P2X₄ Receptor Is a Glycosylated Cardiac Receptor Mediating a Positive Inotropic Response to ATP*

Received for publication, December 18, 2001, and in revised form, February 22, 2002
Published, JBC Papers in Press, February 25, 2002, DOI 10.1074/jbc.M112097200

Bing Hu‡, Carol Senkler‡, Alexander Yang‡, Florentina Soto§, and Bruce T. Liang¶

From the ‡Departments of Medicine and Pharmacology, University of Pennsylvania, Philadelphia, Pennsylvania 19104 and §Max-Planck-Institut fuer Experimentelle Medizin, Abt. 300, Hermann-Rein-Str. 3, Goettingen 37075, Germany

Although P2X receptors are suggested to play a role in synaptic neurotransmission, the specific physiological role of each P2X receptor subtype remains largely unknown. We used cultured chick embryo ventricular myocytes as a model to study a potential physiological role of the P2X₄ receptor in mediating the positive inotropic effect of ATP. The chick P2X₄ receptor (cP2X₄R) mRNA was expressed in the heart and the pharmacological features of the ATP-induced positive inotropic response were similar to those of the cP2X₄R in terms of insensitivity to blockade by known P2 receptor antagonists and the ineffectiveness of adenosine 5'-triphosphate as an agonist. Treatment of myocytes with antisense oligonucleotides specific to the 5'- region of cP2X₄R abrogated the P2 agonist-stimulated ⁴⁰Ca influx. Similarly, antisense oligonucleotide treatment also blocked the 2-methylthio-ATP-stimulated increase in contractile amplitude. The data suggest that the native P2X₄ receptor is involved in mediating the P2 agonist-stimulated response in the heart. In characterizing the biochemical property of the P2X₄ receptor, antibody against cP2X₄R detected a 44-kDa and a 58-kDa protein in the immunoblot. Inhibition of N-linked glycosylation by tunicamycin converted the 58-kDa protein to the 44-kDa protein, suggesting that the 58-kDa protein was a glycosylated P2X₄ receptor. The nonglycosylated 44-kDa P2X₄ receptor was resistant to various detergent/aqueous extraction, consistent with a role of glycosylation in maintaining its detergent solubility and hydrophilicity. Cross-linking the cell surface proteins with N-hydroxysuccinimide-SS-biotin followed by affinity precipitation with streptavidin-conjugated agarose and subsequent immunoblotting with anti-cP2X₄R showed that only the glycosylated 58-kDa P2X₄ receptor was expressed on the cell surface, indicating an important role of glycosylation for the receptor's localization on the plasma membrane. These data revealed a novel physiologic function of the P2X₄ receptor and suggested the importance of N-linked glycosylation in its cell surface expression and detergent solubility.

P2X receptors are nonselective cation channels gated by ATP (for reviews, see Refs. 1–3). Seven isoforms of P2X receptors identified so far show 35–50% sequence identities based on comparison of the homologous region. Each receptor has two transmembrane domains and a large extracellular loop that contains a number of putative N-glycosylation sites and an ATP binding site. Recent human genome data suggested three additional possible P2X receptors in the human genome, and no P2X counterparts were found in the yeast or Drosophila genome (4). Both N- and C-intracellular termini have been shown to be important in determining the rate of desensitization of the P2X receptor (1–3). Biochemical and electrophysiological studies suggested homomeric and heteromeric structures, possibly trimers, as the functional form of P2X receptors (1–3, 5–7).

In vitro and in vivo studies indicated that P2X receptors are involved in a wide range of functions in synaptic transmission and in excitation of smooth muscle and inflammatory cells (1–3). Various phenotypes were found in P2X-deficient mice, such as reduced fertility in P2X₂R knockout male mice (8) and reduced sensitivity to pain in P2X₂R knockout mice (9). However, the physiological function of individual P2X receptors remains largely unknown.

The P2X₄ receptor was chosen for study of its physiological role for the following reasons. First, the pharmacological features of the native P2 receptor-mediated increase in cardiac myocyte contractility were most similar to those of the P2X₄ receptor. Thus, the P2 agonist 2-methylthio-ATP (2-meSATP)¹ is an efficacious agonist at stimulating both the myocyte contractility and the P2X₄ receptor-mediated current, whereas AMP-CPP is a weak agonist (18). Further, the increase in myocyte contractility was insensitive to blockade by the P2 antagonist suramin or pyridoxal phosphate-6-azophenyl-2',4'-disulfonic acid, as was the P2X₄ receptor-mediated increase in current. Finally, the receptor was expressed in the heart, and its cardiac transgenic overexpression resulted in an increased basal contractility of the intact heart, mediated via the endogenous ATP stimulating the overexpressed P2X₄ receptor (11).

Most membrane receptors are glycoproteins. The oligosaccharides are involved in various biological activities including intracellular trafficking, maintenance of receptor stability, recognition of ligand, and proper folding of the receptor. N-Linked glycosylation of the thrombin receptor in T lymphoid cells or of the insulin-like growth factor-1 receptor in Ewing’s sarcoma cells was shown to be important for localization of the receptor on plasma membrane (12, 13). However, glycosylation of the human parathyroid hormone/parathyroid hormone-related protein receptor was not important for their expression at the cell surface or for the integrity of their functional responsiveness (14). N-Linked glycosylation of the rat P2X₂ and P2X₁

¹ The abbreviations used are: 2-meSATP, 2-methylthio-ATP; AMP-CPP, adenosine 5'-triphosphate/triphosphate.
The molecular markers (RNA ladder; Invitrogen) are shown. B with sense or antisense oligonucleotides as described above. The effect (Sigma) for an additional 24–36 h as indicated under washes with fresh growth medium, the myocytes were cultured for an calcium phosphate/DNA precipitates for 6 h at 37°C. Analysis of cP2X4R transcript distribution in several 14-day-old embryo analysis of cP2X4R transcript in the heart was expressed at the same or a 1% SDS-PAGE and transferred to nitrocellulose membrane, followed by probing with the rabbit polyclonal antibody. Following several washes, the nitrocellulose membrane was incubated with peroxidase-conjugated anti-rabbit Ig antibody (1:5000) and developed with an ECL-Plus kit (Amersham Biosciences). The same antibody incubated with its peptide antigen was used as a negative control. Poly(A)+ mRNAs were isolated from tissues obtained from 14-day-old chicken embryos using the FastTrack kit 2.0 (Invitrogen). Ten micrograms of mRNA per tissue were separated in a 1% agarose gel containing formaldehyde, capillary-transferred onto a positively charged nylon membrane (Roche Molecular Biochemicals), and UV-cross-linked. The membrane was hybridized with a [α-32P]dCTP-labeled probe containing the complete coding region of cP2X4. The hybridization was performed in ExpressHyb solution (CLONTECH) at 68°C for 1 h, and the final wash of the membrane was done in 0.1× SSC, 0.1% SDS at 50°C. The membrane was exposed to x-ray film for 3 days.

**RESULTS AND DISCUSSION**

**P2X4 Receptor Is a Native Cardiac Physiological Receptor**—The physiological role of the recently identified ligand-gated P2X receptors is an area of much investigative interest. Whereas some P2X receptor-null mice suggested an important role of the P2X3 receptor in male fertility and of the P2X3 receptor in coding the physiological function. Thus, another objective of the study was to test the possible glycosylation of P2X4 receptor and to study the role of such glycosylation in the cell surface expression of the P2X4 receptor.

**EXPERIMENTAL PROCEDURES**

**DNA Constructs and Preparation of Sense and Antisense Oligonucleotides**—Chick P2X4 full-length clone was isolated from a 17-day-old embryo brain cDNA library (custom made; Stratagene) using as a probe a partial cP2X4 sequence obtained by degenerate PCR on RNA isolated from cultured cardiac ventricular myocytes as previously described (10) and subcloned into the pcDNA3 expression vector. The full-length cDNA was then subcloned in pcDNA3 expression vector as previously described (10). Phosphorothioated sense (CCGGCCGGCCGCGCATGCT) and antisense (GAGCATTCCGGCCGCGCC) oligonucleotides overlapping the 5′-untranslated region and translation start region of chick P2X4 cDNA were synthesized by the Cell Center at the University of Pennsylvania (start code location underlined).

**Preparation of Cultured Cardiac Cells, Transfection with cP2X4 Receptor cDNA, and Measurement of Contractile Amplitude and of 45Ca Uptake**—Cardiac ventricular cells were cultured from chick embryos 14 days in ovo according to previously described procedures (18). Cells were maintained in cultures for 12 h before being exposed to the calcium phosphate/DNA precipitates for 6 h at 37°C (19). After two washes with fresh growth medium, the myocytes were cultured for an additional 24 or 36 h as indicated under “Results.” For treatment with sense and antisense oligonucleotides, 1 μg sense or antisense oligonucleotides were included in calcium phosphate transfection mixtures. After 6 h, the medium was replaced with that containing the oligonucleotides for additional 2 days, at which time the effect on cell contract- tion and 45Ca uptake were measured. For inhibition of N-linked glycosylation, the cells transfected with the cP2X4R cDNA 12 h earlier were washed twice with fresh growth medium, and the cells were then maintained in the culture medium containing 1 μg/ml tunicamycin (Sigma) for an additional 24–36 h. Myocyte cultures were incubated with sense or antisense oligonucleotides as described above. The effect of 2-meSATP-stimulated 45Ca uptake was measured according to a previously described method (18).

**Immunoblotting and RNA Blotting**—Chick P2X4 antisera was raised in rabbit against a peptide (KKYKKVVEDYELGTSET) corresponding to the C terminus of chick P2X4 receptor (Cocalico Biologicals, Inc., Reamstown, PA). Isolated cardiac cells were solubilized in SDS-PAGE sample buffer, and 50 μg of solubilized proteins were resolved by
higher level than in the brain, although lungs have the highest level among the three tissues. In fact, the chick P2X4 receptor cDNA was cloned based on degenerate PCR on RNA isolated from the cultured chick cardiac cell (10). Second, in addition to being expressed in the heart, the pharmacological features of chick P2X4R-mediated ionic current also appeared similar to those of the native P2 receptor that mediated the ATP-induced increase in myocyte contractility (10, 18). Thus, 2-meSATP was a more efficacious agonist than AMP-CPP at the cloned cP2X4 receptor and at stimulating the native chick P2 receptor-mediated increase in myocyte contractility (18). The P2 receptor antagonist suramin (50 μM) did not block the native P2 receptor-mediated increase in myocyte contractile amplitude by 2-meSATP. The percentage of stimulation above basal level in

**Fig. 3.** Cellular effects of sense and antisense oligonucleotides directed at the cP2X4R. Following cultivation of the cardiac ventricular cells, the sense or the antisense oligonucleotides were added in the culture medium, and the effect on the immunoreactive level of cP2X4R was determined. (a), the anti-cP2X4R antibody detected two major bands, 58- and 44–45-kDa proteins, in an immunoblot of membranes from cP2X4R-overexpressing myocytes. Similarly, the corresponding effect on the 2-meSATP (1 μM)-stimulated increase in 45Ca uptake and contractile amplitude was quantitated (b). Values of the basal and P2 agonist-stimulated calcium influx (c) and contractile amplitude (d) under control (con; no treatment), sense, and antisense treatment conditions are shown. The calcium influx data (representative of values taken at the 30-s time point from three experiments) and the contractility tracing (representing 13 cells from four cultures for the antisense treatment and 10 cells from four cultures for sense treatment) were presented as means ± S.E.
response to 2-meSATP was 58 ± 15%, similar to the 53 ± 20% increase when suramin was also present (n = 8 cells from 4 cultures, p > 0.1 test) (Fig. 2). Similarly, the P2 antagonist pyridoxal phosphate-6-azophenyl-2',4'-disulfonic acid (50 μM) did not block the 2-meSATP-stimulated increase in myocyte contractility (data not shown). Although more work is needed, the pharmacological features of the P2 agonist-induced contractile response of the chick embryo cardiac myocyte are similar to those of the response in adult rat and mouse cardiac myocytes (11, 18). Since the P2X4 receptor was also detected in these adult mammalian cardiac myocytes, the present data are consistent with the idea that the chick cardiac cell is a potentially good model to investigate the physiological role and biochemical property of the cardiac P2X4R.

Finally, the cultured chick cardiac cells were amenable to the gene or cDNA transduction approach, which complemented functional (such as calcium influx and contractility) and biochemical (such as cell surface receptor expression) studies. This is an important advantage because of the feasibility in comparing the biochemical and the functional data in the same cell model.

To more definitively determine the function of the P2X4 receptor in mediating the 2-meSATP-stimulated increase in myocyte contractility, antisense oligonucleotide directed at the 5'-untranslated region contiguous with the translation start site of the cP2X4R was prepared. The antisense oligonucleotide against the cP2X4R effectively blocked the expression of the exogenous cP2X4R when both the oligonucleotide and the cP2X4R cDNA were transduced into the myocyte (Fig. 3a). The anti-cP2X4R antibody was unable to detect the native cP2X4R due to a low level expression of the native P2X4R or a low affinity of the antibody for the receptor or both. Therefore, antisense treatment of the myocytes expressing only native P2X4R could not have detected any decrease in its level using this antibody. On the other hand, overexpression of the exogenous recombinant receptor would allow the antibody to detect the receptor and thus the effect of antisense oligonucleotide treatment on the level of the receptor. That the antisense treatment effectively decreased the level of the overexpressed recombinant cP2X4R suggested that the same treatment should also have reduced or eliminated the native P2X4R receptor. Use of the cP2X4R-overexpressing myocyte was to demonstrate that antisense treatment can effectively reduce the level of the receptor.

If the native cP2X4R is involved in mediating the 2-meSATP-stimulated response, one would expect that such antisense oligonucleotide treatment would inhibit such a response. The present data demonstrated that this was indeed the case. Antisense oligonucleotide treatment abrogated the P2 agonist-stimulated 45Ca influx (−17 ± 7.8%, n = 3, ± S.E.) (Fig. 3, b and c). Similarly, antisense oligonucleotide treatment also blocked the 2-meSATP-stimulated increase in contractile amplitude (3.1 ± 1%, n = 13 cells from four cultures) (Fig. 3, b and d). On the other hand, myocytes transfected with pcDNA3 (mock-transfected) or with the sense oligonucleotide showed increases of 41.6 ± 13.8 and 61 ± 20% (n = 3, ± S.E.) in the level of 45Ca influx in response to 2-meSATP, respectively. Sense oligonucleotide-treated myocytes showed a preserved response to 2-meSATP with a 33 ± 3.5% (n = 10 cells from four cultures) increase in contractile amplitude (Fig. 3, b and d). Overall, these data provide more definitive evidence that the P2X4 receptor serves a physiological role, that of mediating an increase in the myocyte contractility and calcium influx in response to ATP.

P2X4 Receptor Is a Glycosylated Protein—Having established an important physiological role for the P2X4 receptor, its further biochemical characterization was carried out. The cP2X4R detected on the Western blot represented the exogenous recombinant chick P2X4R that was overexpressed following transfection of its cDNA. The anti-cP2X4R antibody detected two major bands, 58- and 44–45-kDa proteins, in an immunoblot of membranes from cP2X4R-overexpressing myocytes (Fig. 3a). The detected molecular mass of the P2X4 receptor antibody detected by the antibody against cP2X4R. In other experiments, cP2X4R cDNA-transfected cell were treated with 4% SDS or 1% Triton X-114 for 20 min and centrifuged at 12,000 × g for 15 min. The supernatant was further dissolved in SDS-PAGE sample buffer and blotted with antibody against cP2X4R. The data are representative of three experiments. b, the time course of expression of the 44–45- and 58-kDa proteins was determined following transfection of the cells with cP2X4R cDNA. Cells were solubilized 1, 2, or 3 days after the initiation of transfection (shown in lanes 1, 2, and 3, respectively). The solubilized extracts (in SDS sample buffer) were run on a SDS-page and blotted with antibody against cP2X4R. Data were representative of three experiments.
P2X<sub>4</sub> receptor is glycosylated in cardiac myocytes, similar to the glycosylation of P2X<sub>1</sub> and P2X<sub>2</sub> receptors (15, 16). At later time points following the transfection with exogenous cP2X<sub>4</sub>R cDNA, only the 58-kDa receptor protein was present (Fig. 4b), consistent with a more complete glycosylation of the exogenous cP2X<sub>4</sub>R. Only the 58-kDa P2X<sub>4</sub> receptor was detectable in cardiac myocytes isolated from adult wild type mouse and from P2X<sub>4</sub> receptor transgenic mice, which showed an increased cardiac physiology (11). These data suggested that glycosylation was necessary for maintaining the biochemical property and cell surface expression of the P2X<sub>4</sub> receptor.

**Glycosylation Is Required for Cell Surface Expression of the P2X<sub>4</sub> Receptor and for Maintaining Its Detergent Solubility**—To characterize the role of N-linked glycosylation in modulating the biochemical property of the P2X<sub>4</sub> receptor, studies were carried out to test the hypothesis that glycosylation is needed for cell surface expression of the P2X<sub>4</sub> receptor and for maintaining the detergent solubility of the receptor. Glycosylation has been reported to play an important role in intracellular trafficking and in the cell surface localization of various receptors and many other membrane-associated proteins. To test this possibility on the P2X<sub>4</sub> receptor, cP2X<sub>4</sub>R-overexpressing myocytes were biotinylated to label the cell surface receptor and treated with or without tunicamycin. The labeled receptor was extracted and precipitated by streptavidin-conjugated agarose beads. Immunoblotting with the anti-cP2X<sub>4</sub>R antibody showed that tunicamycin could completely block the cell surface expression of P2X<sub>4</sub> receptor (Fig. 5b), suggesting that N-linked glycosylation is required for its localization on the plasma membrane.

While SDS sample buffer could extract both glycosylated and nonglycosylated P2X<sub>4</sub> receptors, only the glycosylated receptor can be solubilized by 1% Triton X-114 (Fig. 4a). In fact, the nonglycosylated receptor (the 44–45-kDa protein) was resistant to various extraction conditions such as 1% Triton X-114 in the presence or the absence of 1% deoxycholic acid or 0.5 M urea (Fig. 6) or other conditions such as 1% Nonidet P-40 or 1% SDS (not shown). SDS, at the reduced concentration of 1.3%, was able to solubilize the 44–45-kDa nonglycosylated receptor (Fig. 5a). However, only the 58-kDa receptor, and not the 44–45-kDa receptor (also extracted by 1% SDS), could be precipitated by streptavidin (Fig. 5b). Thus, the 44–45-kDa receptor was not labeled with biotin, indicating that the nonglycosylated P2X<sub>4</sub> receptor failed to localize on the plasma membrane. Together, these data suggested that glycosylation was necessary for maintaining the receptor’s detergent solubility property and cell surface expression. Showing that the cell surface localization of the receptor requires glycosylation is clearly important for the function of these receptors, since they are ligand-gated receptor channels on the plasma membrane. Overall, the present study revealed a novel physiological function of the P2X<sub>4</sub> receptor and indicated the importance of N-linked glycosylation in maintaining its biochemical property and cell surface expression.

**Acknowledgments**—We are grateful to Dr. Anja Ruppelt for experimental help and to Dr. Walter Stühmer for generous support.

---

**Fig. 5.** Effect of tunicamycin treatment on the cell surface localization of P2X<sub>4</sub> receptor. *a*, cells transfected with cP2X<sub>4</sub>R cDNA were treated with or without tunicamycin and then subjected to extraction with 1.3% SDS to solubilize the glycosylated and nonglycosylated cP2X<sub>4</sub>R as described in the legend to Fig. 4. After centrifugation at 12,000 g for 15 min, the supernatant was further separated on a SDS-PAGE and blotted with cP2X<sub>4</sub>R antibody. Data were typical of three other experiments. *b*, cardiac ventricular cells were prepared, transfected with cP2X<sub>4</sub>R cDNA, and treated with tunicamycin or not treated. Cells were then subjected to cross-linking with N-hydroxysuccinimide-SS-biotin. Following extraction with 1.3% SDS for 20 min, the supernatant was obtained after centrifugation at 12,000 × g for 15 min. The cP2X<sub>4</sub>R in the supernatant was precipitated by streptavidin-conjugated agarose beads and detected by blotting with the antibody against cP2X<sub>4</sub>R. Data are representative of three other experiments.

**Fig. 6.** Ability of various detergents to solubilize the glycosylated and nonglycosylated cP2X<sub>4</sub> receptor. Cardiac ventricular cells were prepared, transfected with cP2X<sub>4</sub>R cDNA, and subjected to cross-linking with N-hydroxysuccinimide-SS-biotin as described under “Experimental Procedures.” Cells were then subjected to treatment with the various detergent combinations using the method described in the legend to Fig. 5. Part of the supernatant (Supernatant), obtained following the detergent solubilization, and the pellet fraction (Pellet) were dissolved in SDS-PAGE sample buffer and blotted with antibody against cP2X<sub>4</sub>R. Another portion of the supernatant was precipitated with streptavidin-conjugated agarose beads (IP), solubilized in SDS-PAGE, and blotted with antibody against cP2X<sub>4</sub>R. Data are representative of three other experiments.
REFERENCES

1. Khakh, B. S., Burnstock, G., Kennedy, C., King, B. F., North, R. A., Seguela, P., Voigt, M., and Humphrey, P. A. (2001) *Pharmacol. Rev.* **53**, 107–118
2. Soto, F., Garcia-Guzman, M., and Stuhmer, W. (1997) *J. Membr. Biol.* **160**, 91–100
3. Ralevic, V., and Burnstock, G. (1998) *Pharmacol. Rev.* **50**, 413–492
4. Venter, J. C., Adams, M. D., Myers, E. W., Li, P. W., Mural, R. J., Sutton, G. G., Smith, H. O., Yandell, M., Evans, C. A., Holt, R. A., Gocayne, J. D., Amanatides, P., Ballew, R. M., Hason, D. H., Wortman, J. R., et al. (2001) *Science* **291**, 1304–1351
5. Kim, M., Yoo, O. J., and Choe, S. (1997) *Biochem. Biophys. Res. Commun.* **240**, 618–622
6. Nicke, A., Baumert, H. G., Rettinger, J., Eichele, A., Lambrecht, G., Mutschler, E., and Schmalzing, G. (1998) *EMBO J.* **17**, 3016–3028
7. Stoop, H., Thomas, S., Rassendren, F., Kawashima, E., Buell, G., Surprenant, A, and North, R. (1999) *Mol. Pharmacol.* **56**, 973–981
8. Mulryan, K., Gitterman, D. P., Lewis, C. J., Vial, C., Leckie, B. J., Cobb, A. L., Brown, J. E., Conley, K. C., Buell, G., Pritchard, C. A., Evans, R. J. (2000) *Nature* **403**, 86–89
9. Souslova, V., Cesare, P., Ding, Y., Akoplan, A. N., Stanfa, L., Suzuki, R., Carpenter, K., Dickenson, A., Boyce, S., Hill, R., Nebenius-Osthuizen, D., Smith, A. J. H., Kidd, E. J., Wood, J. N. (2000) *Nature* **407**, 1015–1017
10. Ruppelt, A., Liang, B. T., and Soto, S. (1999) *Prog. Brain Res.* **120**, 81–90
11. Ho, B., Mei, Q., Smith, E., Barry, W. H., and Liang, B. T. (2001) *FASEB J.* **15**, 2739–2741
12. Tordai, A., Brass, L. F., Gelfand, E. W. (1995) *Biochem. Biophys. Res. Commun.* **206**, 857–862
13. Girnita, L., Wang, M., Xie, Y., Nilsson, G., Dricu, A., Wejde, J., Larsson, O. (2000) *Anticancer Drug Des.* **15**, 67–72
14. Bisello, A., Greenberg, Z., Behar, V., Rosenblatt, M., Suva, I. J. (1996) *Biocchemistry* **35**, 15890–15895
15. Torres, G. E., Egan, T. M., Voigt, M. M. (1998) *Biochemistry* **37**, 14845–14851
16. Rettinger, J., Aschrafi, A., Schmalzing, G. (2000) *J. Biol. Chem.* **275**, 33542–33547
17. Newbolt, A, Stoop, R., Virginia, C., Surprenant, A, North, R. A., Buell, G., and Rassendren, F. (1998) *J. Biol. Chem.* **273**, 15177–15182
18. Podrasky, E., Xu, D., and Liang, B. T. (1997) *Am. J. Physiol.* **273**, H2380–H2387
19. Xu, H, Miller, J., and Liang, B. T. (1992) *Nucleic Acids Res.* **20**, 6425–6426
20. Rubio, M. E., Soto, F. (2001) *J. Neurosci.* **21**, 641–653
21. Lé, K-T., Villeneuve, P., Ramjaun, A. R., McPherson, P. S., Beaudet, A., Seguela, P. (1998) *Neuroscience* **83**, 177–190
P2X₄ Receptor Is a Glycosylated Cardiac Receptor Mediating a Positive Inotropic Response to ATP

Bing Hu, Carol Senkler, Alexander Yang, Florentina Soto and Bruce T. Liang

J. Biol. Chem. 2002, 277:15752-15757.
doi: 10.1074/jbc.M112097200 originally published online February 25, 2002

Access the most updated version of this article at doi: 10.1074/jbc.M112097200

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 21 references, 8 of which can be accessed free at
http://www.jbc.org/content/277/18/15752.full.html#ref-list-1