Characterization of Gas6, a Member of the Superfamily of G Domain-containing Proteins, as a Ligand for Rse and Axl*  

(Received for publication, October 12, 1995, and in revised form, December 27, 1995)  

Melanie R. Mark‡, JIan Chen‡, R. Glenn Hammonds§, Michael Sadick¶, and Paul J. Godowski††  
From the ‡Department of Molecular Biology, the §Department of Protein Chemistry, and ¶Department of Research Immunochernistry, Genentech, South San Francisco, California 94080  

Rse, Axl, and c-Mer comprise a family of cell adhesion molecule-related tyrosine kinase receptors. Human Gas6 was recently shown to act as a ligand for both human Rse (Godowski et al., 1995) and human Axl (Varnum et al., 1995). Gas6 contains an NH₂-terminal Gla domain followed by four epidermal growth factor-like repeats and tandem globular (G) domains. The G domains are related to those found in sex hormone-binding globulin and to those utilized by laminin and agrin for binding to the dystroglycan complex. A series of Gas6 variants were tested for their ability to bind to Rse and Axl. The Gla domain and epidermal growth factor-like repeats were not required for receptor binding, as deletion variants of Gas6 which lacked these domains bound to the extracellular domains of both Rse and Axl. A deletion variant of Gas6 containing just the G domain region was shown to activate Rse phosphorylation. These results provide evidence that G domains can act as signaling molecules by activating transmembrane receptor tyrosine kinases. Furthermore, they provide a structural link between the activation of cell adhesion receptors and the control of cell growth and differentiation by the G domain-containing superfamily of proteins.  

Specific signals that control the growth and differentiation of cells in developing and adult tissues often exert their effects by binding to and activating cell surface receptors containing an intrinsic tyrosine kinase activity. We recently reported the human and murine complementary DNA sequences of a receptor tyrosine kinase we termed Rse (Mark et al., 1994). A complementary DNA sequence encoding a protein identical to human Rse has also been reported (Tyro3, Lai and Lemke, 1994; Brt, Fujimoto and Yamamoto, 1994; Etk2, Biesecker et al., 1995).  

Rse has about 31% identity sequence with the receptor tyrosine kinases Axl (O'Bryan et al., 1993; Janssen et al., 1991) and c-Mer (Graham et al., 1994). The extracellular domains of the Rse/Axl/c-Mer family are composed of two immunoglobulin-like repeats followed by two fibronectin type III repeats. Together, these proteins define a class of receptor tyrosine kinases whose extracellular domains resemble neural cell adhesion molecules (reviewed by Rutishauser (1993) and Brummendorf and Rathjen (1993)).  

While Rse mRNA is expressed preferentially in the adult brain, it is expressed at lower levels in a number of tissues including kidney, ovary, and testis and in a variety of hematopoietic cell lines (Mark et al., 1994; Lai and Lemke, 1994). Axl and c-Mer are also widely expressed, but the highest levels of Axl and c-Mer mRNA are detected in the heart and skeletal muscle (Graham et al., 1995) and testis, ovary, prostate, lung, and kidney (Graham et al., 1994), respectively.  

Gas6 was initially identified as a product of a gene whose expression is increased in fibroblasts upon growth arrest (Manfioletti et al., 1993). We recently identified Gas6 as a ligand for human Rse (Godowski et al., 1995). Varnum et al. (1995) identified Gas6 as a ligand for human Axl. Gas6 has 46% amino acid identity to Protein S (PS), an abundant serum protein and a negative regulator of the coagulation cascade. Stitt et al. (1995) reported that PS, but not Gas6, was a ligand for Rse. However, those conclusions were based on the analysis of the interactions of human PS and bovine Gas6 with murine Rse. While human PS does indeed bind to murine Rse, we found that even high concentrations of human PS failed to activate human Rse (Godowski et al., 1995). These results have recently been confirmed by Ohashi et al. (1995). Thus, there are no published data indicating that PS is a physiologically relevant ligand for Rse.  

Gas6 contains 678 amino acids, which may be divided into five domains (Fig. 1A). The Gla domain is rich in γ-carboxyglutamic acid (Gla) residues. The corresponding Gla domain of PS mediates its Ca²⁺-dependent binding to negatively charged phospholipids (Dahliback et al., 1986; Hammond et al., 1987). The loop region, containing thrombin-sensitive cleavage sites in PS, but lacking such sites in Gas6, is followed by four epidermal growth factor (EGF)-like repeats. EGF-like repeats are found in a number of proteins which participate in diverse functions such as coagulation and fibrinolysis, cell adhesion, cell growth, and differentiation. These repeats are sufficient to bind to other receptor tyrosine kinases, and are believed to participate directly in protein-protein interactions (Cambell and Bark, 1993). The COOH-terminal regions of Gas6 and PS are homologous to the steroid hormone-binding globulin (SHBG) protein (Gershagen et al., 1987; Hammond et al., 1987) and contain tandem “globular” or G domains (G1 and G2; Joseph and Baker (1994)). G domains, first identified in laminin A chain, are present in a superfamily of proteins that include basement membrane proteins such as laminin A chain, agrin, merosin, and perlecan, as well as the Drosophila regulatory proteins Crumbs, Fat, and Slit (reviewed by Patthy and Nikolic (1993)).  

As a first step in understanding the contributions of the Gla, EGF-like repeats and G domains of Gas6 to interaction with...
Characterization of Gas6 as a Ligand for Rse and Axl

Rse and Axl, we analyzed the relative contributions of these domains in receptor binding and activation. Our data demonstrate the G domains of Gas6 are sufficient for receptor activation. These observations have implications for the mechanism by which other G domain-containing proteins may influence intercellular signaling.

EXPERIMENTAL PROCEDURES

Construction and Expression of Rse, Axl, Protein S, and Gas6 Derivatives—Human PS and Gas6 cDNAs were obtained by PCR using 1 μg of human fetal liver or fetal brain cDNA (Clontech) as template with Pfu DNA polymerase (Stratagene) as described (Mark et al., 1994). The coding sequences of the cDNA clones used for the expression of human PS and Gas6 were identical to those available in GenBank (accession numbers Y00692 and L13720, respectively). Plasmids pRKN.hPS and pRKN.hGas6 were used to express human PS and Gas6, respectively, in stably transfected human fetal kidney 293 cells. Transfected cells were washed free of serum-containing media 4 h after transfection, and refed with media containing 2 μg/ml vitamin K. Expression was verified by metabolic labeling of cultures with [35S]Cys and [35S]Met (Mark et al., 1992) and/or by Western blotting with a polyclonal anti-PS antisemur (Sigma) or a polyclonal anti-Gas6 antibody.2 NH2-terminally tagged versions of Gas6 or PS were constructed by linking the coding sequences for the gD signal sequence and epitope tag (Mark et al., 1994) to coding sequences immediately before the first amino acid of mature Gas6 (gD.Gas6; forward primer 5′-AGCTGGCTAGGCCGTGGTGC-CCGC-CC and PS (forward primer 5′-AGCTGCTGACGAGGAAATTTTATTCTTGAAGA, or amino acids 118 (gD.Gas6118-C; forward primer 5′-GAGTCGGATCCGACAGAGACTGAGTTCC-9 and 279 (gD.Gas6279-C; forward primer 5′-GCCGTGAGACAGCAGGCGGCCTCCAC). The cDNA encoding P2′G′-gD was constructed in a two-step PCR reaction (Mark et al., 1994) to join the coding sequences of amino acids 1–283 of PS to SHBG homology regions (amino acids 279–678) of Gas6.gD. The P2′G′-gD expression vector was constructed in a similar fashion by using coding sequences for amino acids 1–278 of Gas6 to those encoding amino acids 284–676 of PS.gD. The fusion junctions of P2′G′-gD and G2′G′-gD are coding strand 5′-GAGAGTTGCTGAGGACATCTAC-5′ and 5′-GGACACGGAAGGATTGTTTGGAC-9. The reverse primer for gD.Gas6279-C and gD.PS were 5′-GTCGGATCCGACAGAGACTGAGTTCC-9 and 5′-GTCGGATCCGACAGAGACTGAGTTCC-9, respectively. Gas6.gD was constructed by fusing the coding sequences of Gas6 to the COOH-terminal gD tag used for Rse.gD through an Nhel site, which was added by PCR using the primers 5′-ATGGGATCCAGGTGCTG and 5′-CATCTGTAGCAGTGGCCGCTCCAC. The cDNA encoding P2′G′-gD was constructed in a two-step PCR reaction (Mark et al., 1994) to join the coding sequences of amino acids 1–283 of PS to SHBG homology regions (amino acids 279–678) of Gas6.gD. The P2′G′-gD expression vector was constructed in a similar fashion by using coding sequences for amino acids 1–278 of Gas6 to those encoding amino acids 284–676 of PS.gD. The fusion junctions of P2′G′-gD and G2′G′-gD are coding strand 5′-GAGAGTTGCTGAGGACATCTAC-5′ and 5′-GGACACGGAAGGATTGTTTGGAC-9, respectively, where the shill indicates the fusion junction. Rse-Fc (Godowski et al., 1995) and Axl-Fc3 were purified as described (Godowski et al., 1995).

Protein Quantification—The concentration of epitope-tagged Gas6 and Gas6.gD was determined by analyzing an anti-gD monoclonal antibody (SB6; Genentech, Inc., South San Francisco, CA) as a coat/capture antibody. Samples putatively containing epitope-tagged proteins were added to SB6-coated and bovine serum albumin-blocked polystyrene immunoplates (Nunc, Roskilde, Denmark). Following a 2-h incubation at room temperature, biotin-conjugated gD2B (a 6-aminocaproic acid derived from the gD sequence; Genentech, Inc.) was added to each well and allowed to compete with the sample for the SB6 coat for an additional 50 min. After washing the plate 10 times, the wells were incubated with peroxidase-conjugated streptavidin (Zymed Laboratories, Inc., South San Francisco, CA) for 30 min at room temperature. Plates were again washed and TMB Microplate Peroxidase substrate (Kirkegaard & Perry Laboratories, Inc., Gaithersburg, MD) was added to each well. Color was allowed to develop for 10 min, and the reaction was quenched with 100 μl of H2PO4. Absorbance was measured at 450 nm using a Vmax kinetic microplate reader (Molecular Devices). Concentrations of gD-tagged Gas6 or variants were calculated from a standard curve using a purified standard (gD11/Brp120) in a range of 1.56–100 nM.

Binding Assays—For binding to Rse-Fc or Axl-Fc fusion proteins, conditioned media containing 5–10 nm Gas6 or variant protein were preloaded with pansorbin (Calbiochem) for 30 min at room temperature, then incubated with 5 μg of the Rse-Fc fusion protein for 4 h at 4 °C. Fusion proteins were immunoprecipitated with 20 μl of Pansorbin, and the complexes were collected by centrifugation at 14,000 × g for 1 min and then washed three times with PBS containing 0.1% Triton X-100. Precipitates were analyzed by SDS-PAGE under reducing conditions. Western blots of the SDS-PAGE gels were probed with antibody 5B6 as described (Mark et al., 1994).

Protein interaction analysis using BIAcore™ instruments were performed on research grade BIAcore CM5 sensor chips. Running buffer was PBS (10 mM sodium phosphate, pH 7.4, 150 mM sodium chloride) with 0.05% Tween 20. The sensor chip was activated by injection of 20 μl of 1:1 mixture of N-ε-ethyl-N-3-(3-dimethylaminopropyl)carbodiimide hydrochloride and N-hydroxysuccinimide at 5 μl/min flow rate. 20 μl of Rse-Fc at 20 ng/ml in 10 mM sodium acetate, pH 5.0, was injected over the sensor chip, followed by 30 μl of ethanolamine. A total of 3340 response units of Rse-Fc was immobilized on the sensor chip, corresponding to a density of 3.34 nig/ml. Conditioned media containing tagged Gas6 or deletion variants expressed in 293 cells were concentrated in Amicon 10 Centriprep spin concentrators, and the buffer was changed to PBS with 0.05% Tween 20 using Pharmacia PD10 Sephadex columns. The concentrations of tagged Gas6 or deletion variants were determined by ELISA assay using BIAcore protein 5B6 as the detecting antibody. 30 μl of conditioned media containing tagged Gas6 or deletion variants was injected onto the Rse-Fc sensor chip at a flow rate of 10 μl/min by the Kinject method. Proteins were allowed to associate for 20 min in the flow of PBS with 0.05% Tween 20. The sensor chip was regenerated by a short pulse of 2 μl of 10 mM HCl, followed by 2 μl of 10 mM NaOH before the next sample was injected. Sensorsgrams were analyzed with BIASEvaluation 2.1 software from Pharmacia Biosensor AB. Apparent dissociation rate constants (k) and association rate constants (k) were determined by obtaining the sensorsgrams with A + B → AB type I fitting. Equilibrium dissociation constant K was calculated as k/k.

RESULTS

Gas6/Protein S Chimeric Proteins Indicate the G Domains Determine Receptor Specificity—While human Gas6 binds to and activates human Rse, the structurally related protein human PS does not. We constructed and expressed chimeric versions of PS and Gas6 to determine if we could correlate the presence of specific domains of Gas6 with the ability to bind to and activate Rse. These proteins also contain an epitope tag (gD) that allows for quantitative comparison of their properties. P2′G′-gD contains amino acids 1–283 of PS which encode the Gla domain, loop region, and EGF-like repeats of Gas6 encoding the tandem G domains (Fig. 1A). The complementary fusion protein GNPC.gD contains amino acids 1–284 of Gas6 and 284–676 of PS, contains the Gla domain, loop, and EGF-like repeats of Gas6 and the G domains of PS. The fusion junctions of these proteins correspond to intron/exon boundaries dividing the domains containing the EGF-like repeats with the region containing the tandem G domains. The epitope tagged versions of Gas6, PS, P2′G′, or G2′G′ were expressed in stably transfected human embryonic kidney 293 cells and serum-free conditioned media collected from these cells were characterized by ELISA and Western blotting (Fig. 1B) as a means to monitor the size and expression levels of the proteins. Proteins of the expected size were observed, and the expression levels determined by ELISA correlated with the relative expression levels observed by Western blotting.

We compared the ability of tagged Gas6, PS, P2′G′, or G2′G′ to bind to a soluble form of the extracellular domain of Rse termed Rse-Fc. Conditioned media from human 293 cells expressing the variants were incubated with Rse-Fc or, as a control, just the Fc portion of IgG. Proteins that bound to the Fc fusion proteins were recovered from the supernatant with protein A and tagged proteins were revealed by Western blotting of the resolved precipitates. As observed previously (Godowski et al., 1995), Gas6.gD bound to Rse-Fc. The epitope tag at the
carboxyl terminus did not appear to influence receptor binding because Gas6RD bound to Rse-Fc as efficiently as a version containing the N terminus, which provided an epitope tag (Fig. 3) or authentic, untagged Gas6 (data not shown). The binding was specific because Gas6RD did not bind to an irrelevant Fc protein, and PS did not bind to Rse-Fc. Interestingly, PNgC RD, containing the Gla domain and EGF-like repeats of PS and the G domains of Gas6, bound to Rse-Fc. The complementary fusion protein GNPc RD did not bind to Rse-Fc. Thus, the ability to bind to the extracellular domain of Rse correlated with the presence of the G domains of Gas6.

While tagged Gas6 and PNgC were capable of binding to the extracellular domain of Rse in vitro, it was important to compare their ability to activate Rse expressed on the surface of cells. We compared the ability of tagged Gas6, PS, GNPc, and PNgC to induce phosphorylation of Rse expressed in NIH 3T3 cells. Conditioned media from mock-transfected cells or from cells expressing the variants were added to serum-starved NIH 3T3 Rse cells at 37 °C for 5 min. Rse was immunoprecipitated from cell lysates with antibody 19B, a polyclonal antibody directed against the extracellular domain of Rse. Immunoprecipitates were resolved by SDS-PAGE and immunoblotted with anti-phosphotyrosine antibodies (α-pTyr). The blot was stripped and probed with antibody 19B (α-Rse) to control for the amount of Rse on the blot. Sizes of the molecular weight standards are indicated on the left (in kilodaltons).
Characterization of Gas6 as a Ligand for Rse and Axl

Figure 4. Kinetics for binding of Gas6 and deletion variants to Rse-Fc coupled to a BIAcore™ biosensor. Rse-Fc was coupled to the carboxymethylated dextran layer on the surface of the biosensor chip. Either purified Gas6 or buffer-exchanged media containing the indicated proteins were injected over the surface of the biosensor at 160 s. At 340 s, the injector loop was switched to buffer to follow dissociation. No change in RU was observed when buffer-exchanged media from mock-transfected cells were passed over the Rse-Fc chip (data not shown).

Table I

| Ligand             | Association constant ($K_a$) | Dissociation constant ($K_d$) | Equilibrium dissociation constant ($K_{eq}$) |
|--------------------|-----------------------------|------------------------------|---------------------------------------------|
| Gas6               | $4 \times 10^4$             | $6 \times 10^{-4}$           | $4 \times 10^{-3}$                         |
| Gas6-gD            | $1 \times 10^4$             | $6 \times 10^{-4}$           | $6 \times 10^{-3}$                         |
| gD-Gas6118-C       | $1 \times 10^4$             | $7 \times 10^{-4}$           | $7 \times 10^{-3}$                         |
| gD-Gas6279-C       | $1 \times 10^4$             | $7 \times 10^{-4}$           | $7 \times 10^{-3}$                         |

Discussion

Gas6 is secreted ligand with structural homology to members of a superfamily of basement membrane proteins implicated in the growth and differentiation of many cells. A series of Gas6 variants were expressed to begin to define the relative roles of the Gla domain, EGF-like repeats, and G domains in receptor binding and activation. Our deletion studies show that the Gla and EGF-like repeats are not absolutely required for receptor binding or activation, and that the G domain region is sufficient for these activities. The on-rates and off-rates for binding of either full-length Gas6, Gas6118-C, and Gas6279-C to the extracellular domain of Rse were similar. This maps the receptor binding domain of Gas6 to the G domain-containing region. Furthermore, it shows that the Gla and EGF repeats contribute little to receptor binding.

We also quantitated the effects of deletion of the Gla and EGF-like repeats of Gas6 on activation of Rse phosphorylation. In the KIRA assay, we observed that the EC50 values for activation of Rse by epitope-tagged Gas6, Gas6118-C, and Gas6279-C were 12, 11, and 26 nM, respectively.

R. G. Hammonds and H. Raab, personal communication.
Characterization of Gas6 as a Ligand for Rse and Axl

acetylcholine receptors on skeletal muscle fibers is induced by the binding of the G domains of agrin and laminin to dystroglycan-α, a component of the dystrophin receptor (Gee et al., 1991; Campenelli et al., 1994; Gee et al., 1994).

Our results demonstrate that the G domains of Gas6 are necessary and sufficient for a functional interaction with both Rse and Axl and provide direct evidence that G domains can activate receptor tyrosine kinases. Furthermore, they suggest that other G domain-containing proteins may exert their effects by binding to cell adhesion molecule-related proteins and subsequently activating intracellular signaling pathways.

Acknowledgments—We thank Qimin Gu and Audrey Goddard for confirmation of the Gas6 and PS cDNA sequences, Marcel Reichert for support of the KIRA phosphorylation analyses, Andrew Nuljens for development and support of the gD competition ELISA, and Helga Raab for Gas6 purification. We thank Louis Tamayo for graphics, Brian Fendly for antibody 586, Greg Bennett for antibody 198, and the Genentech Oligonucleotide Facility for preparation of digoxin nucleotides.

REFERENCES

Bleske, L. G., Giandola, D. M., and Emerson, S. G. (1995) Oncogene 10, 2239–2242
Brunnendorf, T., and Rathjen, F. G. (1993). J. Neurochem. 61, 1207–1219
Campbell, I. D., and Bork, P. (1993) Curr. Opin. Struct. Biol. 3, 385–392
Campenelli, J. T., Roberts, S. L., Campbell, K. P., and Scheller, R. H. (1994) Cell 77, 663–674
Dahlback, B., Lundwall, A., and Stenflo, J. (1989) Proc. Natl. Acad. Sci. U. S. A. 86, 4199–4203
Fujimoto, J., and Yamamoto, T. (1994) Oncogene 9, 693–698
Gee, S. H., Blacher, R. W., Douville, P. J., Provost, P. R., Yurchenco, P. D., and Carbonetto, S. (1993) J. Biol. Chem. 268, 14972–14980
Hryb, D. J., Montanaro, F., Lindenbaum, M. H., and Carbonetto, S. (1994) Cell 77, 675–686
Gershagen, S. P., Fernlund, P., and Lundwall, A. (1987) FEBS Lett. 220, 129–135
Godowski, P. J., Mark, M. R., Chen, J., Sadick, M. R., Raab, H., and Hammond, R. L. (1995) Cell 82, 355–368
Graham, D. K., Dawson, T. L., Mullane, D. L., Snodgrass, H. R., and Ehr, H. S. (1994) Cell Growth & Diff. 5, 647–657
Graham, D. K., Bowman, G. W., Dawson, T. L., Stanford, W. F., Ehr, H. S., and Snodgrass, H. R. (1995) Oncogene 10, 2349–2359
Hammond, G. L., Underhill, D. A., Smith, C. L., Goping, I. S., Harley, M. J., Musto, N. A., Cheng, C. Y., and Bardin, C. W. (1987) FEBS Lett. 215, 100–104
Hryb, D. J., Khan, M. S., and Rosner, W. (1985) Biochem. Biophys. Res. Commun. 128, 432–440
Hryb, D. J., Khan, M. S., Romas, N. A., and Rosner, W. (1989) J. Biol. Chem. 264, 5378–5383
Janssen, J. W., Schulz, A. G., Steenwoorden, A. C. M., Schmidberger, M., Strehl, S., Ambros, P. F., and Bartram, C. R. (1991) Oncogene 6, 2113–2120
Joseph, D. R., and Baker, M. E. (1992) J. Biol. Chem. 267, 853–858
Manoletti, G., Brancolini, C., Avanzi, G., and Schneider, C. (1993) Mol. Cell. Biol. 13, 4976–4985
Mark, M. R., Loker, N. A., Ziechmeck, T. F., Luis, E. A., and Godowski, P. J. (1992) J. Biol. Chem. 267, 26166–26171
Mark, M. R., Scadden, D. T., Wang, Z., Gu, Q., Goddard, A., and Godowski, P. J. (1994) J. Biol. Chem. 269, 10720–10728
Nakhla, A. M., Khan, M. S., Romas, N. P., and Rosner, W. (1994) Proc. Natl. Acad. Sci. U. S. A. 91, 5402–5405
Noolan, D. M., Fulle, A., Valente, P., Cai, S., Horigan, E., Sasaki, M., Yamada, Y., and Hassell, J. R. (1993) J. Biol. Chem. 268, 22939–22947
O’Bryan, J. P., Frye, R. A., Cogwell, P. C., Niewabauer, A., Kitch, B., Prkoc, D., and Fujiimo, J., and Yamamoto, T. (1994) J. Biol. Chem. 269, 3467–3472
Ohashi, K., Mizuno, K., Kuma, M., Yitaka, M., and Nakamura, T. (1994) Oncogene 9, 669–705
Ohashi, K., Nagata, K., Toyota, J., Nakano, T., Arita, H., Tsuda, H., Suzuki, K., and Mizuno, K. (1995) J. Biol. Chem. 270, 22681–22684
Paborsky, L. R., Fendly, B. M., Fisher, K. L., Lawn, R. M., Marks, J. B., McCray, G., Tate, K. M., Vehar, G. A., and Gorman, C. M. (1990) Protein Eng. 3, 547–553
Pattay, L. (1993) FEBS Lett. 329, 99–103
Pattay, L., and Nikolic, K. (1993) Trends Neurosci. 16, 76–81
Pear, W. S., Nolan, G. P., Scott, M. L., and Baltimore, D. (1993) Proc. Natl. Acad. Sci. U. S. A. 90, 8392–8396
Rothberg, J. M., Jacobs, J. R., Goodman, C. S., and Artavanis-Tsakonas, S. (1990) Nature 344, 1207–1219
Rothschild, U., and Geis, D. R. (1995) Curr. Op. Neurobiol. 3, 709–715
Saito, T., Lai, C., Bruno, J., Radziejewski, C., Mattsson, K., Fisher, J., Geis, D. R., Jones, P. F., Maiakazkii, P., Ryan, T. E., Takeda, N. J., Chen, D. H., DelStefano, P. S., Long, G. L., Basilico, C., Goldfarb, M. P., Lenke, G., Glass, S. J., and Yanopoulos, G. D. (1995) Cell 80, 661–670
Tepass, U., Tiersch, C., and Knust, E. (1995) Cell 81, 797–809
Varmus, B. C., Young, C., Elliott, G., Garda, A., Bartley, T. D., Fridell, Y. -W., Hunt, R. W., Trail, G., Clogston, C., Toso, R. J., Yanagihara, D., Bennett, L., Sybler, M., McEwen, L., Tao, A., Escolar, E., Liu, E., and Yamada, H. K. (1995) Nature 373, 623–626
Characterization of Gas6, a Member of the Superfamily of G Domain-containing Proteins, as a Ligand for Rse and Axl
Melanie R. Mark, Jian Chen, R. Glenn Hammonds, Michael Sadick and Paul J. Godowsk

J. Biol. Chem. 1996, 271:9785-9789.
doi: 10.1074/jbc.271.16.9785

Access the most updated version of this article at http://www.jbc.org/content/271/16/9785

Alerts:
- When this article is cited
- When a correction for this article is posted

Click here to choose from all of JBC's e-mail alerts

This article cites 36 references, 13 of which can be accessed free at http://www.jbc.org/content/271/16/9785.full.html#ref-list-1