Transcriptional Activation of the Proglucagon Gene by Lithium and β-Catenin in Intestinal Endocrine L Cells*

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1 The abbreviations used are: GLP-1, glucagon like peptide 1; CRE, cAMP-response element; β-cat, β-catenin; TCF, T cell factor; cat-TCF, a TCF; TK, thymidine kinase; RIA, radioimmunoassay.

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The proglucagon gene encodes several peptide hormones that regulate blood glucose homeostasis, growth of the small intestine, and satiety. Among them, glucagon-like peptide 1 (GLP-1) lowers blood glucose levels in patients with diabetes and inhibits eating and drinking in fasted rats. Although proglucagon transcription and GLP-1 synthesis were shown to be activated by forskolin and other protein kinase A (PKA) activators, deleting or mutating the cAMP-response element (CRE) only moderately attenuates the proglucagon gene promoter in response to PKA activation. Therefore, PKA may activate proglucagon transcription via a mechanism independent of the CRE motif. Recently, PKA was shown to phosphorylate and inactivate GSK-3β, a key mediator in the Wnt signaling pathway. We show here that lithium, an inhibitor of GSK-3β, activates proglucagon gene transcription and stimulates GLP-1 synthesis in an intestinal endocrine L cell line, GLUTag. The activation was also observed in primary fetal rat intestinal cell (FRIC) cultures, but not in a pancreatic A cell line. Co-transfection of β-catenin, a downstream effector of GSK-3β activities, activated the proglucagon gene promoter without a CRE. Furthermore, forskolin and 8-Br-cAMP phosphorylated GSK-3β at serine 9 in intestinal proglucagon-producing cells, and both lithium and forskolin induced the accumulation of free β-catenin in these cell lines. These observations indicate that the proglucagon gene is among the targets of the Wnt signaling pathway.

The gene encoding proglucagon is expressed in pancreatic A cells and enteroendocrine L cells as well as selective neurons of the central nervous system (1). A single proglucagon mRNA transcript, derived from a common transcription start site, is identical in all the three tissues. Cell type-specific posttranslational processing, however, gives rise to a different profile of proglucagon-derived peptides in each tissue (1, 2). Glucagon is identical in all the three tissues. Cell type-specific posttranslational processing, however, gives rise to a different profile of proglucagon-derived peptides in each tissue (1, 2). Glucagon is produced in the pancreatic A cells. Glucagon-like peptide 1 (GLP-1) and glucagon-like peptide 2 (GLP-2), however, are produced in the endocrine L cells and in selected endocrine neurons in the brain (1, 2). GLP-1 is known as an insulino-tropic hormone. It possesses potent effects to stimulate glucose-dependent insulin secretion, insulin gene expression, and B cell CAMP formation. Other effects of GLP-1 include inhibition of glucagon release and gastric emptying and, possibly, enhancement of peripheral insulin sensitivity (1, 2). In addition, intracerebroventricular administration of GLP-1 was found to powerfully inhibit eating and drinking and alter body weight in fasted rats (3, 4). Furthermore, a recent study shows that peripheral GLP-1 also plays a role in regulating macronutrient selection and food intake (5). GLP-2 was initially identified as a growth factor for the small intestine (6). Recent observations indicate that GLP-2 and its receptor also possess an overlapping function with GLP-1 in regulating gastric emptying and controlling satiety (7, 8).

Numerous studies conducted in the past 15 years have identified more than a dozen transcription factors and signaling molecules that play roles in driving proglucagon gene expression (9–31). However, none of the identified transcription factors or signaling molecules has been found to be tissue type- or cell type-specific.

Protein kinase A (PKA) is able to stimulate proglucagon gene transcription and glucagon or GLP-1 synthesis in both primary and transformed intestinal endocrine cells as well as in rat primary pancreatic islet cultures (11–14, 17). However, deleting or mutating the CAMP-response element (CRE) in the proglucagon gene promoter only partially attenuates its response to PKA activation in the small intestinal proglucagon-producing cell line STC-1 (13, 17). This indicates that PKA may up-regulate proglucagon gene expression via a yet unidentified signaling pathway. Recently, PKA was found to phosphorylate and inactivate the serine/threonine kinase glycogen synthase kinase 3β (GSK-3β) in several cell lineages (32, 33). These observations prompted us to ask whether this newly identified function of PKA is engaged in proglucagon gene transcription. GSK-3β is a major mediator of the Wnt signaling pathway (34–36). In normal epithelial cells, adenosomatous polyposis coli together with GSK-3β and Axin bind to and phosphorylate β-catenin (β-cat), targeting β-cat for proteasomal mediated degradation. In embryonic cells, Wnt signals inactivate GSK-3β, resulting in free β-cat accumulation. Free
β-catenin then forms a bipartite transcription factor with a T cell factor (TCF/lymphoid enhancer factor (LEF), namely cat/TCF, activating the Wnt responsive or cat/TCF target genes (36)). Another notable inhibitor of GSK-3β is lithium, which mimics the function of the Wnt signals in embryonic cells (37, 38).

In this study, using cultivated intestinal endocrine cell lines as well as primary intestinal cells in culture, we examined whether Wnt signaling pathway/molecules mediate the effect of PKA in activating proglucagon gene transcription and GLP-1 synthesis.

**EXPERIMENTAL PROCEDURES**

**Materials**—Tissue culture medium and serum and oligonucleotides were purchased from Invitrogen. Radioisotopes were obtained from Amersham Biosciences. Forskolin, 3-isobutyl-1-methylxanthine (IBMX), 8-Br-cAMP, and 8-Br-cGMP were purchased from Sigma.

**Plasmids**—Construction of the wild type and mutant rat proglucagon/luciferase (LUC) reporter gene plasmids have been described previously (15, 17, 39). β-catenin expression plasmids were provided by Dr. Eric Fearon (40). The TCF-TK-LUC reporter gene construct (TOPFLASH) was provided by Dr. Bert Vogelstein (41). The G2-TK-LUC fusion gene was generated by inserting one copy of the rat proglucagon gene G2 enhancer-like element (9) into the TK-LUC plasmid. The DNA sequence for the top strain of G2 is AGGCACAAGAGTAAATAAAAAGTTTCCGGCCCTCTG (9). It contains a potential binding site (AAGTTTC) for the bipartite transcription factor cat/TCF (35).

**Cell Culture, Transfection, and LUC Reporter Gene Analysis**—The intestinal GLUTag, STC-1, and the pancreatic InR1-G9 cell lines were grown and maintained in Dulbecco’s modified Eagle’s medium supplemented with appropriate serum (39). To examine the effects of lithium and forskolin on proglucagon mRNA expression, GLP-1 synthesis, and secretion, cells were grown in the medium containing the appropriate serum overnight. 6 h prior to the experiment, serum-containing medium was withdrawn, and serum-free medium was added. For the LUC reporter gene analysis, the intestinal endocrine cell lines GLUTag and STC-1 were transfected using LipofectAMINE (Invitrogen) per the manufacturer’s instructions, whereas the InR1-G9 cell line was transfected by a method of calcium precipitation (42).

**Radioimmunoassay (RIA) for GLP-1**—As described previously (14, 31), peptides from the media were extracted by adding 1% (v/v) trifluoroacetic acid followed by passage twice through a cartridge of C18 SepPak. An RIA for immunoreactive GLP-1 was carried out using an antisera directed toward the C-terminal end of GLP-1 (Affinity Research Products Ltd., Mamhead, UK), which has previously been demonstrated to detect predominantly GLP-1<sup>-[3-3H]</sup> in GLUTag and FRIC cells (14).

**RESULTS**

**Lithium Activates Proglucagon mRNA Expression**—Lithium is a notable inhibitor of GSK-3β. We hypothesized that if PKA activates proglucagon gene transcription via inactivating GSK-3β, lithium may also activate proglucagon mRNA expression. We examined this hypothesis in the mouse large intestinal GLUTag (13, 45–47) and the mouse small intestinal STC-1 (14) cell lines, as well as in the primary (FRIC) rat intestinal cultures. After incubation with 10 μM LiCl for 4 h, both cell lines demonstrated ~3-fold increased proglucagon mRNA expression (Fig. 1A). In the GLUTag cells, the activation was still observable after 12 h (3.9-fold). After a 24-hour incubation in two experiments, we still observed an enhanced proglucagon mRNA expression. Results from one experiment are presented in Fig. 1A. In two other experiments, however, proglucagon mRNA expression returned to untreated levels by 24 h (data not shown). In the STC-1 cell line, activated proglucagon gene expression was substantial during the whole experimental procedure (Fig. 1B). Fig. 1A also shows that proglucagon mRNA expression in the GLUTag cells is activated by the PKA acti-
vators forskolin and IBMX as we demonstrated previously (14).
Similarly, an 8-h treatment of FRIC cultures with 10 mM LiCl
enhanced proglucagon mRNA expression 3-fold (Fig. 1C).

Lithium Activates the Rat Proglucagon Gene Promoter—The
effect of lithium on proglucagon mRNA levels could be a result
of enhanced transcription or reduced degradation or both. We
therefore examined whether lithium activates the proglucagon
gene promoter. Fig. 2A shows that, after being transfected into
the GLUTag cells, the expression of the −1.1-kb and the
−472-bp promoter constructs was activated 5-fold by forsko-
l in. The evolutionarily conserved CRE is located between −291
and −298 bp (9). When the wild type −302-bp promoter was
examined, −6-fold activation by forskolin was observed. How-
ever, even with the CRE-mutated [−302(M)] or deleted (−290)
reporter gene constructs, forskolin still generated −3−3.5-fold
activation. This is consistent with previous studies on the
STC-1 cell line (13, 17). LiCl treatment also activated all the
five promoter constructs examined by 2−3-fold, including those
that carry a mutated [−302(M)] or deleted (−290) CRE. Lith-
ium was also found to activate the proglucagon gene promoter
constructs when transfected into the STC-1 cell line (Fig. 2B).

The stimulatory effect of LiCl on the proglucagon promoter was
found to be dose-dependent (Fig. 2B) and was observable
within 4 to 12 h after the treatment (data not shown).

To determine whether lithium also activates proglucagon
gene expression in the pancreatic A cells, we conducted the
above analyses against the hamster pancreatic A cell line InR1-
G9. No appreciable activation on either proglucagon gene pro-
moter or endogenous proglucagon mRNA expression was ob-
served by LiCl treatment in this cell line (data not shown).

Lithium Stimulates GLP-1 Synthesis—We next examined
the effect of lithium on GLP-1 synthesis and secretion in the
GLUTag cell line and FRIC cultures. We demonstrated previ-
ously that GLP-1 synthesis and secretion in those cells could be
activated by forskolin or other PKA activators (11, 14, 44−47).
Incubating the GLUTag cells with 10 mM LiCl for 4 h did not
affect either the synthesis or secretion of GLP-1 (data not
shown). When the incubation time was extended to 8 h, a
1.7-fold increase in the GLP-1 content of cell was observed (p <
0.01, Fig. 3A). In the same period, no change in GLP-1 secretion
was observed as determined by an RIA of the cell free medium
(Fig. 3B). Similarly, FRIC cultures were treated without or
with 10 mM LiCl or 10 mM KCl (as control) for 8 h or 24 h before the assessment of GLP-1 synthesis and secretion. Consistent with the stimulatory effect of LiCl on the GLUTag cells, LiCl also increased GLP-1 synthesis in FRIC cultures to 1.5-fold of the control (p < 0.05) after a 24 h incubation (Fig. 4). No substantial effects of LiCl on GLP-1 synthesis were observed at 8 h or on GLP-1 secretion at either 8 or 24 h (Fig. 4).

**β-CAT Transfection Activates the Proglucagon Gene Promotor without a Functional CRE**—Lithium inactivates GSK-3β and induces free β-cat accumulation in other cell lineages (48). If lithium indeed stimulates proglucagon transcription via inactivation of GSK-3β, overexpressing β-cat should mimic the effect of lithium treatment. A reporter gene construct, namely TOPFLASH, has been widely utilized to examine the cat-TCF activity in colon cancer and other cell lines (41, 48–50). In this plasmid, the expression of a LUC reporter gene is driven by a minimum TK promoter fused with three copies of the TCF binding site. As shown in Fig. 5, this fusion gene is dose dependently activated by LiCl but not by KCl when transfected into the GLUTag cells.

We then directly examined the effect of the wild type and the constitutively active mutant β-cat, S33Y, on the expression of the proglucagon gene promoter. The S33Y mutant β-cat is resistant to degradation because it cannot be phosphorylated by GSK-3β (41). Co-transfection with the wild type β-cat cDNA stimulated all of the proglucagon gene promoter constructs examined, except for the −1.1-kb promoter construct, by 1.7–2.0-fold (Fig. 6). Compared with the wild type β-cat, the S33Y mutant β-cat activated all five constructs more effectively, varying from 2.5- to 4-fold (Fig. 6). In addition, the S33Y mutant β-cat was able to activate the −1.1 kb proglucagon gene promoter construct. The activation was also independent of the CRE motif on the proglucagon gene promoter (Fig. 6).

The consensus binding site for cat/TCF has been suggested to be WWGTTC (35). We localized several potential cat/TCF binding sites on the proglucagon gene promoter. One such binding site is downstream of −290 bp and is a part of the G2 enhancer-like element (9). This site is conserved among the...
Proglucagon Transcription by Lithium and β-Catenin

PKA Phosphorylates GSK-3β, and Both LiCl and PKA Activators Induce Free β-Cat Accumulation—Recent studies have shown that PKA phosphorylates and inactivates GSK-3β in the HEK293 and NIH 3T3 cell lines (32). In addition, PKA was also found to phosphorylate and inactivate GSK-3β in neuronal cells, and GSK-3β inactivation is linked to the inhibition of neuronal cell apoptosis (33). To further examine our hypothesis that Wnt pathway mediates the activation of proglucagon gene transcription by PKA, we examined GSK-3 phosphorylation and inactivation by PKA in the intestinal endocrine cell lines. As shown in Fig. 8, treating the GLUTag cells with forskolin or 8-Br-cAMP induced the phosphorylation of GSK-3α at serine 21 and GSK-3β at serine 9. In contrast, treating the GLUTag cells with 8-Br-cGMP did not change the phosphorylation status of GSK-3α or GSK-3β (Fig. 8).

We then asked whether lithium treatment or PKA activation would lead to free β-cat accumulation in the two intestinal proglucagon-producing cell lines. The amount of β-cat in cell membrane and cell cytosol was examined by Western blot analysis following a cellular fractionation procedure (43). In four separate experiments we observed free β-cat accumulation in response to either LiCl or forskolin treatment, with accumulation levels ranging from 2.5 to 10-fold. Data from one experiment are shown in Fig. 9. In this experiment, incubating the GLUTag cells with 10 mM LiCl for 2 h led to an ~3-fold increase in cytosolic β-cat, whereas the change in β-cat in the membrane portion was minimal. Similarly, treating the GLUTag cells with forskolin for 2 h led to a 2.9-fold increase in cytosolic β-cat accumulation, whereas the change of β-cat in the membrane portion was minimal (Fig. 9A). We also found that treatment of the GLUTag cells with 10 nM insulin for 2 h generated an appreciable effect on free β-cat accumulation (2.9-fold), whereas the effect at 2 or 4 h was minimal (Fig. 9A). Similar results regarding free β-cat accumulation in response to these three reagents were obtained in examining the STC-1 cell line (Fig. 9B).

DISCUSSION

In mammals, a single proglucagon gene encodes three major peptide hormones expressed in three different tissues (1). These hormones play critical roles in blood glucose homeostasis, the growth of small intestines, and satiety (1–8). Uniquely, different hormones encoded by the same proglucagon gene may possess opposite roles, such as glucagon versus GLP-1 in blood glucose homeostasis, or overlapping roles, such as GLP-1 and GLP-2 in mediating satiety (7, 8). Therefore, it is desirable and interesting to explore the molecular mechanisms that underlie proglucagon gene transcription and the biosynthesis of each individual hormone in a cell-specific manner. Although previous studies have identified numerous transcription factors and signals that may up-or down-regulate proglucagon gene transcription, none of the identified factors specifically regulate proglucagon transcription in pancreatic A cells only or in intestinal endocrine cells only (9–26, 51–53). Our observations indicated that lithium (as an inhibitory agent of GSK-3β) and β-cat/TCF selectively up-regulates proglucagon gene transcription and GLP-1 synthesis in the endocrine cells of the gut but not in a pancreatic A cell line. It will be interesting to explore the molecular mechanisms underlying this cell type-specific transactivation event.

A typical CRE element is located on both human and rat
proglucagon gene promoters (9, 17, 22). Previous studies in the mouse small intestinal STC-1 cell line have shown that deleting or mutating this CRE motif generates only partial attenuation in response to PKA activation (13, 17). We show here that this is also true for the GLUTag cell line. These observations further support our proposal that PKA stimulates proglucagon gene transcription not only via phosphorylation of the CRE-binding proteins (CREB) but also through pathways that cross-talk with PKA.

The cross-talk between the Wnt signaling pathway and PKA or G-protein-coupled receptors has been realized very recently. Meigs et al. (54) have shown that constitutively activated Ga12 and Ga13 interact with E-cadherin to cause the release of β-catenin and the subsequent stimulation of cat/TCF-mediated transcription. The chimeric receptor of the ligand binding and transmembrane domains of the β2 adrenergic receptor and the cytoplasmic domains of Frizzled-1 can also stimulate the cat/TCF transcriptional activation through a mechanism that appears to involve signaling through Gaq and/or Gα13 (55). Similarly, Fujino et al. (56) reported the phosphorylation of GSK-β and stimulated TCF-mediated transcription by prostaglandin E2 through the EP2 or EP4 receptor, likely via PKA and phosphatidylinositol 3-kinase, respectively. Furthermore, treating fibroblasts and neuronal cells with forskolin or 8-Br-cAMP may lead to enhanced phosphorylation and inactivation of GSK-3β (32, 33). Very recently, Yusta et al. (57) reported that GLP-2 inhibits cell apoptosis via its G protein-coupled receptor in association with PKA-dependent inactivation of GSK-3β. Here we extend these observations into the intestinal proglucagon-producing cell (Fig. 8). This, in combination with our observation that both lithium and forskolin activated the G2-TK fusion promoter (Fig. 7), supports our notion that PKA activates proglucagon gene transcription via cross-talk with the Wnt pathway. Previous studies have implicated G2 in regulating proglucagon gene expression via binding with members of the Foxa transcription factor family (20, 51–53). In addition, by examining the effect of membrane depolarization on proglucagon gene expression in pancreatic A cell lines, Furstenau et al. (23) identified a calcium-response element within this enhancer-like element (23). Further studies are needed to examine the binding of cat/TCF to G2 and determine how G2 is implicated in specifying cell type-specific regulation of proglucagon gene transcription in response to lithium and other signaling molecules.

Although phosphorylation and inactivation of GSK-3β by PKA, PKB/Akt, and PKC have been demonstrated in several cell lineages (32, 33, 57–62), whether this inactivation leads to free β-catenin accumulation and therefore induce the accumulation of free β-catenin, which will form a complex with a given TCF. The bipartite transcription factor cat/TCF then stimulates proglucagon mRNA transcription, possibly through interacting with the G2 enhancer-like element (9, 23). PKA is able to activate proglucagon gene transcription via the evolutionarily conserved CRE element on the proglucagon gene promoters (9, 11–14, 17). In addition, PKA may also activate proglucagon gene transcription by inactivating the GSK-3β and inducing the accumulation of free β-catenin. It is recognized however, that GSK-3β inactivation and free β-catenin accumulation in response to PKA could be two separate events. Glu, proglucagon gene. A dotted line shows the CRE pathway documented previously (11–14, 17).

Fig. 9. Both LiCl and forskolin stimulate free β-catenin accumulation in intestinal endocrine cell lines. GLUTag (A) and STC-1 (B) cells were grown in the absence or presence of 10 nm insulin (Ins), 10 mM LiCl, or 10 μM forskolin (For) plus 10 μM BMX for the indicated periods of time. Cytosolic and membrane β-catenin were examined by Western blot analysis as described under “Experimental Procedures.” The same membranes were stripped followed by hybridization with an anti-β-catenin antibody (loading control). After the densitometric analyses of the photograms, the effect of each chemical on β-catenin appearance in both cytosol and membrane fractions were calculated as the fold change versus the untreated cells normalized against β-actin.

Fig. 10. A diagram showing mechanisms for the regulation of proglucagon gene transcription by lithium and PKA. Lithium is able to inactivate GSK-3β and therefore induce the accumulation of free β-catenin, which will form a complex with a given TCF. The bipartite transcription factor cat/TCF then stimulates proglucagon mRNA transcription, possibly through interacting with the G2 enhancer-like element (9, 23). PKA is able to activate proglucagon gene transcription via the evolutionarily conserved CRE element on the proglucagon gene promoters (9, 11–14, 17). In addition, PKA may also activate proglucagon gene transcription by inactivating the GSK-3β and inducing the accumulation of free β-catenin. It is recognized however, that GSK-3β inactivation and free β-catenin accumulation in response to PKA could be two separate events. Glu, proglucagon gene. A dotted line shows the CRE pathway documented previously (11–14, 17).
serine 9 and inactivation of GSK-3β enzymatic activity in a number of epithelial cell lines. However, free β-cat levels were not altered in their study. In contrast, both Wnt and lithium induced free β-cat accumulation but did not affect the phosphorylation status of GSK-3β (58). Based on these observations, Ding et al. (58) hypothesized that insulin and the Wnt signals regulate GSK-3β through different mechanisms and therefore lead to a distinct downstream event and that the phosphorylation of GSK-3β at serine 9 may not be sufficient to induce free β-cat accumulation. In our study, we also found that treating the intestinal endocrine cell lines with 10 mM insulin for 2 h generated no effect on free β-cat accumulation. However, the effect began to be observable after 4 h and was enhanced after 12 h (Fig. 9). More importantly, we demonstrated that, in the intestinal proglucagon producing cells, forskolin treatment led to enhanced free β-cat accumulation within 2 h. We speculate that, in these particular endocrine cell lines, phosphorylation of GSK-3β at serine 9 by PKA may be sufficient to inactivate its ability to facilitate β-cat degradation. However, at this stage, we cannot eliminate the possibility that phosphorylation of GSK-3β by PKA and the accumulation of free β-cat in response to PKA are two independent events. Nevertheless, our results clearly indicate that PKA activates proglucagon gene transcription in the endocrine L cells at least in part through accumulation of free β-cat. A model illustrating our current understanding of the mechanisms underlying proglucagon gene activation by PKA and lithium is shown in Fig. 10.

Our results also provide a potential molecular mechanism for the similar effect of lithium and GLP-1 to suppress food and water intake (3, 4, 63–70). Scientists have observed for a number of years that peripheral administration of lithium in rats causes a spectrum of effects, including reduced food/water intake, decreased salt ingestion after sodium depletion, and induction of robust conditioned taste aversions (63–70). Our results also provide a potential molecular mechanism for the downstream target genes of cat-TCF or the Wnt signaling pathway. Our results provide a novel mechanism by which PKA up-regulates proglucagon gene transcription in the intestinal endocrine L cells and a potential explanation as to how lithium controls food/water intake. Additional studies are required to examine proglucagon gene transcription and GLP-1 synthesis in the brain in response to lithium and other Wnt signaling molecules.

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REFERENCES

1. Kieffer, T. J., and Habener, J. F. (1999) Endocr. Rev. 20, 876–913
2. Drucker, D. J. (2002) Gastroenterology 122, 531–544
3. Turton, M. D., O’Shea, D., Gunn, I., Heak, S. A., Edwards, C. M., Meeran, K., Choi, S. J., Taylor, G. M., Heath, M. M., Lambert, P. D., Wilding, J. P. H., Smith, D. M., Ghatel, M. A., Herbert, J., and Bloom, S. R. (1996) Nature 381, 69–72
4. Meeran, K., O’Shea, D., Edwards, C. M., Turton, M. D., Heath, M. M., Gunn, I., Abusamra, S., Rossi, M., Smal, C. J., Goldstone, A. P., Taylor, G. M., Sunter, D., Steele, J., Choi, S. J., Ghatel, M. A., and Bloom, S. R. (1999) Endocrinology 140, 2494–2500
5. Peters, C. T., Choi, Y. H., Brubaker, P. L., and Anderson, G. H. (2001) J. Nutr. 131, 2164–2170
6. Drucker, D. J., Erlich, P., Asa, S. L., and Brubaker, P. L. (1996) Proc. Natl. Acad. Sci. U. S. A. 93, 7911–7916
7. Lovshin, J., Estall, J., Yusta, B., Brown, T. J., and Drucker, D. J. (2001) J. Biol. Chem. 276, 21449–21452
8. Woldemihret, M., Wettergren, A., Hartmann, B., and Holst, J. J. (1998) Scand. J. Gastroenterol. 33, 828–832
9. Filippeg, J., Drucker, D. J., Koppel, W., Jepelal, L., Musilovin, Z., and Habener, J. F. (1988) Mol. Cell. Biol. 8, 4877–4888
10. Drucker, D. J., and Asa, S. (1988) J. Biol. Chem. 263, 13475–13478
11. Drucker, D. J., and Brubaker, P. L. (1989) Proc. Natl. Acad. Sci. U. S. A. 86, 3983–3987
12. Drucker, D. J., Campos, R., Reynolds, R., Stobie, K., and Brubaker, P. L. (1991) Endocrinology 128, 394–400
13. Gajic, D., and Drucker, D. J. (1993) Endocrinology 132, 1055–1062
14. Drucker, D. J., Jin, T., Asa, S., Young, T., and Brubaker, P. L. (1994) Mol. Endocrinol. 8, 1646–1655
15. Jin, T., and Drucker, D. J. (1995) Mol. Endocrinol. 9, 1306–1320
16. Wang, M., and Drucker, D. J. (1995) J. Biol. Chem. 270, 12646–12652
17. Lu, P., Jin, T., and Drucker, D. J. (1996) Endocrinology 137, 3710–3716
18. Hussain, M. A., Lee, J., Miller, C. P., and Habener, J. F. (1997) Mol. Cell. Biol. 17, 7186–7194
19. Cordier-Bussat, M., Bernard, C., Levenez, F., Klages, N., Laser-Ritz, B., Philippe, J., Chayvialle, J. A., and Cuber, J. C. (1998) Diabetes 47, 1038–1045
20. Kaestner, K. H., Katz, J., Liu, Y., Drucker, D. J., and Schultz, G. (1999) Genes Dev. 13, 495–504
21. BeimSchwe, S., Neubauer, A., Herzig, S., Grzeskowiak, R., Diedrich, T., Cierny, I., Schols, D., Aleje, T., and Knepe, W. (1999) Mol. Endocrinol. 13, 718–728
22. Nian, M., Drucker, D. J., and Irwin, D. (1999) Am. J. Physiol. 278, G829-G837
23. Purstenau, U., Schwanger, M., Blume, R., Jendrusch, E. M., and Knepe, W. (1999) J. Biol. Chem. 274, 5851–5860
24. Siemann, M., Blume, R., Grapentin, D., Oetjen, E., Schwanger, M., and Knepe, W. (1999) Mol. Pharmacol. 55, 1094–1100
25. Herzig, S., Fuzesi, L., and Knepe, W. (2000) J. Biol. Chem. 275, 27789–27799
26. Ritz-Laser, B., Estreich, A., Gauthier, B., and Philipp, J. (2000) J. Biol. Chem. 275, 32708–32715
27. Dhanvantari, S., Izzo, A., Jansen, E., and Brubaker, P. L. (2001) Endocrinology 142, 57–62
28. Xiao, Q., Boushey, R. P., Cino, M., Drucker, D. J., and Brubaker, P. L. (2000) Am. J. Physiol. Regul. Integr. Comp. Physiol. 278, R1057-R1063
29. Lee, Y. C., Asa, S. L., and Drucker, D. J. (1992) J. Biol. Chem. 267, 10705–10708
30. Efrati, S., Teitelman, G., Anwar, M., Ruggiero, D., and Hanahan, D. (1988) Neuron 1, 605–613
31. Attini, Y., Hansoti, B., and Brubaker, P. L. (2002) Endocrinology 143, 2420–2426
32. Fang, X., Yu, S. X., Lu, Y., Bast, R. C., Woodgett, J. R., and Mills, G. B. (2000) Proc. Natl. Acad. Sci. U. S. A. 97, 11960–11965
33. Li, M., Wang, X., Meinert, M. K., Laseng, T., Birnbaum, M. J., and Heidenreich, K. A. (2000) Mol. Cell. Biol. 20, 9356–9363
34. Kinzler, K. W., Nilbert, M. C., Su, L. K., Vogelstein, B., Bryan, T. M., Levy, D. H., Smith, K. J., Preisler, H. D., McKeanie, D., Finneer, R., Markham, A., Groffen, J., Boksski, M. S., Alscl, S. P., Fori, A., Ando, H., Miyoshi, Y., Miki, Y., Nishisyo, I., and Nakamura, Y. (1991) Science 253, 651–655
35. He, C. T., Spark, A. B., Rago, C., Hermskeng, H., Zawel, L., da Costa, L. T., Morin, P. J., Vogelstein, B., and Kinzler, K. W. (1998) Science 281, 1509–1512
36. van Es, J. H., Giles, R. H., and Clevers, H. C. (2001) Exp. Cell Res. 264, 126–134
37. Kao, R. K., and Elinson, R. P. (1998) Biol. Cell 90, 585–589
38. Salinas, P. C. (1999) Biochem. Soc. Symp. 65, 101–109
39. Jin, T., and Drucker, D. J. (1996) Mol. Cell. Biol. 16, 9–26
40. Kolligs, F. T., Hu, G., Dang, C. V., and Fearon, E. R. (1999) Mol. Cell. Biol. 19, 5696–5706
41. Morin, P. J., Sparks, A. B., Korinek, V., Barker, N., Clevers, H., Vogelstein, B., and Kinzler K. W. (1997) Science. 275, 1787–1790
42. Xu, F., Li, H., and Jin, T. (1999) Endocrinology 139, 4108–4114
43. Morin, P. J., Sparks, A. B., Korinek, V., Barker, N., Clevers, H., Vogelstein, B., and Kinzler K. W. (1997) Science. 275, 1787–1790
44. Brubaker, P. L., Lee, Y. C., and Drucker, D. J. (1997) Proc. Natl. Acad. Sci. U. S. A. 94, 15696–15703
45. Xu, F., Li, H., and Jin, T. (1999) J. Biol. Chem. 274, 34310–34316
46. Stambolic, V., Ruel, L., and Woodgett, J. R. (1996) Cell Growth & Differ. 8, 1349–1357
47. Brubaker, P. L., Lee, Y. C., and Drucker, D. J. (1992) J. Biol. Chem. 267, 20728–20732
48. Meigs, T. E., Fields, T. A., McKee, D. D., and Casey, P. J. (2001) Proc. Natl. Acad. Sci. U. S. A. 98, 5191–5196
49. Williams, J. M., and Woodgett, J. R. (1997) Mol. Endocrinol. 11, 313–321
50. van Dijk, G., and Thiele, T. E. (1999) Biochem. J. 338, 501–506
51. Gauthier, B. R., Schwitzgebel, V. M., Zaiko, M., Mamin, A., Ritz-Laser, B., and Philippe, J. (2002) Mol. Endocrinol. 16, 176–183
52. Gauthier, B. R., Schwitzgebel, V. M., Zaiko, M., Mamin, A., Ritz-Laser, B., and Philippe, J. (2002) Mol. Endocrinol. 16, 176–183
53. Diedrich, T., Furstenau, U., and Knepel, W. (1997) Biol. Chem. 378, 89–98
54. Meigs, T. E., Fields, T. A., McKee, D. D., and Casey, P. J. (2001) Proc. Natl. Acad. Sci. U. S. A. 98, 5191–5196
55. Liu, T., DeCostanzo, A. J., Liu, X., Wang, H., Hallagan, S., Moon, R. T., and Malbon, C. C. (2001) Science 292, 1718–1722
56. Fujino, H., West, K. A., and Regan, J. W. (2002) J. Biol. Chem. 277, 2614–2619
57. Yusta, B., Estall, J., and Drucker, D. J. (2002) J. Biol. Chem. 277, 24896–24906
58. Ding, W. W., Trends, H., and McCormick, F. (2000) J. Biol. Chem. 275, 323–3248
59. Chen, R. H., Ding, W. W., and McCormick, F. (2000) J. Biol. Chem. 275, 17894–17899
60. Cross, D. A., Alessi, D. R., Cohen, P., Andjelkovich, M., and Hemmings, B. A. (1995) Nature 378, 785–789
61. Goode, N., Hughes, K., Woodgett, J. R., and Parker P. J. (1992) J. Biol. Chem. 267, 16779–16882
62. Fang, X., Yu, S., Tanyi, J. L., Lu, Y., Woodgett, J. R., and Mills, G. B. (2002) Mol. Cell. Biol. 22, 2099–2110
63. Ritter, S., McGlone, J. J., and Kelley, K. W. (1980) Brain Res. 201, 501–506
64. McCann, M. J., Verbalis, J. G., and Stricker, E. M. (1989) Am. J. Physiol. 256, R463–R468
65. van Dijk, G., and Thiele, T. E. (1999) Peptides 33, 406–414
66. Montrose-Rafizadeh, C., Yang, H., Rodgers, B. D., Biday, A., Pritchette, L. A., and Eng, J. (1997) J. Biol. Chem. 272, 21201–21206
67. Thiele, T. E., Seeley, R. J., D’Alessio, D., Eng, J., Bernstein, I. L., Woods, S. C., and van Dijk, G. (1998) Brain Res. 801, 164–170
68. Rinaman, L. (1999) Am. J. Physiol. 277, R1557–R1564
69. Seeley, R. J., Blake, K., Rushing, P. A., Bensit, S., Eng, J., Woods, S. C., and D’Alessio, D. (2000) J. Neurosci. 20, 1616–1621
70. Rinaman, L. (1999) Am. J. Physiol. 277, R582–R590
71. Anini, Y., and Brubaker, P. L. (2003) Diabetes 52, in press
72. Rocca, A. S., and Brubaker, P. L. (1999) Endocrinology 140, 1687–1694
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