Inhibition of Insulin-induced Glucose Uptake by Atypical Protein Kinase C Isotype-specific Interacting Protein in 3T3-L1 Adipocytes*

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Atypical protein kinase C (PKC) isotype-specific interacting protein (ASIP) specifically interacts with the atypical protein kinase C isozymes PKCα and PKCζ. ASIP and atypical PKC, as well as their Caenorhabditis elegans counterparts (PAR-3 and PKC-3, respectively), are thought to coordinately participate in intracellular signaling that contributes to the maintenance of cellular polarity and to the formation of junctional complexes. The potential role of ASIP in other cellular functions of atypical PKC was investigated by examining the effect of overexpression of ASIP on insulin-induced glucose uptake, previously shown to be mediated through PKCζ, in 3T3-L1 adipocytes. When overexpressed in these cells, which contain PKCa but not PKCζ, ASIP was co-immunoprecipitated with endogenous PKCα but not with PKCζ or with Akt. The subcellular localization of PKCα was altered in cells overexpressing ASIP. Overexpression of ASIP inhibited insulin stimulation of both glucose uptake and translocation of the glucose transporter GLUT4 to the plasma membrane, but it did not inhibit glucose uptake induced by either growth hormone or hyperosmolarity both of which promote glucose uptake in a PKCα-independent manner. Moreover, glucose uptake stimulated by a constitutively active mutant of PKCα, but not that induced by an active form of Akt, was inhibited by ASIP. Insulin-induced activation of PKCα, but not that of phosphoinositide 3-kinase or Akt, was also inhibited by overexpression of ASIP. These data suggest that overexpression of ASIP inhibits insulin-induced glucose uptake by specifically interfering with signals transmitted through PKCα.

Members of the protein kinase C (PKC) family of serine-threonine kinases were originally identified as effectors of lipid-mediated intracellular signaling (1, 2). Three classes of PKC isozymes have been defined: conventional PKC, consisting of PKCa, PKCb1, PKCb2, and PKCγ; novel PKC, comprising PKCd, PKCe, PKCζ, and PKCε; and atypical PKC, including PKCα and PKCζ (2, 3). Atypical PKC isozymes are distinct from other members of the PKC family in that they are not activated by diacylglycerol or phorbol esters (1–4). These isozymes are thought to act downstream of phosphoinositide (PI) 3-kinase. PKCζ is activated in vitro by phosphatidylinositol 3,4,5-trisphosphate (5), a cellular product of PI 3-kinase action, whereas PKCα was shown in transfected cells to contribute in a PI 3-kinase-dependent manner to trans-activation of the 12-O-tetradecanoylphorbol-13-acetate (phorbol ester)-responsive element induced by platelet-derived growth factor or epidermal growth factor (4). Furthermore, both the phosphorylation and activity of atypical PKC stimulated by growth factors were shown to be inhibited by either a pharmacological inhibitor of or a dominant negative mutant of PI 3-kinase (4, 6–9).

Atypical PKC isotype-specific interacting protein (ASIP) was recently identified as a protein that specifically associates with atypical PKC (10). ASIP does not contain any known catalytic domain but possesses three PDZ domains (10), which mediate protein-protein interactions at the plasma membrane (11). PAR-3, the Caenorhabditis elegans ortholog of ASIP, also interacts with the worm counterpart of atypical PKC (PKC-3) (12). Worm embryos lacking PAR-3 show severe defects in asymmetrical cell division, and PKC-3-depleted cells show a phenotype highly similar to that of PAR-3-depleted cells (12). Bazooka, a protein essential for asymmetrical cell division in Drosophila neuroblasts, has been identified as a fly ortholog of ASIP and PAR-3 (13, 14). Although a fly homolog of atypical PKC has not been identified, these observations, together with the fact that ASIP and PKCα colocalize at junctional complexes in mamalian cells (10), suggest that ASIP and atypical PKC coordinately function in the maintenance of cellular polarity and cell-cell interaction.

We have now investigated whether ASIP also contributes to other cellular functions of atypical PKC. Stimulation of glucose uptake by insulin in its target cells, such as skeletal muscle and adipocytes, is an important biological action of this hormone. Pharmacological and molecular biological evidence has revealed that PI 3-kinase plays a major role in insulin-induced glucose uptake (15–17). Although the precise mechanism by which activation of PI 3-kinase results in glucose uptake remains unclear, we and others have shown that a kinase-deficient mutant of atypical PKC inhibits insulin-induced glucose uptake (7, 8) and that a constitutively active mutant of atypical PKC stimulates glucose uptake in quiescent cells (8). Furthermore, microinjection of antibodies specific to PKCα inhibited insulin-induced translocation of the glucose transporter GLUT4 to the plasma membrane (18). These results support...
the hypothesis that atypical PKC participates in the signaling pathway by which insulin stimulates glucose uptake.

To elucidate whether ASIP contributes to insulin stimulation of glucose uptake, we have examined the effects of overexpression of ASIP on glucose uptake induced by insulin and by various other stimuli, as well as on the activation of signaling molecules by insulin, in cultured adipocytes. We now provide evidence that overexpression of ASIP inhibits insulin-induced glucose uptake by specifically interfering with signaling mediated through PKCα.

**EXPERIMENTAL PROCEDURES**

**Cells and Antibodies—**3T3-L1 preadipocytes (American Type Culture Collection) were maintained and induced to differentiate into adipocytes as described previously (8). Polyclonal antibodies to PKCα that were generated in response either to a peptide corresponding to amino acids 197–213 (α197) or to a glutathione S-transferase fusion protein containing amino acids 190–240 (α190) of mouse PKCα were as described (8). Polyclonal antibodies to ASIP (10), to GLUT4 (19), or to Akt were generated in response either to a peptide corresponding to amino acids 190–240 (αe190), to PKCα (αPKCα), or to Akt, or to T7 (lower panels). The positions of molecular size standards (in kilodaltons) are indicated on the left. Data are representative of three independent experiments.

**Glucose Uptake—**Glucose uptake was assayed as described previously (8). In brief, 3T3-L1 adipocytes infected (or not) with adenovirus vectors were incubated for 16 h in Dulbecco’s modified Eagle’s medium containing 5.6 mM glucose and 0.5% fetal bovine serum. The cells were washed twice with DB buffer (140 mM NaCl, 2.7 mM KCl, 1 mM CaCl2, 1.5 mM KH2PO4, 8 mM Na2HPO4 (pH 7.4), 0.5 mM MgCl2) and then incubated in DB buffer with 100 nM insulin for 10 min, growth hormone (GH) (0.5 µg/ml) for 10 min, or 300 mM sorbitol for 60 min. DB buffer (1 ml) containing bovine serum albumin (1 mg/ml) and 0.1 mM 2-deoxy-D-[1,2-3H]glucose (1 µCi) was added to each well, and after 5 min, the cells were washed with DB buffer and then solubilized with 0.1% (w/v) SDS. The radioactivity incorporated into the cells was measured with a liquid scintillation counter.

**RESULTS**

Coprecipitation of ASIP with PKCα from 3T3-L1 Adipocytes—ASIP was identified as a protein that specifically interacted with the atypical PKC isoforms PKCα and PKCζ (10). We therefore first examined whether ASIP binds to atypical PKC in 3T3-L1 adipocytes, which express PKCα exclusively (8). The cells were infected (or not) with an adenovirus vector that encodes T7 epo- tagged ASIP (AxCAASIP), lyzed, and subjected to immunoprecipitation with antibodies to T7 or to PKCα, or with control immunoglobulin G (IgG). The resulting immunoprecipitates were then subjected to immunoblot analysis with antibodies to either T7 or PKCα, but only when the cells had been infected with AxCAASIP (Fig. 1). Conversely, a protein of ~80 kDa, once with buffer A containing 1 M NaCl, and once with a solution containing 20 mM Tris-HCl (pH 7.5), 5 mM MgCl2, and 1 mM EGTA. The precipitates were then incubated for 14 min at 30 °C with 0.4 µCi of [γ-32P]ATP in a reaction mixture (25 µl) containing 35 mM Tris-HCl (pH 7.5), 10 mM MgCl2, 0.5 mM EGTA, 0.1 mM CaCl2, 40 µM unlabelled ATP, and 30 µM myelin basic protein as substrate. All protein kinase reactions were terminated by the addition of SDS sample buffer, and the reaction mixtures were then fractionated by SDS-polyacrylamide gel electrophoresis. The radioactivity incorporated into substrates was determined with a Fuji BAS 2000 image analyzer.

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The results are shown in Table 1. Cells were infected (or not) with adenovirus vectors encoding T7 or to PKCα, or with control IgG. The resulting immunoprecipitates were then subjected to immunoblot analysis with antibodies to ASIP or to PKCα (upper panels), to PKCα (monoclonal antibody), to PKCα, or Akt (IP) and subjected to immunoblot analysis with antibodies to T7 or to PKCα, or to Akt (lower panels). The positions of molecular size standards (in kilodaltons) are indicated on the left. Data are representative of three independent experiments.
kDa, corresponding to the molecular size of PKC, was recognized by antibodies to PKCα in the immunoprecipitates prepared with antibodies to PKCα or to T7. No marked immunoreactive bands were detected in the immunoprecipitates prepared with control IgG. These data indicate that PKCα associates with ASIP in 3T3-L1 adipocytes. The immunoprecipitates with antibodies to PKCα prepared from noninfected 3T3-L1 adipocytes did not react to antibodies to ASIP (data not shown). This may be because a relatively small amount of ASIP protein is expressed in the cells.

To investigate the specificity of the interaction between ASIP and PKCα, we performed similar experiments with antibodies to PKCe or to Akt. However, the 180-kDa protein recognized by antibodies to PKCe at T7 was not evident in the immunoprecipitates prepared with antibodies to either PKCe or Akt, and neither PKCe nor Akt was detected in the immunoprecipitates prepared with antibodies to T7 (Fig. 1). The interaction of ASIP with PKCα thus appeared to be specific, consistent with previous observations (10). Treatment of the cells with insulin did not affect the interaction between ASIP and PKCα (data not shown).

Effect of ASIP on the Subcellular Localization of PKCα—We next investigated the effect of overexpression of ASIP on the subcellular localization of PKCα. 3T3-L1 adipocytes infected (or not) with AxCAASIP were lysed, and LDM, PM, and cytosolic fractions were prepared by differential centrifugation. Equal amounts of protein from each fraction were then subjected to immunoblot analysis with antibodies to ASIP or to PKCα (monoclonal antibody). Data are representative of three independent experiments.

Inhibition of Insulin-induced Glucose Uptake by ASIP—
Given that PKCα is thought to participate in insulin-induced glucose uptake and GLUT4 translocation in 3T3-L1 adipocytes (8, 18), we examined the effects of overexpression of ASIP on these actions of insulin. Infection of the cells with AxCAASIP resulted in a dose-dependent increase in the amount of ASIP protein as assessed by immunoblot analysis; the amount of ASIP in cells infected at a multiplicity of infection (m.o.i.) of 150 plaque-forming units (pfu/cell) was ~20 times that of endogenous ASIP (Fig. 3A). Insulin induced a ~7-fold increase in glucose uptake in 3T3-L1 adipocytes within 10 min (Fig. 3A).
Expression of ASIP inhibited insulin stimulation of glucose uptake in a dose-dependent manner, with ~50% inhibition apparent at an m.o.i. of 150 pfu/cell; ASIP had little effect on the basal level of glucose uptake apparent in the absence of insulin. We next investigated the effect of ASIP on the translocation of GLUT4 by subcellular fractionation. Treatment of noninfected cells with insulin reduced the amount of GLUT4 in the LDM fraction and increased the amount of GLUT4 in the PM fraction (Fig. 3A). Infection of cells with AxCAASIP, at an m.o.i. of 150 pfu/cell, markedly inhibited the insulin-induced decrease in the amount of GLUT4 in the LDM fraction as well as the increase in its abundance in the PM fraction. The extent of ASIP-induced inhibition of GLUT4 translocation was similar to that of ASIP-induced inhibition of glucose uptake. The amount of GLUT4 in total cell lysates was not affected by overexpression of ASIP (data not shown). These results suggest that overexpression of ASIP inhibited insulin-induced glucose uptake by preventing the intracellular translocation of GLUT4.

Effects of ASIP on Glucose Uptake Induced by Various Stimuli—GH and hyperosmolarity each stimulate glucose uptake by promoting translocation of GLUT4 (24, 25). The observation that glucose uptake induced by these stimuli was not inhibited by a dominant negative mutant of PKCζ (8) suggests that it is independent of PKCζ. Infection of 3T3-L1 adipocytes with AxCAASIP at an m.o.i. of 150 pfu/cell, a virus dose sufficient to inhibit insulin-induced glucose uptake by ~50%, had no effect on hyperosmolarity induced glucose uptake and actually potentiated GH-induced glucose uptake (Fig. 3C), suggesting that expression of ASIP inhibits glucose uptake by specifically interfering with signaling through PKCζ.

To examine further this hypothesis, we investigated the effect of ASIP on glucose uptake stimulated by a constitutively active mutant of PKCζ. The ΔζPD mutant lacks the pseudosubstrate domain and exhibits a kinase activity markedly greater than that of the wild-type enzyme (8). Given that ASIP binds to the kinase domain of atypical PKC (10), it is likely that ASIP also interacts with ΔζPD. Consistent with our previous observation (8), expression of ΔζPD stimulated glucose uptake in quiescent 3T3-L1 adipocytes. Overexpression of ASIP inhibited ΔζPD-induced glucose uptake in a dose-dependent manner (Fig. 4A). Expression of constitutively active mutants of Akt also stimulates glucose uptake and translocation of GLUT4 in adipocytes (26, 27). Indeed, infection of 3T3-L1 adipocytes with AxCAMyr-Akt, an adenovirus vector that encodes a constitutively active mutant of Akt (22), stimulated glucose uptake in 3T3-L1 adipocytes in the absence of insulin (Fig. 4B). However, expression of ASIP did not inhibit Myr-Akt stimulation of glucose uptake, suggesting that ASIP does not interrupt signaling through Akt.

Effects of ASIP on Various Signaling Molecules Activated by Insulin—To elucidate further the mechanism by which ASIP inhibits insulin-induced glucose uptake, we investigated the effects of overexpression of this protein on the activation of various signaling molecules by insulin. Insulin-induced activation of PI 3-kinase or of Akt was not affected by ASIP (Fig. 5, A and B). Furthermore, overexpression of ASIP had no effect on the insulin-induced translocation of Akt to the PM fraction (Fig. 5C).

Finally, we examined the effect of ASIP overexpression on kinase activity immunoprecipitated with antibodies to PKCζ. Exposure of cells to insulin increased the amount of kinase activity, measured with myelin basic protein as substrate, in such immunoprecipitates by a factor of ~1.8 (Fig. 6A). Overexpression of ASIP inhibited the insulin-induced kinase activity in a dose-dependent manner, with complete inhibition apparent at an m.o.i. of 150 pfu/cell (Fig. 6A). The amount of PKCζ protein in the immunoprecipitates prepared with antibodies to PKCζ was increased by expression of ASIP in a dose-dependent manner (data not shown), with an ~2-fold increase apparent at an m.o.i. of 150 pfu/cell (Fig. 6B).

**DISCUSSION**

Given that cell permeable pharmacological inhibitors or activators of atypical isozymes of PKC are not available, the cellular functions of these isozymes have been explored with the use either of pseudosubstrate peptides that specifically compete with endogenous substrates for kinase activity or of mutant enzymes that either are constitutively active or act in a dominant negative manner. Such tools have revealed putative roles for atypical PKC in oocyte maturation (28), mitogenesis (29), protection of cells from apoptosis (30), cell differentiation (31), and insulin-induced glucose uptake (7, 8). Another approach to investigating the cellular functions of signaling molecules is to identify proteins that directly interact with them. Several proteins that associate with atypical PKC have been identified, including p62/ZIP (32), LIP (33), PAR-4 (34), UNC-76 (35), and the small GTP-binding protein RAS (36). The cDNA that encodes ASIP was also isolated as a result of screening an expression library with PKCζ as a probe (10).

With the use of adenovirus-mediated gene transfer, we have now shown that overexpression of ASIP in 3T3-L1 adipocytes inhibited insulin stimulation of both glucose uptake and GLUT4 translocation. GH and hyperosmolarity (sorbitol) also stimulate glucose uptake but in a manner insensitive to a dominant negative mutant of PKCζ (8), suggesting that these stimuli induce glucose uptake through a PKCζ-independent
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Our observation that overexpression of ASIP did not inhibit glucose uptake in response to either GH or hyperosmolarity suggests that ASIP attenuates glucose uptake not by an effect on a common component of the transport machinery but by interfering with specific signals transmitted through PKCα. This hypothesis was further supported by the observation that ASIP inhibited glucose uptake induced by a constitutively active mutant of PKCα but not that induced by an active form of Akt. It is not clear why overexpression of ASIP enhanced glucose uptake induced by GH. Given that a dominant negative mutant of PKCα did not exhibit such an effect (8), it is not likely that prevention of signaling through PKCα was responsible for this action of ASIP. We therefore cannot exclude the possibility that overexpression of ASIP affects not only PKCα-mediated signaling but also an unidentified signaling pathway that contributes to GH-induced glucose uptake.

The results of the present and previous (26, 27) studies have shown that constitutively active mutants of Akt stimulate both glucose uptake and the translocation of GLUT4 in quiescent cells. However, studies on the effects of kinase defective Akt mutants on insulin-induced glucose uptake or translocation of GLUT4 have produced conflicting results (20, 37, 38). It is thus unclear whether Akt plays a role in insulin-stimulated glucose uptake. We have now shown that the activation of Akt by insulin was not affected in cells overexpressing ASIP, whereas insulin-induced glucose uptake in these cells was inhibited by ~50%. Moreover, insulin-induced translocation of Akt to the plasma membrane, thought to be an important step in Akt activation (39), was not inhibited by ASIP, consistent with the observation that ASIP did not associate with Akt in intact cells. It is not likely that ASIP inhibits insulin-induced glucose uptake by interfering with signaling downstream of Akt, because ASIP had no effect on glucose uptake induced by a constitutively active mutant of Akt. Although our data do not exclude a role for Akt in insulin stimulation of glucose uptake, insulin-induced activation of Akt alone is likely not sufficient to fully stimulate glucose uptake, at least in 3T3-L1 adipocytes.

Immunocytochemical analysis of MDCKII and NIH 3T3 cells has revealed that endogenous ASIP is specifically localized to regions of cell-cell contact, most likely at tight junctions, whereas PKCα not only colocalizes with ASIP in these regions but is also distributed throughout the cytoplasm (10). With the use of subcellular fractionation analysis, we have now shown that overexpression of ASIP alters the subcellular localization of endogenous PKCα in 3T3-L1 adipocytes. Whereas PKCα was abundant in the LDM and cytosolic fractions of control cells, overexpression of ASIP resulted in a decrease in the amount of PKCα in the cytosolic fraction and in marked and small in-

Fig. 5. Effects of overexpression of ASIP on insulin-induced activation of PI 3-kinase and Akt. A and B, 3T3-L1 adipocytes were infected with AxCAASIP at the indicated m.o.i. (pfu/cell) and then incubated in the absence or presence of 100 nM insulin for 5 min (A) or 10 min (B). Cells were then lysed and subjected to immunoprecipitation with antibodies to phosphotyrosine (A) or to Akt (B), and the resulting precipitates were assayed for PI 3-kinase and Akt activity, respectively. Data are expressed as fold stimulation relative to the activities of noninfected, nonstimulated cells and are the mean ± S.E. from three independent experiments. C, 3T3-L1 adipocytes were infected (or not) with AxCAASIP at an m.o.i. of 150 pfu/cell and incubated in the absence or presence of 100 nM insulin for 15 min, and then subjected to subcellular fractionation for isolation of the PM fraction. This fraction (~20 μg of protein) was then subjected to immunoblot analysis with antibodies to Akt2. Data are representative of three independent experiments.

Fig. 6. Effect of overexpression of ASIP on insulin-induced activation of PKCA. A, 3T3-L1 adipocytes were infected with AxCAASIP at the indicated m.o.i. (pfu/cell) and then incubated in the absence or presence of 100 nM insulin for 5 min. The cells were then lysed and subjected to immunoprecipitation with antibodies to PKCA (α197), and the resulting precipitates were assayed for kinase activity with myelin basic protein as substrate. Data are expressed as fold stimulation relative to the activity of noninfected, nonstimulated cells and are the mean ± S.E. from three independent experiments. B, 3T3-L1 adipocytes were infected (or not) with AxCAASIP at an m.o.i. of 150 pfu/cell and incubated in the absence or presence of 100 nM insulin for 5 min. Cells were then lysed and subjected to immunoprecipitation with antibodies to PKCα (α197), and the resulting precipitates were subjected to immunoblot analysis with antibodies to PKCα (monoclonal antibody). Data are representative of three independent experiments.
creases in the amounts of the isozyme in the LDM and PM fractions, respectively. The observation that recombinant ASIP was also abundant in the LDM and PM fractions is consistent with the notion that the altered subcellular localization of PKCa is attributable to its direct interaction with ASIP.

The mechanism by which ASIP inhibits PKCa signaling in the pathway that leads to glucose uptake remains unclear. Our observation that the amount of PKCa immunoprecipitated by antibodies to this isozyme from cells overexpressing ASIP was about twice that precipitated from control cells might be explained if one ASIP molecule interacts with multiple PKCa molecules or if ASIP forms a multimeric complex in cells. Despite the increase in the amount of PKCa protein, the kinase activity in the immunoprecipitates prepared from cells overexpressing ASIP was lower than in the precipitates prepared from control cells, suggesting that PKC activity is suppressed when it is bound to ASIP. It is thus possible that overexpression of ASIP inhibits PKCa activity in the cells, thereby resulting in inhibition of insulin-induced glucose uptake. At present, we do not know the mechanism how overexpression of ASIP reduces PKCa activity. Because the activity of PKC isoenzymes is regulated by its phosphorylation (3), it is possible that ASIP affects phosphorylation of specific residues of PKCa.

Biochemical and genetic evidence suggests that ASIP, as well as its worm (PAR-3) and fly (Bazooka) counterparts, participate in asymmetrical cell division or maintenance of cellular polarity (10, 12–14). Because ASIP does not contain any known catalytic domain but does possess PDZ domains, which are thought to mediate protein-protein interactions, it is possible that ASIP acts as a cellular scaffold that provides a location for atypical PKC to transmit signals that establish cell polarity. Moreover, ASIP is phosphorylated in vitro (10), and our recent experiments showed that ASIP is phosphorylated in intact cells via a PKC-dependent manner. It is possible that overexpression of ASIP may displace PKCa from a compartment in which it plays a role in glucose uptake or may compete with an unidentified substrate that is involved in glucose uptake or may compete with molecules with which it interacts, to mediate signaling that leads to glucose uptake should help to clarify the molecular mechanism of this important biological effect of insulin.

REFERENCES

1. Nishizuka, Y. (1992) Science 258, 607–613
2. Nishizuka, Y. (1995) FASEB J. 9, 484–496
3. Mellor, H., and Parker, P. J. (1998) Biochem. J. 332, 281–282
4. Akimoto, K., Takahashi, R., Moriya, S., Nishikawa, N., Takayanagi, J., Kimura, K., Fukui, Y., Osada, S.-I., Mizuno, K., Hirai, S.-I., Kazulauskas, A., and Ohno, S. (1996) EMBO J. 15, 788–798
5. Nakanishi, H., Brewer, K. A., and Exton, J. H. (1993) J. Biol. Chem. 268, 13–16

Y. Tamai and S. Ohno, manuscript in preparation.

6. Mendez, R., Kollmorgen, G., White, M. F., and Rhoads, R. E. (1997) Mol. Cell. Biol. 17, 5184–5192
7. Bandopadhayay, G., Standaert, M. L., Zhao, L., Binghi, Y., Arigoni, A., Galloway, L., Karnam, P., Moscat, J., and Farese, R. V. (1997) J. Biol. Chem. 272, 2551–2558
8. Kotani, K., Ogawa, W., Matsumoto, M., Kitamura, T., Sakauke, H., Hino, Y., Miyake, K., Sano, W., Akimoto, K., Ohno, S., and Kasuga, M. (1998) Mol. Cell. Biol. 18, 6971–6982
9. Romanelli, A., Martin, K. A., Toker, A., and Blenis, J. (1999) Mol. Cell. Biol. 19, 2921–2928
10. Izumi, Y., Hirose, T., Tamai, Y., Hirai, S., Nagashima, Y., Fujimoto, T., Tabuse, Y., Kemphues, K. J., and Ohno, S. (1998) J. Cell Biol. 143, 95–106
11. Fanning, A. S., and Andersen, J. M. (1999) J. Clin. Invest. 103, 767–772
12. Tabuse, Y., Izumi, Y., Piano, F., Kemphues, K. J., Miwa, J., and Ohno, S. (1998) Development 125, 3607–3614
13. Schober, M., Schaefer, M., and Knobil, J. A. (1999) Nature 402, 548–551
14. Wodorz, A., Ramrath, A., Kuchinke, U., and Knust, E. (1999) Nature 402, 544–547
15. Holman, G. D., and Kasuga, M. (1997) Diabetes 46, 991–1003
16. Ogawa, W., Matozaki, T., and Kasuga, M. (1998) Mol. Cell. Biochem. 182, 13–22
17. Sakauke, H., Ogawa, W., Takata, M., Kuroda, S., Kotani, K., Matsumoto, M., Sakauke, M., Nishio, S., Ueno, H., and Kasuga, M. (1997) Mol. Endocrinol. 11, 1552–1562
18. Nishizuka, Y. (1992) Science 258, 607–613
19. Kiyoshima, T., Nakanishi, H., Ueno, H., and Kasuga, M. (1997) Mol. Endocrinol. 11, 1552–1562
20. Bandyopadhyay, G., Eisele, M., Matsunami, H., and Nishizuka, Y. (1999) J. Biol. Chem. 274, 332–337
21. Kitamura, T., Takahashi, K., Nakashima, H., Uchiyama, H., and Matsumoto, K. (1999) Mol. Cell. Biol. 19, 6765–6774
22. Hashimimoto, M., and James, D. E. (1997) J. Biol. Chem. 272, 6971–6982
23. Tabuse, Y., Kuroda, S., Hino, Y., Ogawa, W., Sakauke, M., Matsumoto, M., Maeda, T., Konishi, H.,ocka, S., and Kasuga, M. (1998) Mol. Cell. Biol. 18, 3708–3717
24. Miyake, S., Makinori, M., Kagey, Y., Harada, S., Sato, Y., Takamori, K., Tokuda, C., and Saito, I. (1996) Proc. Natl. Acad. Sci. U. S. A. 93, 1200–1204
25. Kitamura, T., Kitamura, Y., Kuroda, S., Hino, Y., Ando, M., Kotani, K., Konishi, H., Matsuzaki, H., Kikka, W., Ogawa, W., and Kasuga, M. (1998) Mol. Cell. Biol. 19, 6286–6296
26. Simpson, I. A., Yer, D. H., Hissin, P. J., Wardzala, L. J., Kariell, E., Salans, L. B., and Cushman, S. W. (1983) Biochim. Biophys. Acta 763, 393–407
27. Tannen, J. W., Leinfield, K., Mueckler, M. M., and Lien, C. K. (1992) Biochem. J. 282, 105–114
28. Toyoda, N., Robinson, F. W., Smith, M. M., Flanagan, J. E., and Kono, T. (1986) J. Biol. Chem. 261, 2117–2122
29. Kohn, A. D., Summers, S. A., Birnbaum, M. J., and Roth, R. A. (1996) J. Biol. Chem. 271, 3172–3178
30. Tanti, J. F., Grillo, S., Gremeaux, T., Cofer, P. J., Van-Obberghen, E., and Le-Marchand-Brustel, Y. (1997) Endocrinology 138, 2005–2010
31. Dominguez, I., Diaz-Meco, M. T., Munico, M. M., Berra, E., Garcia de Herreros, A., Cortan, M. E., Sanz, L., and Moscat, J. (1992) Mol. Cell. Biochem. 12, 3776–3783
32. Berra, E., Diaz-Meco, M. T., Dominguez, I., Munico, M. M., Sanz, L., Lozano, J., Chapkin, R. S., and Moscat, J. (1997) Cell 74, 555–563
33. Berra, E., Munico, M. M., Sanz, L., Frutos, S., Diaz-Meco, M., and Moscat, J. (1997) Mol. Cell. Biol. 17, 4346–4354
34. Wooton, M. W., Zhou, G., Seibenhener, M. L., and Coleman, E. S. (1994) Proc. Natl. Acad. Sci. U. S. A. 91, 6191–6196
35. Diaz-Meco, M. T., Munico, M. M., Sanchez, P., Lozano, J., and Moscat, J. (1996) Mol. Cell. Biol. 16, 105–114
36. Diaz-Meco, M. T., Lozano, J., Munico, M. M., Berra, E., Frutos, S., Sanz, L., and Moscat, J. (1996) Cell 86, 777–786
37. Wang, Q., Somwar, R., Bilan, P. J., Liu, Z., Jin, J., Woodgett, J. R., and Klip, A. (1999) Mol. Cell. Biol. 19, 4008–4018
38. Coffer, P. J., Jin, J., and Woodgett, J. R. (1998) Biochem. J. 335, 1–13
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