1 was localized near the cell surface other than to Golgi apparatus in GalT V-AS/SHG44 cells

To elucidate the mechanism of down-regulation of the expression of GalT V may control maturation of integrin β1, we examined cellular localization of integrin β1 using immunofluorescence confocal microscopy analysis. The GalT V-AS/SHG44 and vector/SHG44 were stained for integrin β1 (in red) and C6-NBD (in green), a special dye labeling Golgi apparatus. The yellow color present in the merged image indicates colocalization between integrin β1 and Golgi apparatus. We found that integrin β1 was present in Golgi apparatus in vector/SHG44 cells (Fig. 4, upper panel); however, in GalT V-AS/SHG44 cells, integrin β1 was localized near the cell surface and formed small intracellular clusters (Fig. 4, lower panel, red arrow). These results suggested that the down-regulation of the expression of GalT V could promote trafficking of integrin β1 from Golgi apparatus to cell surface.

Down-regulation of GalT V strengthened the interaction between integrin β1 and α5, increased the cell adhesion ability to FN and the level of FAK phosphorylation

We want to further investigate whether down-regulation of GalT V could affect the function of integrin. As expected, GalT V-AS/SHG44 cells demonstrated that the interaction between integrin β1 and α5 subunits strengthened (Fig. 5A) and the cell adhesion ability to FN increased (Fig. 5B). FAK colocalized with integrin, and its phosphorylation state and tyrosine kinase activity were also regulated by binding of cells to ECM [21,22]. As shown in Fig. 5C, the tyrosine phosphorylation level of FAK was significantly enhanced in GalT V-AS/SHG44 cells.

Discussion

GnT V and its product, β1,6 GlcNAc branching on the cell surface glycans were positively correlated with the metastasis-potential of some malignancies. Cells transfected with GnT V cDNA were shown to decrease adhesion to FN and increase cell migration, and cell invasion through matrigel [23]. It has been reported that the expression of GnT V could regulate glycosylation and function of α5β1 integrins. Over-expression of GnT V in HT1080 cells was shown to reduce α5β1 integrins clustering, inhibit α5β1 integrin-mediated adhesion, and promote cell motility [19].

The gene expression level of GalT V but not other β1,4-GalTs was strongly correlated with those of GnT V whose activity was shown to increase by malignant transformation [13]. To explore if GalT V also can affect glycosylation

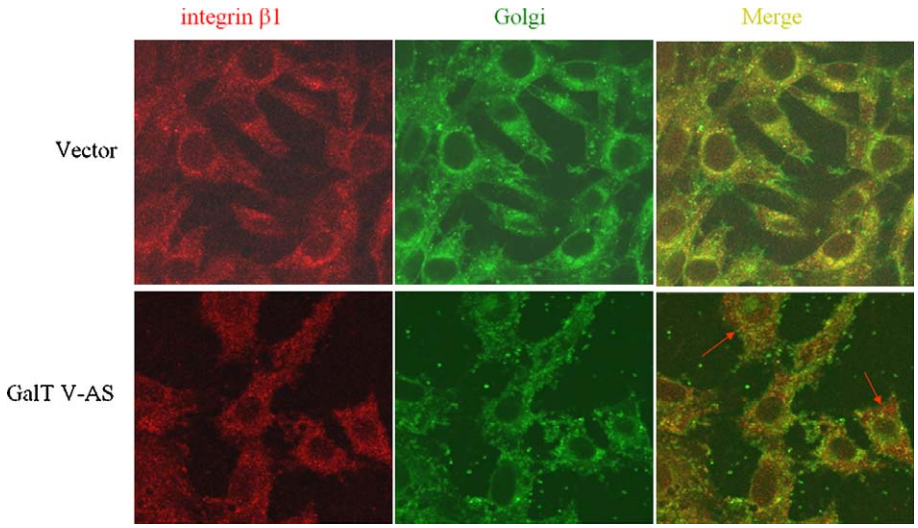

Fig. 4. Reduction in the GalT V expression affects the subcellular localization of integrin β1 subunit. After fixed and permeabilized, vector/SHG44 G44 cells (upper panel) or GalT V-AS/SH cells (lower panel) reacted with anti-integrin β1 mouse monoclonal antibody followed by incubation with rhodamine-conjugated goat anti-mouse IgG and C6-NBD, a special dye labeling Golgi apparatus. Images were captured and analyzed with a Zeiss confocal microscope (40× zoom). Integrin β1 is in red and the Golgi marker is in green. The yellow image is a red/green merge to show colocalization. In GalT V-AS/SHG44 cells, the integrin β1 was localized near the cell surface and formed small intracellular clusters (red arrow). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this paper.)

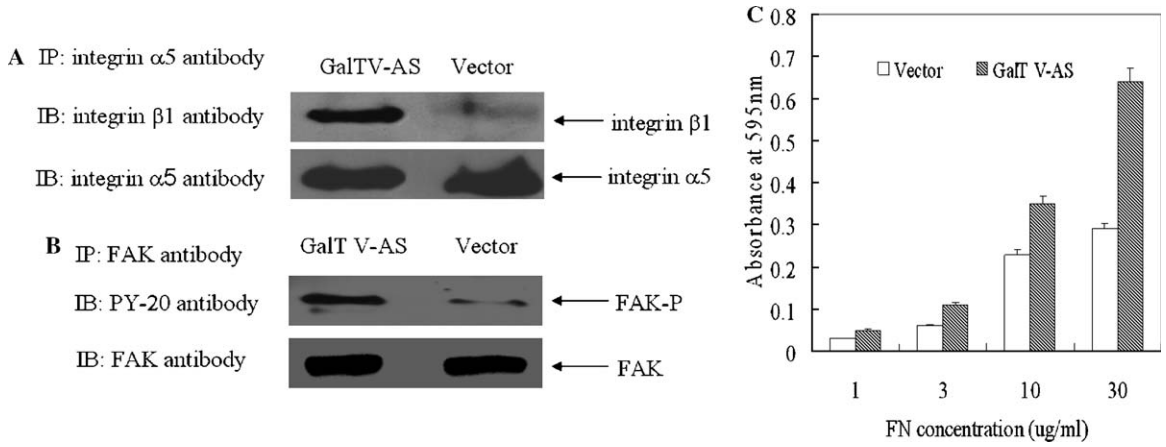

Fig. 5. Over-expression of GalT V strengthened the interaction between integrin β1 and α5 subunit, increased cell adhesion ability to FN and the level of FAK phosphorylation. (A) Endogenous integrin β1 subunit interacts with integrin α5 subunit in GalT V-AS/SHG44 cells and vector/SHG44 cells. Immunoprecipitation was performed with monoclonal anti-integrin α5 antibody. Co-immunoprecipitation protein was probed with indicated antibodies. (B) After incubation on FN (15 μg/ml) for 30 min, the cell lysates were immunoprecipitated by monoclonal anti-FAK antibody. The level of FAK and phosphor-FAK was detected by indicated antibodies. (C) GalT V-AS/SHG44 cells demonstrated more adhesion ability to fibronectin. GalT V-AS/SHG44 and vector/SHG44 cells (3×10^4) were applied to 96-well plates coated with poly-lysine (100 μg/ml), 1% BSA or increasing concentrations (1, 3, 10, and 30 μg/ml) of FN and incubated at 37 °C for 30 min. Adherent cells were crystal violet and absorbance of each well was determined at 595 nm. Each bar represents the mean (±SD) of triplicate determinations.

and expression of cell surface α5β1 integrin, SHG44 cells in which GalT V is highly expressed are stably transfected with GalT V-AS or pcDNA3.0 vectors. Increased cell surface β1 integrin is found in GalT V-AS/SHG44 cells. This result is confirmed in HeLa cells. Real-time RT-PCR analysis reveals the identical mRNA level of integrin β1 in GalT V-AS/SHG44 cells and vector/SHG44 cells, ruling out the possibility that the enhancement of cell surface integrin β1 by transfection with GalT V-AS is caused by increased the mRNA transcription level of integrin β1. The cell surface integrin α5 showed unchanged though the mRNA level of integrin α5 is increased to 4.81 times in GalT V-AS/SHG44 cells, suggesting the special role of GalT V in regulation on the expression of integrin β1.

To elucidate further the mechanism by which reduction in the expression of GalT V could increase the cell surface abundant of integrin β1, we performed Western blots to investigate the expression of the total cytoplasmic pools of β1 integrin. The HeLa cells transfection with GalT V-AS showed increased level of both p105 and p125.

However, stable transfection with GalT V-AS to SHG44 cells had no effect on the total level of β1 integrin in cellular lysates, but showed increased p125 and extremely reduced p105. These results suggested that the mechanisms of down-regulation of the expression of GalT V promoting the expression of cell surface integrin β1 were depended on the cell types as GnT V [7,19].

Because malignant degree of astrocytoma may be correlated with the expression of GalT V [16], we are interested in what happened in GalT V-AS/SHG44 cells. First, we want to identify that the p105 was the precursor form of integrin β1. As previously reported, the p105 and p125 bands represent integrin β1 isoforms that are in difference stages of maturation. After synthesized as an 87 kDa polypeptide, integrin β1 becomes partially glycosylated by ER glycosyltransferases, generating a high mannose, precursor form of 105 kDa. These N-linked mannose-rich glycans can be dissociated by Endo H. The integrin β1 precursor complex with a precursor form of an integrin α-subunit and then the heterodimer is transported to the Golgi apparatus, where the α and β subunits accept completely glycosylation by Golgi glycosyltransferases to generate the mature form with the Endo H-resistant N-linked glycans. The mature heterodimer is subsequently transported to the cell surface. In the Endo H digestion experiments, we confirmed that the new band about 85 kDa was the product of deglycosylated p105. This molecular mass was consistent with the unglycosylated integrin β1 core peptide. These results identified the p105 as the precursor form of integrin β1.

GalT V could effectively galactosylate the GlcNacβ1 → 6Man group of the highly branched *N*-glycans. So it is possible that down-regulation of the expression of GalT V by transfection with antisense cDNA could affect the homeostasis of Golgi apparatus and then alter the glycosylation or transportation of integrin β1 in Golgi. To verify the hypothesis, we investigate the subcellular localization of integrin β1. Compared with vector/SHG44 cells in which integrin β1 are localized in Golgi apparatus, the GalT V-AS/SHG44 cells demonstrate more integrin β1 localized near the cell surface and in small intracellular clusters. The results suggested that down-regulation of the expression of GalT V could directly or indirectly promote trafficking of integrin β1 from Golgi apparatus to cell surface. Consistent with these results, the GalT V-AS/SHG44 cells were shown to strengthen the interaction between integrin β1 and α5 subunit, increase cell adhesion to FN and the level of FAK phosphotyrosine as anticipated.

In our previous reports, the gene expression of GalT V increased in the process of astrocytoma progress and the over-expression of GalT V promoted the growth of astrocytoma cell line [15,16]. The GalT V-AS/SHG44 cells demonstrate the suppression of tumor development in experiment animals [17]. On the basis of these results, GalT V could represent a novel target in glioma therapy. But it has been reported that the expression of α5β1 integrin was up-regulated in glioma [24]. So there may be other molecular(s) involved in the process that GalT V knockdown suppresses the glioma development. We have found that a 61 kDa protein was galactosylated in astrocytoma but not in normal brain tissues [15] and more efforts are in proceeding.

Acknowledgments

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References

[1] J.W. Dennis, S. Laferte, C. Waghorne, M.L. Breitman, R.S. Kerbel, Beta 1-6 branching of Asn-linked oligosaccharides is directly associated with metastasis, Science 236 (1987) 582–585.
[2] B. Fernandes, U. Sagman, M. Auger, M. Demetrio, J.W. Dennis, Beta 1-6 branched oligosaccharides as a marker of tumor progression in human breast and colon neoplasia, Cancer Res. 51 (1991) 718–723.
[3] S. Hakomori, Tumor malignancy defined by aberrant glycosylation and sphingo(glyco)lipid metabolism, Cancer Res. 56 (1996) 5309–5318.
[4] M. Asada, K. Furukawa, K. Segawa, T. Endo, A. Kobata, Increased expression of highly branched *N*-glycans at cell surface is correlated with the malignant phenotypes of mouse tumor cells, Cancer Res. 57 (1997) 1073–1080.
[5] M. Yoshimura, Y. Ihara, Y. Matsuzawa, N. Taniguchi, Aberrant glycosylation of E-cadherin enhances cell–cell binding to suppress metastasis, J. Biol. Chem. 271 (1996) 13811–13815.
[6] H. Yamamoto, Y. Kaneko, A. Rebbaa, E.G. Bremer, J.R. Moskal, alpha2,6-Sialyltransferase gene transfection into a human glioma cell line (U373 MG) results in decreased invasivity, J. Neurochem. 68 (1997) 2566–2576.
[7] H.B. Guo, I. Lee, B.T. Bryan, M. Pierce, Deletion of mouse embryo fibroblast *N*-acetylglucosaminyltransferase V stimulates alpha5beta1 integrin expression mediated by the protein kinase C signaling pathway, J. Biol. Chem. 280 (2005) 8332–8342.
[8] P. Guo, Y. Zhang, J.H. Zhao, L.Y. Wang, H.B. Guo, X.Y. Zhang, H.L. Chen, Regulation on the expression and N-glycosylation of integrins by *N*-acetylglucosaminyltransferase V, Biochem. Biophys. Res. Commun. 310 (2003) 619–626.
[9] G.J. Mizejewski, Role of integrins in cancer: survey of expression patterns, Proc. Soc. Exp. Biol. Med. 222 (1999) 124–138.
[10] S.K. Akiyama, K.M. Yamada, Biosynthesis and acquisition of biological activity of the fibronectin receptor, J. Biol. Chem. 262 (1987) 17536–17542.
[11] B. De Strooper, F. Van Leuven, G. Carmeliet, H. Van Den Berghe, J.J. Cassiman, Cultured human fibroblasts contain a large pool of precursor beta 1-integrin but lack an intracellular pool of mature subunit, Eur. J. Biochem. 199 (1991) 25–33.
[12] M. Amado, R. Almeida, T. Schwientek, H. Clausen, Identification and characterization of large galactosyltransferase gene families: galactosyltransferases for all functions, Biochim. Biophys. Acta 1473 (1999) 35–53.
[13] T. Sato, K. Shirane, M. Kido, K. Furukawa, Correlated gene expression between beta-1,4-galactosyltransferase V and *N*-acetylglucosaminyltransferase V in human cancer cell lines, Biochem. Biophys. Res. Commun. 276 (2000) 1019–1023.
[14] K. Shirane, T. Sato, K. Segawa, K. Furukawa, Involvement of beta-1,4-galactosyltransferase V in malignant transformation-associated changes in glycosylation, Biochem. Biophys. Res. Commun. 265 (1999) 434–438.

[15] S. Xu, X. Zhu, S. Zhang, S. Yin, L. Zhou, C. Chen, J. Gu, Over-expression of beta-1,4-galactosyltransferase I, II, and V in human astrocytoma, J. Cancer Res. Clin. Oncol. 127 (2001) 502–506.
[16] S. Xu, J. Gu, Over-expression of Beta-1,4-Galactosyltransferase V increases the growth of astrocytoma cell line, J. Exp. Clin. Cancer Res. 21 (2002) 187–192.
[17] J. Jiang, X. Chen, J. Shen, Y. Wei, T. Wu, Y. Yang, H. Wang, H. Zong, J. Yang, S. Zhang, J. Xie, X. Kong, W. Liu, J. Gu, beta 1,4-galactosyltransferase V functions as a positive growth regulator in glioma, J. Biol. Chem. (2006), M504489200, in press.
[18] A.M. Salicioni, A. Gaultier, C. Brownlee, M.K. Cheezum, S.L. Gonias, Low density lipoprotein receptor-related protein-1 promotes beta1 integrin maturation and transport to the cell surface, J. Biol. Chem. 279 (2004) 10005–10012.
[19] H.B. Guo, I. Lee, M. Kamar, S.K. Akiyama, M. Pierce, Aberrant N-glycosylation of beta1 integrin causes reduced alpha5beta1 integrin clustering and stimulates cell migration, Cancer Res. 62 (2002) 6837–6845.
[20] W.S. Argraves, S. Suzuki, H. Arai, K. Thompson, M.D. Pierschbacher, E. Ruoslahti, Amino acid sequence of the human fibronectin receptor, J. Cell Biol. 105 (1987) 1183–1190.
[21] E. Zamir, B. Geiger, Molecular complexity and dynamics of cell–matrix adhesions, J. Cell Sci. 114 (2001) 3583–3590.
[22] J.T. Parsons, Focal adhesion kinase: the first ten years, J. Cell Sci. 116 (2003) 1409–1416.
[23] M. Demetriou, I.R. Nabi, M. Coppolino, S. Dedhar, J.W. Dennis, Reduced contact-inhibition and substratum adhesion in epithelial cells expressing GlcNAc-transferase V, J. Cell Biol. 130 (1995) 383–392.
[24] Y. Fukushima, T. Ohnishi, N. Arita, T. Hayakawa, K. Sekiguchi, Integrin alpha3beta1-mediated interaction with laminin-5 stimulates adhesion, migration and invasion of malignant glioma cells, Int. J. Cancer 76 (1998) 63–72.