PANCREAPEDIA
Exocrine Pancreas Knowledge Base

APA AMERICAN PANCREATIC ASSOCIATION

ERK Activation and Its Role in Pancreatic Acinar Cell Function

John A. Williams and Bryan J. Holtz

Department of Molecular and Integrative Physiology, University of Michigan, Ann Arbor MI 48109

Email: jawillms@med.umich.edu

Version 1.0, September 12, 2017 DOI: 10.3998/panc.2017.06

1. ERK Belongs to the MAPK Family

The Extracellular Signal-Regulated Kinase (ERK) pathway is the best understood of the Mitogen Activated Protein Kinase (MAPK) cascades (29, 40). The pathways are named for the central kinase that affects cell function, ERK, JNK, p38 MAPK and ERK5. These MAPKs are synthesized by cytoplasmic ribosomes but can migrate into the nucleus when activated. Each MAPK pathway consists of a least three kinase components generically termed MAP3K, MAP2K and MAPK which sequentially activate the downstream component. For the ERK pathway these kinases are Raf, MEK and ERK (**Figure 1**). The ERK pathway is activated by growth factors, mitogens, hormones and some neurotransmitters which bind to tyrosine kinase and G protein coupled receptors. The JNK and p38 MAPK pathways are most often activated by cytokines and cell stress. At many levels of the three pathways there are multiple forms such as ERK1 and ERK2 (ERK1/2) and JNK1/2/3 (40). These multiple species are more closely related and in some cases have the same actions. For the ERK pathway Human ERK1 and ERK2 are 84% identical and all known stimuli activate both forms (3). By contrast, the p38 Map Kinase has four forms (α, β, γ, δ) which may have different regulation and actions. The MAPK pathways are often organized by scaffolding proteins (28, 42) For ERK the best studied scaffold is KSR1 (Kinase Suppressor of Ras-1) which binds all three members of the ERK kinase cascade. For JNK the cascade may be organized by the binding protein JIP-1. MAPK cascade components are all inactivated by phosphatases including pSer/Thr phosphatases such as pp2A, Tyr phosphatases or in the case of the MAPKs themselves by dual specificity phosphatases or DUSPS that dephosphorylate both Ser/Thr and Tyr (23, 26, 40). There are ten catalytically active DUSPS arranged in three families by their nuclear or cytosolic localization.

The MAPK cascades all have multiple actions in both the nucleus and cytosol. In the nucleus, ERK and its downstream effectors such as p90 Ribosomal S6 Kinase (RSK) phosphorylate ternary complex factors such as ELK-1 and thereby stimulate transcription of early response genes such as Fos and Egr1 and are important in initiating mitogenesis (43, 54). In the cytoplasm, ERK and another downstream kinase, MAPK-interacting protein kinase (MNK-1) phosphorylate specific translational factors including eIF4E and eIF4G as well as $cPLA_2$

(cytoplasmic phospholipase A2) ERK also localizes to other organelles including endosomes, caveolae, Golgi and cytoskeleton (53).

This review will first consider what is known about activation of ERK1/2 in pancreatic acinar cells and then cover what is known regarding the actions of ERK in this cell.

2. Activation of ERK pathway in Pancreatic Acinar Cells (Figure 1)

ERK activation is usually monitored by following the dual phosphorylation of the Thr and Tyr residues in the Thr-Glu-Tyr activation sequence brought about by MEK as there are a number of good phosphospecific antibodies directed at this epitope. It can also be shown by phosphorylation of myelin basic protein either in a test tube or by an in gel technique

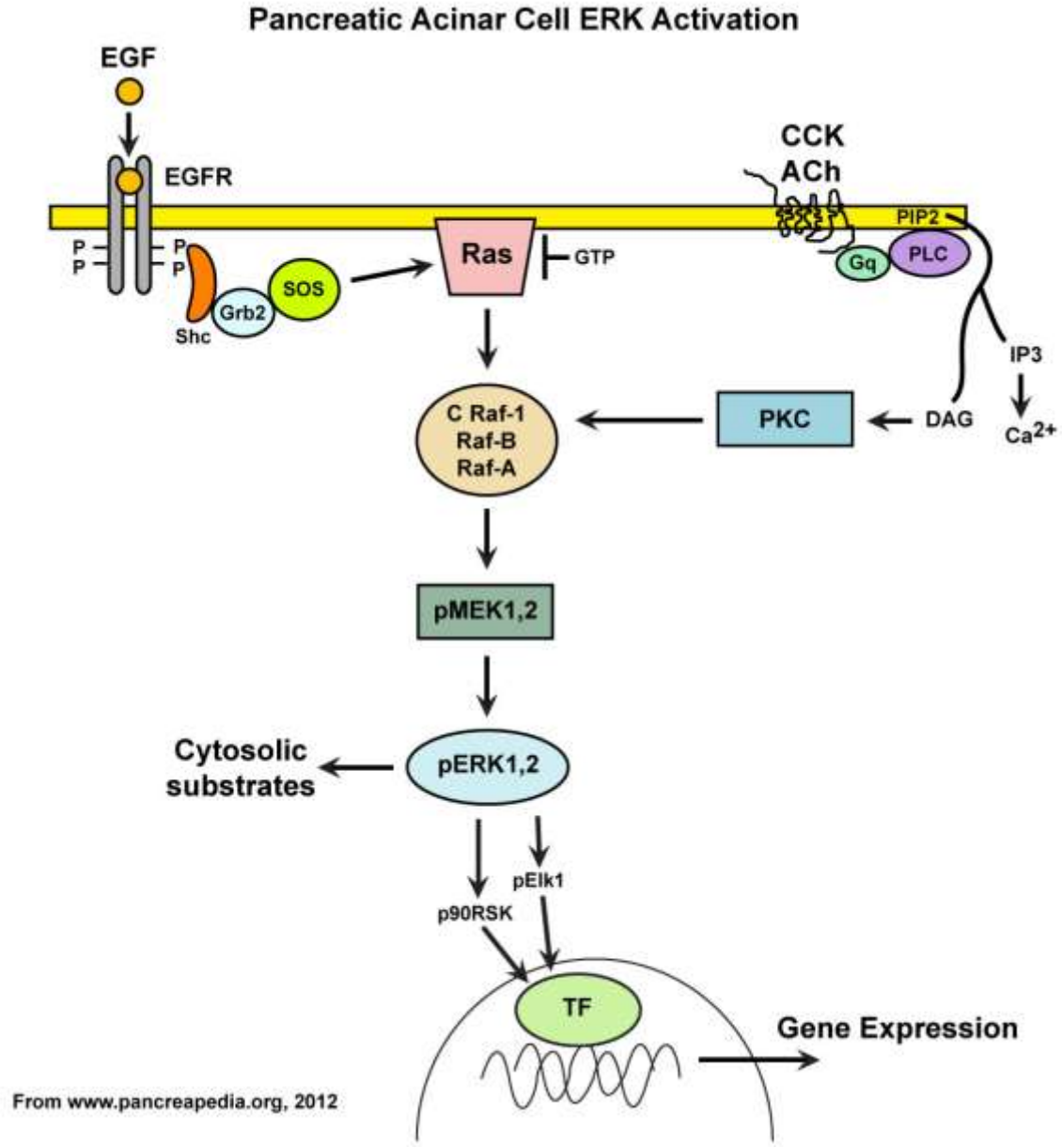

Figure 1. Pancreatic Acinar Cell ERK activation.

following gel electrophoresis and renaturization. Both Western blots and the in gel kinase procedure reveal the two forms of ERK at approximately 44 and 42 kDa; in fact, the molecules were originally referred to as p42 and p44 MAPK with p42 being what is now referred to as ERK2 and p44 now being ERK1. Using isolated rat or mouse pancreatic acini in vitro, ERK1/2 is activated by CCK, bombesin, substance P, and carbachol, all of which activate G protein coupled receptors coupled to G_q and calcium mobilization but not by secretin or VIP which activate receptors coupled to G_s and cAMP formation (10, 12, 15, 16, 41). By contrast, human acini show an increase in pERK in response to cholinergic agonist but not CCK although this was ascribed to the absence of CCK receptors (24). ERK1/2 is also activated by EGF, HGF, IGF-1 and other growth factors which activate Tyr kinase containing receptors (1, 10, 51). Most mechanistic studies have utilized CCK and EGF as they induce the most robust activation. Both of these agonists activate ERK1/2 within minutes in vitro with the response sustained for at least an hour. CCK effects are seen at 3 or 10 pM which is slightly higher than that required to mobilize Ca^{2+} or stimulate digestive enzyme secretion. TGF-β has also been shown to activate ERK1/2 in pancreatic acini with the effect mediated by Smad4 (46). In vivo, there is little change in phospho ERK between fasting and refeeding chow but phospho ERK shows a large increase after refeeding chow with trypsin inhibitor that increases plasma CCK to around 10 pM and induces adaptive pancreatic growth (18, 47, 48). In another type of growth, p42 and p44 MAPK were shown to be activated during pancreatic regeneration following partial pancreatectomy (33).

ERK appears to be activated in pancreatic acinar cells by the canonical pathway of RAF – MEK – ERK as RafA, RafB and c-Raf1 as well as MEK1 and MEK2 are all present and activated by CCK and EGF (15, 16). EGF activates this pathway by activating Ras and is not blocked by inhibiting Protein Kinase C (PKC) (10). Whether CCK activates Ras is unclear with different reports indicating activation (13, 16) or lack of activation (10). The two studies showing activation used high concentrations of CCK. In addition the effects of CCK to activate ERK were blocked by a PKC inhibitor but not by dominant negative Ras (9, 35). It appears that the CCK1 receptor and receptors for other agonists that activate Gq primarily activate PKC via Ca^{2+} and diacylglycerol (DAG) and thereby activate the ERK pathway and the ERK mediator RSK (2). In some other cell types a pertussis toxin (PTx) sensitive G protein is involved in ERK activation. In AR42J cells derived from a rat pancreatic tumor, PTx partially inhibited ERK activation in response to CCK, EGF and phorbol ester (37). However, this was shown to be due to disinhibition of adenylyl cyclase signaling. Gastrin or CCK2 receptors have also been shown to be able to activate ERK (9, 14). In contrast to rodent pancreatic acini, in AR42J cells the action of CCK to induce ERK activation was mediated by the CCK2 receptor, the tyrosine kinase Yes and transactivation of the EGFR (36). This transactivation is known to occur in some cells but in rat pancreatic acinar cells there has been no CCK induced EGFR activation observed (11). Another mechanism for ERK activation in some cells is through G protein coupled receptors kinases that phosphorylate the receptor which then binds β-arrestins which recruit other signaling molecules and mediate the prolonged activation of ERK (32). However, a β-arrestin mechanism has not

been described for pancreatic acini or the CCK1R.

There is only a little information on the inactivation of ERK in acinar cells. In one study, pancreatic DUSP mRNA levels were very low but DUSP 5, -6, and -10 were induced by caerulein hyperstimulation such that they could be considered early response genes (21).

3. Actions of ERK1/2 in pancreas cells

ERK1/2 as protein kinases have broad substrate specificity for Ser or Thr residues upstream from Pro and can phosphorylate a large number of proteins with 659 target sites listed in a recent compendium (49). There are also 296 reported ERK-interacting proteins whose function may thereby be influenced by ERK (50). These include a number of molecules known to be important in pancreatic function including Stim1 which regulates Ca^{2+} entry channels (38) and Raptor (4) which promotes activation of the mTOR complex 1 (TORC1). However, most of the cellular responses of ERK signaling cascade include proliferation, differentiation, angiogenesis, survival, and metastasis. Many of the experimental studies have used MEK inhibitors as MEK is the only known activator of ERK1/2. The first MEK inhibitor was PD98059 which is a flavone and highly insoluble in water (17). Another MEK inhibitor U0126 was discovered shortly thereafter and is slightly more potent. These inhibitors have been used for *in vitro* studies of ERK signaling but were not very useful for in vivo studies due to poor solubility and short half-life. This led to the development of PD325901 and later Trametinib (GSK1120212) which are longer acting and effective experimentally in vivo (45).

In pancreas, as in other cell types, most studies of ERK action have focused on growth, proliferation and regeneration as ERK1/2 is recognized as a master regulator of the cell cycle focused on the G1 to S phase transition (31). To study adaptive growth mediated by CCK, Holtz et al fed mice trypsin inhibitor and showed that both PD325901 and Trametinib blocked acinar cell mitogenesis and pancreatic growth (22). The drugs were effective when fed orally by gavage or mixed into food and a single bolus dose was effective for at least 12 hours. Moreover, the drugs had no effect on other signaling pathways including mTOR, JNK, and STAT3. Cell cycle proteins including cyclin D1, D3 and E as well as PCNA and BrdU incorporation into DNA were inhibited. In vitro, inhibiting ERK with U0126 or PD98059 blocked proliferation of acinar cell monolayer cultures (19).

ERK activation has also been shown to play a role in cytokine production by pancreatic acinar cells. In isolated mouse acinar cells, PD98059 decreased production of MCP-1, MCP1α and MIP-2 induced by Substance P (41) and in rat pancreatic fragments, PD98059 reduced production of TNF-α and IL-1β induced by cerulein (44). The stimulation of cytokine production in both cases involved AP-1 transcription factor that is also blocked by inhibitors of JNK. EGR-1 is another early response gene whose expression in AR42J cells was shown to be blocked by ERK inhibitor PD98059 or overexpression of DUSP-1 (MKP-1) (25). MEK inhibition by either targeted shRNA or Trametanib has also been shown to reduce inflammatory cytokines in cerulein induced

chronic pancreatitis (20). Some of this activation of MAPKs may be mediated by reactive oxygen species as hydrogen peroxide and menadione strongly activated all three MAPKs and the activation by CCK was reduced by antioxidants (8).

4. ERK1/2 and Pancreatic Disease

Active ERK1/2 has been observed in both pancreatitis and pancreatic cancer and localized to acinar cells, inflammatory cells and PanINs, the precursor lesion to PDAC (Pancreatic ductal adenocarcinoma). Early studies evaluated the role of ERK1/2 in acute pancreatitis utilized PD 98059 and U0126 dissolved in DMSO that was administered IP to rats or mice; in both studies modest to moderate inhibition of cerulein-induced pancreatitis was observed (5, 30, 34). Inhibition of ERK in isolated acinar cells with PD98059 blocked the upregulation of the Neurokinin 1 receptor induced by cerulein (27). More recent studies in vivo using the much longer acting and water soluble inhibitors PD 325901 or Trametanib both of which block ERK activity by the oral route had no effect on acute pancreatitis but could reverse chronic pancreatitis (6, 20). The ERK inhibitors also prevented the regeneration that occurs after pancreatitis through mitogenesis of acinar cells. These studies are complicated by the fact that ERK is present in more than one cell type. The evidence is clearer for a role in JNK in acute pancreatitis with ROS being one cause of activation.

Studies in PDAC are clearer as in mouse models with active Ras or with chronic pancreatitis, inhibition of ERK by PD 325901 or targeted shRNA to MEK prevented the development of ADM (Acinar Ductal Metaplasia) and PanINs (6, 20). ERK activation has also been reported to play a role in epithelial to mesenchymal transition induced by TGF-β (39). MEK inhibitors block growth of some but not all PDAC derived cell lines (7). Unfortunately, MEK inhibitors by reducing ERK feedback on Ras signaling have also activated other pathways including PI-3K - Akt which serve to maintain carcinogenesis (52). Current clinical trials of MEK inhibitors also often include comparison to combined therapy with both ERK and AKT inhibitors.

5. References

1. **Aparicio IM, Garcia-Marin LJ, Andreolotti AG, Bodega G, Jensen RT and Bragado MJ**. Hepatocyte growth factor activates several transduction pathways in rat pancreatic acini. *Biochim Biophys Acta* 1643(1-3): 37-46,2003. PMID: 14654226.
2. **Bragado MJ, Dabrowski A, Groblewski GE and Williams JA**. CCK activates p90rsk in rat pancreatic acini through protein kinase C. *Am J Physiol* 272(3 Pt 1): G401-407,1997. PMID: 9124559.
3. **Busca R, Pouyssegur J and Lenormand P**. ERK1 and ERK2 Map Kinases: Specific Roles or Functional Redundancy? *Front Cell Dev Biol* 4: 53,2016. PMID: 27376062.
4. **Carriere A, Romeo Y, Acosta-Jaquez HA, Moreau J, Bonneil E, Thibault P, et al.** ERK1/2 phosphorylate Raptor to promote Ras-dependent activation of mTOR complex 1 (mTORC1). *J Biol Chem* 286(1): 567-577,2011. PMID: 21071439.
5. **Clemons AP, Holstein DM, Galli A and Saunders C**. Cerulein-induced acute pancreatitis in the rat is significantly ameliorated by treatment with MEK1/2 inhibitors U0126 and PD98059. *Pancreas* 25(3): 251-259,2002. PMID: 12370536.
6. **Collins MA, Yan W, Sebolt-Leopold JS and Pasca di Magliano M**. MAPK signaling is required for dedifferentiation of acinar cells and development of pancreatic intraepithelial neoplasia in mice. *Gastroenterology* 146(3): 822-834 e827,2014. PMID: 24315826.

7. **Collisson EA, Trejo CL, Silva JM, Gu S, Korkola JE, Heiser LM, et al.** A central role for RAF-->MEK-->ERK signaling in the genesis of pancreatic ductal adenocarcinoma. *Cancer Discov* 2(8): 685-693,2012. PMID: 22628411.
8. **Dabrowski A, Boguslowicz C, Dabrowska M, Tribillo I and Gabryelewicz A**. Reactive oxygen species activate mitogen-activated protein kinases in pancreatic acinar cells. *Pancreas* 21(4): 376-384,2000. PMID: 11075992.
9. **Dabrowski A, Detjen KM, Logsdon CD and Williams JA**. Stimulation of both CCK-A and CCK-B receptors activates MAP kinases in AR42J and receptor-transfected CHO cells. *Digestion* 58(4): 361-367,1997. PMID: 9324163.
10. **Dabrowski A, Groblewski GE, Schafer C, Guan KL and Williams JA**. Cholecystokinin and EGF activate a MAPK cascade by different mechanisms in rat pancreatic acinar cells. *Am J Physiol Cell Physiol* 273(5 Pt 1): C1472-1479,1997. PMID: 9374631.
11. **Dabrowski A, VanderKuur JA, Carter-Su C and Williams JA**. Cholecystokinin stimulates formation of shc-grb2 complex in rat pancreatic acinar cells through a protein kinase C-dependent mechanism. *J Biol Chem* 271(43): 27125-27129,1996. PMID: 8900204.
12. **Daniluk J and Dabrowski A**. The effect of concomitant stimulation with cholecystokinin and epidermal growth factor on extracellular signal-regulated kinase (ERK) activity in pancreatic acinar cells. *J Physiol Pharmacol* 58(3): 441-453,2007. PMID: 17928641.
13. **Daniluk J, Liu Y, Deng D, Chu J, Huang H, Gaiser S, et al.** An NF-kappaB pathway-mediated positive feedback loop amplifies Ras activity to pathological levels in mice. *J Clin Invest* 122(4): 1519-1528,2012. PMID: 22406536.
14. **Daulhac L, Kowalski-Chauvel A, Pradayrol L, Vaysse N and Seva C**. Src-family tyrosine kinases in activation of ERK-1 and p85/p110-phosphatidylinositol 3-kinase by G/CCKB receptors. *J Biol Chem* 274(29): 20657-20663,1999. PMID: 10400698.
15. **Duan RD and Williams JA**. Cholecystokinin rapidly activates mitogen-activated protein kinase in rat pancreatic acini. *Am J Physiol* 267(3 Pt 1): G401-408,1994. PMID: 7943237.
16. **Duan RD, Zheng CF, Guan KL and Williams JA**. Activation of MAP kinase kinase (MEK) and Ras by cholecystokinin in rat pancreatic acini. *Am J Physiol Gastrointest Liver Physiol* 268(6 Pt 1): G1060-1065,1995. PMID: 7611406.
17. **Dudley DT, Pang L, Decker SJ, Bridges AJ and Saltiel AR**. A synthetic inhibitor of the mitogen-activated protein kinase cascade. *Proc Natl Acad Sci U S A* 92(17): 7686-7689,1995. PMID: 7644477.
18. **Guo L, Sans MD, Gurda GT, Lee SH, Ernst SA and Williams JA**. Induction of early response genes in trypsin inhibitor-induced pancreatic growth. *Am J Physiol Gastrointest Liver Physiol* 292(2): G667-677,2007. PMID: 17095753.
19. **Guo L, Sans MD, Hou Y, Ernst SA and Williams JA**. c-Jun/AP-1 is required for CCK-induced pancreatic acinar cell dedifferentiation and DNA synthesis in vitro. *Am J Physiol Gastrointest Liver Physiol* 302(12): G1381-1396,2012. PMID: 22461029.
20. **Halbrook CJ, Wen HJ, Ruggeri JM, Takeuchi KK, Zhang Y, di Magliano MP, et al.** Mitogen-activated Protein Kinase Kinase Activity Maintains Acinar-to-Ductal Metaplasia and Is Required for Organ Regeneration in Pancreatitis. *Cell Mol Gastroenterol Hepatol* 3(1): 99-118,2017. PMID: 28090569.
21. **Hofken T, Keller N, Fleischer F, Goke B and Wagner AC**. Map kinase phosphatases (MKP's) are early responsive genes during induction of cerulein hyperstimulation pancreatitis. *Biochem Biophys Res Commun* 276(2): 680-685,2000. PMID: 11027531.
22. **Holtz BJ, Lodewyk KB, Sebolt-Leopold JS, Ernst SA and Williams JA**. ERK activation is required for CCK-mediated pancreatic adaptive growth in mice. *Am J Physiol Gastrointest Liver Physiol* 307(7): G700-710,2014. PMID: 25104499.
23. **Huang CY and Tan TH**. DUSPs, to MAP kinases and beyond. *Cell Biosci* 2(1): 24,2012. PMID: 22769588.
24. **Ji B, Bi Y, Simeone D, Mortensen RM and Logsdon CD**. Human pancreatic acinar cells lack functional responses to cholecystokinin and gastrin. *Gastroenterology* 121(6): 1380-1390,2001. PMID: 11729117.
25. **Kaufmann A, Rossler OG and Thiel G**. Expression of the transcription factor Egr-1 in pancreatic acinar cells following stimulation of cholecystokinin or Galphaq-coupled designer receptors. *Cell Physiol Biochem* 33(5): 1411-1425,2014. PMID: 24853800.
26. **Kidger AM and Keyse SM**. The regulation of oncogenic Ras/ERK signalling by dual-specificity mitogen activated protein kinase phosphatases (MKPs). *Semin Cell Dev Biol* 50: 125-132,2016. PMID: 26791049.
27. **Koh YH, Tamizhselvi R and Bhatia M**. Extracellular signal-regulated kinase 1/2 and c-Jun NH2-terminal kinase, through nuclear factor-kappaB and activator protein-1, contribute to caerulein-induced expression of substance P and neurokinin-1 receptors in pancreatic acinar cells. *J Pharmacol Exp Ther* 332(3): 940-948,2010. PMID: 20007404.

28. **Kolch W**. Coordinating ERK/MAPK signalling through scaffolds and inhibitors. *Nat Rev Mol Cell Biol* 6(11): 827-837,2005. PMID: 16227978.
29. **Krishna M and Narang H**. The complexity of mitogen-activated protein kinases (MAPKs) made simple. *Cell Mol Life Sci* 65(22): 3525-3544,2008. PMID: 18668205.
30. **Mazzon E, Impellizzeri D, Di Paola R, Paterniti I, Esposito E, Cappellani A, et al.** Effects of mitogen-activated protein kinase signaling pathway inhibition on the development of cerulein-induced acute pancreatitis in mice. *Pancreas* 41(4): 560-570,2012. PMID: 22228051.
31. **Meloche S and Pouyssegur J**. The ERK1/2 mitogen-activated protein kinase pathway as a master regulator of the G1- to S-phase transition. *Oncogene* 26(22): 3227-3239,2007. PMID: 17496918.
32. **Miller WE and Lefkowitz RJ**. Expanding roles for beta-arrestins as scaffolds and adapters in GPCR signaling and trafficking. *Curr Opin Cell Biol* 13(2): 139-145,2001. PMID: 11248546.
33. **Morisset J, Aliaga JC, Calvo EL, Bourassa J and Rivard N**. Expression and modulation of p42/p44 MAPKs and cell cycle regulatory proteins in rat pancreas regeneration. *Am J Physilo Cell Physiol* 277(5 Pt 1): G953-959,1999. PMID: 10564100.
34. **Namkung W, Yoon JS, Kim KH and Lee MG**. PAR2 exerts local protection against acute pancreatitis via modulation of MAP kinase and MAP kinase phosphatase signaling. *Am J Physiol Gastrointest Liver Physiol* 295(5): G886-894,2008. PMID: 18755806.
35. **Nicke B, Tseng MJ, Fenrich M and Logsdon CD**. Adenovirus-mediated gene transfer of RasN17 inhibits specific CCK actions on pancreatic acinar cells. *Am J Physiol Gastrointest Liver Physiol* 276(2 Pt 1): G499-506,1999. PMID: 9950825.
36. **Piiper A, Elez R, You SJ, Kronenberger B, Loitsch S, Roche S, et al.** Cholecystokinin stimulates extracellular signal-regulated kinase through activation of the epidermal growth factor receptor, Yes, and protein kinase C. Signal amplification at the level of Raf by activation of protein kinase Cepsilon. *J Biol Chem* 278(9): 7065-7072,2003. PMID: 12496267.
37. **Piiper A, Gebhardt R, Kronenberger B, Giannini CD, Elez R and Zeuzem S**. Pertussis toxin inhibits cholecystokinin- and epidermal growth factor-induced mitogen-activated protein kinase activation by disinhibition of the cAMP signaling pathway and inhibition of c-Raf-1. *Mol Pharmacol* 58(3): 608-613,2000. PMID: 10953055.
38. **Pozo-Guisado E, Campbell DG, Deak M, Alvarez-Barrientos A, Morrice NA, Alvarez IS, et al.** Phosphorylation of STIM1 at ERK1/2 target sites modulates store-operated calcium entry. *J Cell Sci* 123(Pt 18): 3084-3093,2010. PMID: 20736304.
39. **Principe DR, Diaz AM, Torres C, Mangan RJ, DeCant B, McKinney R, et al.** TGFbeta engages MEK/ERK to differentially regulate benign and malignant pancreas cell function. *Oncogene* 36(30): 4336-4348,2017. PMID: 28368414.
40. **Raman M, Chen W and Cobb MH**. Differential regulation and properties of MAPKs. *Oncogene* 26(22): 3100-3112,2007. PMID: 17496909.
41. **Ramnath RD, Sun J, Adhikari S and Bhatia M**. Effect of mitogen-activated protein kinases on chemokine synthesis induced by substance P in mouse pancreatic acinar cells. *J Cell Mol Med* 11(6): 1326-1341,2007. PMID: 18205703.
42. **Ramos JW**. The regulation of extracellular signal-regulated kinase (ERK) in mammalian cells. *Int J Biochem Cell Biol* 40(12): 2707-2719,2008. PMID: 18562239.
43. **Roskoski R, Jr.** ERK1/2 MAP kinases: structure, function, and regulation. *Pharmacol Res* 66(2): 105-143,2012. PMID: 22569528.
44. **Samuel I, Zaheer A and Fisher RA**. In vitro evidence for role of ERK, p38, and JNK in exocrine pancreatic cytokine production. *J Gastrointest Surg* 10(10): 1376-1383,2006. PMID: 17175457.
45. **Sebolt-Leopold JS and Bridges AJ**. Road to PD0325901 and beyond: The MEK Inhibitor Quest. *Kinase Inhibitor Drugs*. Hoboken, NJ, John Wiley & Sons, Inc.**:** 203-227, 2009.
46. **Simeone DM, Zhang L, Graziano K, Nicke B, Pham T, Schaefer C, et al.** Smad4 mediates activation of mitogen-activated protein kinases by TGF-beta in pancreatic acinar cells. *Am J Physiol Cell Physiol* 281(1): C311-319,2001. PMID: 11401854.
47. **Tashiro M, Dabrowski A, Guo L, Sans MD and Williams JA**. Calcineurin-dependent and calcineurin-independent signal transduction pathways activated as part of pancreatic growth. *Pancreas* 32(3): 314-320,2006. PMID: 16628088.
48. **Tashiro M, Samuelson LC, Liddle RA and Williams JA**. Calcineurin mediates pancreatic growth in protease inhibitor-treated mice. *Am J Physiol Gastrointest Liver Physiol* 286(5): G784-790,2004. PMID: 14684381.
49. **Ünal EB, Uhlitz F and Blüthgen N**. A compendium of ERK targets. *FEBS Lett* 591(17): 2607-2615,2017. PMID: 28675784.

50. **von Kriegsheim A, Baiocchi D, Birtwistle M, Sumpton D, Bienvenut W, Morrice N, et al.** Cell fate decisions are specified by the dynamic ERK interactome. *Nat Cell Biol* 11(12): 1458-1464,2009. PMID: 19935650.
51. **Watanabe H, Saito H, Rychahou PG, Uchida T and Evers BM**. Aging is associated with decreased pancreatic acinar cell regeneration and phosphatidylinositol 3-kinase/Akt activation. *Gastroenterology* 128(5): 1391-1404,2005. PMID: 15887120.
52. **Won JK, Yang HW, Shin SY, Lee JH, Heo WD and Cho KH**. The crossregulation between ERK and PI3K signaling pathways determines the tumoricidal efficacy of MEK inhibitor. *J Mol Cell Biol* 4(3): 153-163,2012. PMID: 22561840.
53. **Yao Z and Seger R**. The ERK signaling cascade--views from different subcellular compartments. *Biofactors* 35(5): 407-416,2009. PMID: 19565474.
54. **Yoon S and Seger R**. The extracellular signal-regulated kinase: multiple substrates regulate diverse cellular functions. *Growth Factors* 24(1): 21-44,2006. PMID: 16393692.