**CLINICAL VIGNETTE**

# Treatment of Pancreatic Cancer: Where Are We Now?

Tina Roosta, Shahryar Ashouri, M.D.

Pancreatic adenocarcinoma was the 10th most common cause of cancer in 2010 with an estimated incidence of 43,140 in the United States. However, what is more striking is that estimated deaths reached 36,800, making it the 4th most common cause of cancer death in 2010[1]. This disproportional mortality reflects treatment inefficacy for pancreatic cancer. In fact, compared to the other four major causes of cancer-related death—lung, colon, breast, and prostate— whose death rates are declining, pancreatic cancer death rates have remained relatively stable. What adds to this challenge is that less than 20% of patients with pancreatic cancer present with localized disease at the time of diagnosis[2]. According to data from the Surveillance, Epidemiology, and End Results (SEER) registry, 8% are localized, 25.9% have regionally spread, 50.5% have metastatic disease, and 15.5% are unstaged at time of diagnosis[3]. With the advent of gemcitabine in 1996, five-year survival for pancreatic cancer patients has modestly improved from 3% to 6%. However, the prognosis remained poor with a median overall survival of 3.5 months and with only 5.3% of patients alive at the 3-year mark from 1998-2005[3]. However, there have been strides to improve treatment by focusing on the molecular pathogenesis of pancreatic adenocarcinoma.

It has become widely known that pancreatic cancer arises from a dynamic interplay between tumor epithelial cells and tumor stromal cells mediated by multiple signaling pathways. Such pathways include but are not limited to: KRAS, CDKN2A, TGF-B, TP53, EGFR, and NOTCH path-ways[4]. Recently, gemcitabine has been supplemented by other therapies, such as EGFR inhibitors like erlotinib. Although the survival benefit was modest from this addition, it paved the way for the initiation of other clinical trials using this molecular approach. Concomitantly, studies are being undertaken to identify predictive markers to measure sensitivities to established drugs (gemcitabine and erlotnib) in order to stratify which patients will receive the most benefit from a given drug depending on their tumor profile.

This review will summarize the trials that have led to the current standard of therapy, highlight the most important signaling pathways along with the in vitro and in vivo trials that have attempted to target these pathways, and conclude by looking at the development of predictive markers with current treatment regimens.

Many clinical trials have been conducted to evaluate adjuvant treatment regimens for pancreatic cancer. In 1997, a phase III randomized control trial[5] compared gemcitabine to 5-FU, demonstrating an increased overall median survival and increased 12-month survival rate in the gemcitabine group versus 5-FU group (5.7 months vs. 4.4 months and 18% vs. 2% respectively). This pivotal trial brought gemcitabine in the limelight as the standard of therapy over 5-FU in the adjuvant setting. Subsequently, in 2001-2004, the ESPAC-1 trial[6] examined the efficacy of adjuvant 5-FU chemotherapy versus chemoradiotherapy versus observation alone. This multicenter trial showed a significant survival benefit for those who received adjuvant chemotherapy compared to those who did not—21% vs. 8% respectively. It also demonstrated a deleterious effect on survival in those patients who received chemoradiotherapy, conferring a five-year survival rate of 10% vs. 20% in the chemoradiotherapy group vs. control group, respectively.

The ESPAC-1 trial stimulated further study of adjuvant chemotherapy. In 2008, the CONKO-001 trial[7] randomized 368 patients to receive adjuvant chemotherapy with gemcitabine vs. observation alone. The results showed a statistically significant difference in median disease free survival in the gemcitabine arm vs. observation arm (13.4 months vs. 6.9 months), longer median survival (22.8 mos vs. 20.2 mos), and overall 5-year survival (21% vs. 9%). Following this, two studies, the RTOG 97-04 and ESPAC-3 trial, sought to compare gemcitabine to 5-FU in order to confirm or negate the original 1997 clinical trial attesting to gemcitabine's superiority. In the RTOG 97-04 trial[8], 451 patients were randomized to either 5-FU or gemcitabine for three weeks prior to chemoradiation and 12 weeks after chemoradiation

therapy. The results showed a median survival of 20.5 months and 3-year survival of 31% vs. a median survival of 16.9 months and 3-year survival of 22% in the gemcitabine arm vs. 5-FU arm, respectively (p=.05). Furthermore, in the ESPAC-3 trial[9], 1,088 patients were randomized post-resection to receive gemcitabine or 5-FU. The median overall survival was 23.6 months vs. 23.0 months in the gemcitabine and 5-FU group, respectively and so, did not demonstrate a significant difference.

Although the trials delineated above are inconsistent with respect to chemoradiotherapy and which chemotherapy is most effective, the National Comprehensive Cancer Network guidelines recommended that patients receive systemic gemcitabine followed by 5-FU chemoradiation or systemic chemotherapy alone with gemcitabine, 5-FU, or capecitabine[7]. However, in 2010, a phase III trial randomized 360 patients to gemcitabine vs. FOLFIRINOX (5FU/leucovorin, irinotecan, oxaliplatin) to assess overall survival in metastatic pancreatic patients. The FOLFIRINOX group displayed significantly longer overall survival, progression-free survival, better response rate, along with more treatment-related toxicity than the gemcitabine arm[10]. Although this trial demonstrated a treatment regimen more effective than gemcitabine, predictive markers are needed to determine which patients will benefit more from this aggressive approach.

Currently, the molecular pathogenesis of pancreatic adenocarcinoma has been a primary focus, leading to clinical trials involving targeted therapies. With the completion of genomic sequencing of pancreatic adenocarcinomas (sample size of 28), a total of 275 different genes have been found to be mutated. Of these 275 mutations, only 39 were repeated and only four of these genes (KRAS, CDKN2A, TP53, SMAD4) had a prevalence rate greater than 7%, posing a challenge to targeted therapies[4]. We know that pancreatic adenocarcinoma arises from ductal epithelium. However, it has been more recently emphasized that the invasive progression of this tumor is not solely due to the tumor cells alone; in fact, there is a dynamic interplay between the tumor cells and the surrounding stromal cells through multiple signaling pathways[4]. Understanding the dual compartment model of pancreatic cancer offers insight into alterations to the molecular framework that drive the tumor compartment, stromal compartment, or both.

We will focus on the following important signaling pathways and related in vivo and in vitro trials: KRAS, EGFR, TGF-B, CDKN2A, TP53, and NOTCH pathways. The KRAS oncogene is a membrane-bound protein with GTP-ase activity that affects multiple signaling pathways involved in cell-cycle progression, cytoskeletal alterations, and inhibition of apoptosis[11]. Mutations in KRAS occur early on in pancreatic cancer and appear to be a key driver in tumorigenesis; in fact, about ninety percent of pancreatic adenocarcinomas have a mutation in the KRAS oncogene. Mouse models with mutant forms of KRAS[12] demonstrated an increase in number and grade of neoplastic lesions.

Unfortunately, attempts at targeting KRAS have been unsuccessful; however, it may be a predictive marker for other targeted therapies, which will be discussed below. The epidermal growth factor receptor (EGFR) family is a group of tyrosine kinase receptors—Her1 (ErbB-1), Her2 (ErbB-2), Her3 (ErbB-3), and Her4 (ErbB-4)—which bind ligands such as epidermal growth factor (EGF) and transforming growth factor-a (TGF-a). Binding of the ligands results in downstream signaling of multiple pathways, such as Ras, phos-phoinositol-3 kinase, and phospholipase C. In pancreatic cancer cells lines, amplification of ErbB-1 and ErbB-2 genes occur most frequently, whereas there is little expression of this gene in normal pancreatic ductal epithelium[13]. It is thought that alterations in this receptor affect the tumor compart-ment, while overexpression of the ligands (EGF and TGF-a) affects the stromal compartment. In fact, it is not only the tumor cells that overproduce these ligands, it is also the desmoplastic cells in the stromal compartment[14]. From these findings, clinical trials were conducted with multiple EGFR inhibitors. In 2007, a phase III clinical trial investigated the efficacy of dual therapy with gemcitabine and cetuximab, an anti-EGFR monoclonal antibody used in the treatment of colon cancer, versus gemcitabine alone against locally advanced or metastatic pancreatic cancer[15]. The median survival was 6.5 months in the gemcitabine-cetuximab arm and 6 months in the gemcitabine arm, yielding no significant benefit in overall survival with the addition of cetuximab[15]. Similarly in 2007, another phase III trial was conducted to determine the efficacy of dual therapy with gemcitabine and erlotinib, an anti-ErbB-1/Her1 drug, versus gemcita-bine alone. The patients in the gemcitabine-erlotinib arm had an increased overall survival, one-year survival (23% vs. 17%), and progression-free survival[16]. This study was groundbreaking, as it was the first to find adding an agent to gemcitabine improved survival. In 2011, a random-

ized phase II trial looked at the addition of yet another EGFR inhibitor panitumumab to the gemcitabine-erlotinib treatment regimen to examine whether it had superiority over the gemcitabine-erlotinib regimen.

The progression-free survival was 3.3 months in the panitumumab-gemcitabine-erlotinib arm vs. 2.0 months in the gemcitabine-erlotinib arm, demonstrating overall survival benefit with dual inhibition of the EGFR pathway[3]. Yet another signaling pathway involved in pancreatic cancer is the transforming growth factor-B (TGF-B) pathway. It is activated through binding of serine-threonine receptors, which in turn phosphorylate receptor-mediated SMADS (SMAD, SMAD2, SMAD3, SMAD4). These receptor-mediated SMADS translocate to the nucleus and transcribe genes involved in cell-cycle progression. The most common mutation is that of SMAD4 (DPC4), deleted in over fifty percent of pancreatic adeno-carcinomas[17,18]. Loss of SMAD4 appears late in the progression of pancreatic cancer, facilitating the epithelial-to-mesenchymal transition which defines the more invasive and aggressive phenotype[19-20]. The TGF-B pathway also classically fits into the dual compartment model of pancreatic cancer, having effects on both tumor epithelial cells and the stromal cells. In fact, a transgenic mouse model with TGF-B overexpression demonstrated increased collagen synthesis by pancreatic stromal cells, resulting in a desmoplastic reaction[21].

Additionally, it appears that the loss of SMAD4 in combination with KRAS mutation results in tumorigenesis[22]. Bardeesy et al. created a mouse model with deletion of SMAD4 alone vs. deleted SMAD4 and KRAS mutation[22]. The model with deleted SMAD4 alone did not have carcinogenic properties; however, the deleted SMAD4 on KRAS mutation showed an accelerated growth of tumor both in the tumor and stromal compartments[22]. This study suggests that cross-talk occurs between these multiple signaling pathways, supporting a multi-targeted approach. In 2012, a mouse model was created to test this hypothesis by targeting both EGFR and TGF-B pathways, resulting in effective tumor inhibition and suggesting that this mechanism of treatment could be an upcoming therapeutic approach[23].

Furthermore, CDKN2A is a tumor suppressor gene that encodes p16; therefore, loss of CDKN2A results in cell-cycle progression through the phosphorylation of the Rb protein[24-26]. About ninety-five percent of pancreatic adeno-carcinomas have a loss of CDKN2A and thus, p16[19]. A transgenic mouse model with loss of p16 on top of KRAS mutation demonstrated an earlier and more aggressive cancer phenotype than KRAS mutation alone[27]. In vitro pancreatic cells treated with a CDK inhibitor plus gemcitabine showed consid-erable success by inhibiting tumor growth, sensitizing cells to gemcitabine-induced apoptosis, and down-regulating angiogenesis[28]. TP53, another well-known tumor suppressor gene, encodes p53; loss of TP53 results in a cascade of events bypassing integral DNA checkpoints and apoptotic signals. TP53 mutations are found in fifty to seventy-five percent of pancreatic tumors, with missense point mutations being the most common type of mutation[29-31]. Multiple in vivo mouse models[32-33] demonstrated that mutations in both p53 and KRAS resulted in early and more progressive tumorigenesis, transforming preinvasive lesions induced by KRAS to invasive and metastatic carcinomas from the addition of the p53 mutation. Furthermore, the NOTCH pathway is an embryogenic pathway that prevents terminal differentiation of cells. When ligands bind to NOTCH receptors, the enzyme γ-secretase cleaves the receptor's intracellular domain so that it can translocate to the nucleus and transcribe its respective genes[34].

Researchers have demonstrated increased expression of NOTCH genes in early neoplastic lesions as well as in chronic inflammation from cancer or pancreatitis[35]. NOTCH also appears to play a role in interacting with other signaling pathways such as EFGR and NF-kB[36]. In vitro and in vivo studies have attempted to inhibit the NOTCH pathway by inhibiting γ-secretase, resulting in less progression to invasive carcinoma[37]. Others have further investigated the role of the stromal component of pancreatic adenocarcinoma. Angiogenesis is vital to growth and progression of many tumors. Although microvessel density is fairly low in pancreatic adenocarcinoma, it has been established that angiogenesis plays a central role in the early parts of tumor progression[38-39]. Also, pancreatic cancer is associated with a significant desmoplastic reaction involving inflammatory cells; thus, it has been demonstrated that the number of macrophages and mast cells correlate with microvessel density[40-42]. In 2010, a phase III trial by the Cancer and Leukemia Group B investigated the efficacy of bevacizumab (a VEGF inhibitor) plus gemcitabine vs. gemcitabine alone after encouraging results from a multicenter phase II trial. The median overall survival was 5.8 months vs. 5.9 months, the median progression-free survival, 3.8 months vs. 2.9 months and the overall response rates, 13% vs. 10%

in the gemcitabine-bevacizumab vs. gemcitabine-placebo group, respectively[43]. In comparison to gemcitabine alone, the combination of gemcitabine-bevacizumab did not show a major benefit. A phase II study was soon underway in 2011 to investigate the efficacy of gemcitabine plus sorafenib, a multi-targeted tyrosine kinase inhibitor for receptors such as VEGF, PDGF, and RAF[3]. Although this was a well-tolerated regimen, it did not have promising results. In order to further elucidate these findings, the BAYPAN study, a double blind phase III randomized controlled trial, was conducted in 2012[44]. Again, this trial confirmed that the addition of sorafenib to gemcitabine does not improve overall survival. Currently, a phase I/II study initiated in 2011 is examining the combination of gemcitabine and PCI-27483, a Factor VIIa inhibitor. The trial is still ongoing; however, animal models with this drug have demonstrated tumor growth inhibition at 2.5-3.0 times change in prothrombin time[3]. Clearly, understanding the molecular pathogenesis of pancreatic cancer has led to relevant clinical trials and discoveries; however, the challenge comes from finding the correct combination of systemic drugs and targeted therapies to control the progression of this highly malignant cancer.

With an understanding of molecular targets and the creation of novel drugs targeting these molecular hotspots, identifying predictive markers in individual tumors will help direct the most beneficial of these novel drugs. The development of predictive markers has been set on gemcitabine, as it has been standard therapy since 1996. In 2007, predictive markers for gemcitabine resistance were identified in human pancreatic cancer cell lines[45]. It was discovered that the genes RRM1, RRM2, and hENT1 were increased in cells resistant to gemcitabine, whereas dCK was decreased[45].

Subsequently in 2010, these same four genes were analyzed as predictive markers to determine gemcitabine sensitivity in pancreatic cancer cell lines[46]. Quantitative analysis revealed that high dCK, low RRM2, and high GEM score (the combined score) predicted a longer disease-free survival for those treated with gemcitabine[46]. Multiple studies have also recently looked at the RNA binding protein, HuR, as a potential promising predictive marker for gemcitabine therapy[47-48]. HuR works by stabilizing dCK, which is responsible for phosphorylating gemcitabine into its active metabolites. Much like HER2/neu and herceptin, the presence of HuR is associated with a worse prognosis and stage for pancreatic cancer; however, its presence is also associated with better outcomes when treated with gemcitabine[47-48]. In regards to erlotinib, a phase III trial in 2012 comparing two sequences—gemcitabine/erlotinib followed by capecitabine vs. capecitabine/erlotinib followed by gemcitabine—incidentally discovered that KRAS wildtype status correlated with improved overall survival[49]. This sheds light on KRAS acting as a predictive marker for those treated with erlotinib, much as it is used for treatment with cetuximab.

Despite substantial improvements in systemic chemotherapy and a better understanding of the molecular pathogenesis of pancreatic cancer, it still remains one of the most aggressive and deadly cancers of all time. It is becoming apparent that a multi-targeted approach works best for pancreatic cancer to inhibit the multiple signaling pathways that enhance tumorigenesis. Predictive markers for the systemic or targeted therapies that have demonstrated improved overall survival in clinical trials are being identified. In this way, therapy can be individualized depending on the tumor profile, a paradigm shift that has occurred for other cancers that have shown a steady increase in survival rates. Rather than exposing a patient to a drug to merely experience its toxic side effects with limited improvement in survival time, we can stratify patients who will benefit and avoid treating those who will not. We are hopeful that improved insights and new technology will lead to a more promising future for pancreatic cancer, in which the diagnosis will one day be less of a death sentence and more of a chronic disease.

## REFERENCES

1. **Jemal A, Siegel R, Xu J, Ward E**. Cancer statistics, 2010. *CA Cancer J Clin*. 2010 Sep-Oct;60(5):277-300. Epub 2010 Jul 7. Erratum in: CA Cancer J Clin. 2011 Mar-Apr;61(2):133-4. PubMed PMID: 20610543.
2. **Li D, Xie K, Wolff R, Abbruzzese JL**. Pancreatic cancer. *Lancet*. 2004 Mar 27;363(9414):1049-57. Review. PubMed PMID: 15051286; PubMed Central PMCID:PMC3062508.
3. **Oberstein PE, Saif MW**. First-line treatment for advanced pancreatic cancer. Highlights from the "2011 ASCO Gastrointestinal Cancers Symposium". San Francisco, CA, USA. January 20-22, 2011. *JOP*. 2011 Mar 9;12(2):96-100. PubMed PMID: 21386629.
4. **McCleary-Wheeler AL, McWilliams R, Fernandez-Zapico ME**. Aberrant signaling pathways in pancreatic cancer: a two compartment view. *Mol Carcinog*. 2012 Jan;51(1):25-39. doi: 10.1002/mc.20827. PubMed PMID: 22162229; PubMed Central PMCID: PMC3253704.
5. **Burris HA 3rd, Moore MJ, Andersen J, Green MR, Rothenberg ML, Modiano MR, Cripps MC, Portenoy RK, Storniolo AM, Tarassoff P, Nelson R, Dorr FA, Stephens CD, Von Hoff DD**. Improvements in survival and

clinical benefit with gemcitabine as first-line therapy for patients with advanced pancreas cancer: a randomized trial. *J Clin Oncol*. 1997 Jun;15(6):2403-13. PubMed PMID: 9196156.

6. **Neoptolemos JP, Stocken DD, Friess H, Bassi C, Dunn JA, Hickey H, Beger H, Fernandez-Cruz L, Dervenis C, Lacaine F, Falconi M, Pederzoli P, Pap A, Spooner D, Kerr DJ, Büchler MW**; European Study Group for Pancreatic Cancer. A randomized trial of chemoradiotherapy and chemotherapy after resection of pancreatic cancer. *N Engl J Med*. 2004 Mar 18 ;350(12):1200-10. Erratum in: N Engl J Med. 2004 Aug 12;351(7):726. PubMed PMID: 15028824.
7. **Katz MH, Fleming JB, Lee JE, Pisters PW**. Current status of adjuvant therapy for pancreatic cancer. *Oncologist*. 2010;15(11):1205-13. Epub 2010 Nov 2. PubMed PMID: 21045189; PubMed Central PMCID: PMC3227902.
8. **Regine WF, Winter KA, Abrams RA, Safran H, Hoffman JP, Konski A, Benson AB, Macdonald JS, Kudrimoti MR, Fromm ML, Haddock MG, Schaefer P, Willett CG, Rich TA**. Fluorouracil vs gemcitabine chemotherapy before and after fluorouracil-based chemoradiation following resection of pancreatic adenocarcinoma: a randomized controlled trial. *JAMA*. 2008 Mar 5;299(9):1019-26. Erratum in: JAMA. 2008 Apr 23/30;299(16):1902. PubMed PMID: 18319412.
9. **Neoptolemos JP, Moore MJ, Cox TF, Valle JW, Palmer DH, McDonald AC, Carter R, Tebbutt NC, Dervenis C, Smith D, Glimelius B, Charnley RM, Lacaine F, Scarfe AG, Middleton MR, Anthoney A, Ghaneh P, Halloran CM, Lerch MM, Oláh A, Rawcliffe CL, Verbeke CS, Campbell F, Büchler MW**; European Study Group for Pancreatic Cancer. Effect of adjuvant chemotherapy with fluorouracil plus folinic acid or gemcitabine vs observation on survival in patients with resected periampullary adenocarcinoma: the ESPAC-3 periampullary cancer randomized trial. *JAMA*. 2012 Jul 11;308(2):147-56. PubMed PMID: 22782416.
10. **Conroy T, Desseigne F, Ychou M, Bouché O, Guimbaud R, Bécouarn Y, Adenis A, Raoul JL, Gourgou-Bourgade S, de la Fouchardière C, Bennouna J, Bachet JB, Khemissa-Akouz F, Péré-Vergé D, Delbaldo C, Assenat E, Chauffert B, Michel P, Montoto-Grillot C, Ducreux M**; Groupe Tumeurs Digestives of Unicancer; PRODIGE Intergroup. FOLFIRINOX versus gemcitabine for metastatic pancreatic cancer. *N Engl J Med*. 2011 May 12;364(19):1817-25. PubMed PMID: 21561347.
11. **Hruban RH, van Mansfeld AD, Offerhaus GJ, van Weering DH, Allison DC, Goodman SN, Kensler TW, Bose KK, Cameron JL, Bos JL**. K-ras oncogene activation in adenocarcinoma of the human pancreas. A study of 82 carcinomas using a combination of mutant-enriched polymerase chain reaction analysis and allele-specific oligonucleotide hybridization. *Am J Pathol*. 1993 Aug;143(2):545-54. Review. PubMed PMID: 8342602; PubMed Central PMCID: PMC1887038.
12. **Hingorani SR, Petricoin EF, Maitra A, Rajapakse V, King C, Jacobetz MA, Ross S, Conrads TP, Veenstra TD, Hitt BA, Kawaguchi Y, Johann D, Liotta LA, Crawford HC, Putt ME, Jacks T, Wright CV, Hruban RH, Lowy AM, Tuveson DA**. Preinvasive and invasive ductal pancreatic cancer and its early detection in the mouse. *Cancer Cell*. 2003 Dec;4(6):437-50. PubMed PMID: 14706336.
13. **Williams TM, Weiner DB, Greene MI, Maguire HC Jr**. Expression of c-erbB-2 in human pancreatic adenocarcinomas. *Pathobiology*. 1991;59(1):46-52. PubMed PMID: 1675057.
14. **Korc M, Chandrasekar B, Yamanaka Y, Friess H, Buchier M, Beger HG**. Overexpression of the epidermal growth factor receptor in human pancreatic cancer is associated with concomitant increases in the levels of epidermal growth factor and transforming growth factor alpha. *J Clin Invest*. 1992 Oct;90(4):1352-60. PubMed PMID: 1401070; PubMed Central PMCID: PMC443180.
15. **Philip PA, Benedetti J, Corless CL, Wong R, O'Reilly EM, Flynn PJ, Rowland KM, Atkins JN, Mirtsching BC, Rivkin SE, Khorana AA, Goldman B, Fenoglio-Preiser CM, Abbruzzese JL, Blanke CD**. Phase III study comparing gemcitabine plus cetuximab versus gemcitabine in patients with advanced pancreatic adenocarcinoma: Southwest Oncology Group-directed intergroup trial S0205. *J Clin Oncol*. 2010 Aug 1;28(22):3605-10. Epub 2010 Jul 6. PubMed PMID: 20606093; PubMed Central PMCID: PMC2917315.
16. **Moore MJ, Goldstein D, Hamm J, Figer A, Hecht JR, Gallinger S, Au HJ, Murawa P, Walde D, Wolff RA, Campos D, Lim R, Ding K, Clark G, Voskoglou-Nomikos T, Ptasynski M, Parulekar W**; National Cancer Institute of Canada Clinical Trials Group. Erlotinib plus gemcitabine compared with gemcitabine alone in patients with advanced pancreatic cancer: a phase III trial of the National Cancer Institute of Canada Clinical Trials Group. *J Clin Oncol*. 2007 May 20;25(15):1960-6. Epub 2007 Apr 23. PubMed PMID: 17452677.
17. **Jones S, Zhang X, Parsons DW, Lin JC, Leary RJ, Angenendt P, Mankoo P, Carter H, Kamiyama H, Jimeno A, Hong SM, Fu B, Lin MT, Calhoun ES, Kamiyama M, Walter K, Nikolskaya T, Nikolsky Y, Hartigan J, Smith DR, Hidalgo M, Leach SD, Klein AP, Jaffee EM, Goggins M, Maitra A, Iacobuzio-Donahue C, Eshleman JR, Kern SE, Hruban RH, Karchin R, Papadopoulos N, Parmigiani G, Vogelstein B, Velculescu VE, Kinzler KW**. Core signaling pathways in human pancreatic cancers revealed by global genomic analyses. *Science*. 2008 Sep 26;321(5897):1801-6. Epub 2008 Sep 4. PubMed PMID: 18772397; PubMed Central PMCID: PMC2848990.
18. **Maitra A, Hruban RH**. Pancreatic cancer. *Annu Rev Pathol*. 2008;3:157-88. Review. PubMed PMID: 18039136; PubMed Central PMCID: PMC2666336.
19. **Wilentz RE, Iacobuzio-Donahue CA, Argani P, McCarthy DM, Parsons JL, Yeo CJ, Kern SE, Hruban RH**. Loss of expression of Dpc4 in pancreatic intraepithelial neoplasia: evidence that DPC4 inactivation occurs late in neoplastic progression. *Cancer Res*. 2000 Apr 1;60(7):2002-6. PubMed PMID: 10766191.
20. **Ellenrieder V, Hendler SF, Boeck W, Seufferlein T, Menke A, Ruhland C, Adler G, Gress TM**. Transforming growth factor beta1 treatment leads to an epithelial-mesenchymal transdifferentiation of pancreatic cancer cells requiring extracellular signal-regulated kinase 2 activation. *Cancer Res*. 2001 May 15;61(10):4222-8. PubMed PMID: 11358848.
21. **Aoyagi Y, Oda T, Kinoshita T, Nakahashi C, Hasebe T, Ohkohchi N, Ochiai A**. Overexpression of TGF-beta by infiltrated granulocytes correlates with the expression of collagen mRNA in pancreatic cancer. *Br J Cancer*. 2004 Oct 4;91(7):1316-26. PubMed PMID: 15365564; PubMed Central PMCID: PMC2409911.
22. **Bardeesy N, Cheng KH, Berger JH, Chu GC, Pahler J, Olson P, Hezel AF, Horner J, Lauwers GY, Hanahan D, DePinho RA**. Smad4 is dispensable for normal pancreas development yet critical in progression and tumor biology of pancreas cancer. *Genes Dev*. 2006 Nov 15;20(22):3130-46. PubMed PMID: 17114584; PubMed Central PMCID: PMC1635148.

23. **Deharvengt S, Marmarelis M, Korc M**. Concomitant targeting of EGF receptor, TGF-beta and SRC points to a novel therapeutic approach in pancreatic cancer. *PLoS One*. 2012;7(6):e39684. Epub 2012 Jun 27. PubMed PMID: 22761868; PubMed Central PMCID: PMC3384603.
24. **Schutte M, Hruban RH, Geradts J, Maynard R, Hilgers W, Rabindran SK, Moskaluk CA, Hahn SA, Schwarte-Waldhoff I, Schmiegel W, Baylin SB, Kern SE, Herman JG**. Abrogation of the Rb/p16 tumor-suppressive pathway in virtually all pancreatic Carcinomas. *Cancer Res*. 1997 Aug 1;57(15):3126-30. PubMed PMID: 9242437.
25. **Caldas C, Hahn SA, da Costa LT, Redston MS, Schutte M, Seymour AB, Weinstein CL, Hruban RH, Yeo CJ, Kern SE**. Frequent somatic mutations and homozygous deletions of the p16 (MTS1) gene in pancreatic adenocarcinoma. Nat Genet. 1994 Sep;8(1):27-32. Erratum in: *Nat Genet* 1994 Dec;8(4):410. PubMed PMID: 7726912.
26. **Rozenblum E, Schutte M, Goggins M, Hahn SA, Panzer S, Zahurak M, Goodman SN, Sohn TA, Hruban RH, Yeo CJ, Kern SE**. Tumor-suppressive pathways in pancreatic carcinoma. *Cancer Res*. 1997 May 1;57(9):1731-4. PubMed PMID: 9135016.
27. **Bardeesy N, Aguirre AJ, Chu GC, Cheng KH, Lopez LV, Hezel AF, Feng B, Brennan C, Weissleder R, Mahmood U, Hanahan D, Redston MS, Chin L, Depinho RA**. Both p16(Ink4a) and the p19(Arf)-p53 pathway constrain progression of pancreatic adenocarcinoma in the mouse. *Proc Natl Acad Sci U S A*. 2006 Apr 11 ;103(15):5947-52. Epub 2006 Apr 3. PubMed PMID: 16585505; PubMed Central PMCID:PMC1458678.
28. **Subramaniam D, Periyasamy G, Ponnurangam S, Chakrabarti D, Sugumar A, Padigaru M, Weir SJ, Balakrishnan A, Sharma S, Anant S**. CDK-4 inhibitor P276 sensitizes pancreatic cancer cells to gemcitabine-induced apoptosis. *Mol Cancer Ther*. 2012 Jul;11(7):1598-608. doi: 10.1158/1535-7163.MCT-12-0102. Epub 2012 Apr 24. PubMed PMID: 22532602; PubMed Central PMCID: PMC3392497.
29. **Redston MS, Caldas C, Seymour AB, Hruban RH, da Costa L, Yeo CJ, Kern SE**. p53 mutations in pancreatic carcinoma and evidence of common involvement of homocopolymer tracts in DNA microdeletions. *Cancer Res*. 1994 Jun 1;54(11):3025-33. PubMed PMID: 8187092.
30. **Scarpa A, Capelli P, Mukai K, Zamboni G, Oda T, Iacono C, Hirohashi S**. Pancreatic adenocarcinomas frequently show p53 gene mutations. *Am J Pathol*. 1993 May;142(5):1534-43. PubMed PMID: 8494051; PubMed Central PMCID: PMC1886920.
31. **Barton CM, Staddon SL, Hughes CM, Hall PA, O'Sullivan C, Klöppel G, Theis B, Russell RC, Neoptolemos J, Williamson RC**, et al. Abnormalities of the p53 tumour suppressor gene in human pancreatic cancer. *Br J Cancer*. 1991 Dec;64(6):1076-82. Erratum in: Br J Cancer 1992 Mar;65(3):485. PubMed PMID: 1764370; PubMed Central PMCID: PMC1977874.
32. **Hingorani SR, Wang L, Multani AS, Combs C, Deramaudt TB, Hruban RH, Rustgi AK, Chang S, Tuveson DA**. Trp53R172H and KrasG12D cooperate to promote chromosomal instability and widely metastatic pancreatic ductal adenocarcinoma in mice. *Cancer Cell*. 2005 May;7(5):469-83. PubMed PMID: 15894267.
33. **Wagner M, Greten FR, Weber CK, Koschnick S, Mattfeldt T, Deppert W, Kern H, Adler G, Schmid RM**. A murine tumor progression model for pancreatic cancer recapitulating the genetic alterations of the human disease. *Genes Dev*. 2001 Feb 1;15(3):286-93. PubMed PMID: 11159909; PubMed Central PMCID: PMC312620.
34. **Mullendore ME, Koorstra JB, Li YM, Offerhaus GJ, Fan X, Henderson CM, Matsui W, Eberhart CG, Maitra A, Feldmann G**. Ligand-dependent Notch signaling is involved in tumor initiation and tumor maintenance in pancreatic cancer. *Clin Cancer Res*. 2009 Apr 1;15(7):2291-301. Epub 2009 Mar 3. PubMed PMID: 19258443; PubMed Central PMCID: PMC2711441.
35. **Miyamoto Y, Maitra A, Ghosh B, Zechner U, Argani P, Iacobuzio-Donahue CA, Sriuranpong V, Iso T, Meszoely IM, Wolfe MS, Hruban RH, Ball DW, Schmid RM, Leach SD**. Notch mediates TGF alpha-induced changes in epithelial differentiation during pancreatic tumorigenesis. *Cancer Cell*. 2003 Jun;3(6):565-76. PubMed PMID: 12842085.
36. **Wang Z, Banerjee S, Li Y, Rahman KM, Zhang Y, Sarkar FH**. Down-regulation of notch-1 inhibits invasion by inactivation of nuclear factor-kappaB, vascular endothelial growth factor, and matrix metalloproteinase-9 in pancreatic cancer cells. *Cancer Res*. 2006 Mar 1;66(5):2778-84. PubMed PMID: 16510599.
37. **Plentz R, Park JS, Rhim AD, Abravanel D, Hezel AF, Sharma SV, Gurumurthy S, Deshpande V, Kenific C, Settleman J, Majumder PK, Stanger BZ, Bardeesy N**. Inhibition of gamma-secretase activity inhibits tumor progression in a mouse model of pancreatic ductal adenocarcinoma. *Gastroenterology*. 2009 May;136(5):1741-9.e6. Epub 2009 Jan 14. PubMed PMID: 19208345.
38. **Esposito I, Kayed H, Keleg S, Giese T, Sage EH, Schirmacher P, Friess H, Kleeff J**. Tumor-suppressor function of SPARC-like protein 1/Hevin in pancreatic cancer. *Neoplasia*. 2007 Jan;9(1):8-17. PubMed PMID: 17325739; PubMed Central PMCID: PMC1803032.
39. **Sharma S, Green KB**. The pancreatic duct and its arteriovenous relationship: an underutilized aid in the diagnosis and distinction of pancreatic adenocarcinoma from pancreatic intraepithelial neoplasia. A study of 126 pancreatectomy specimens. *Am J Surg Pathol*. 2004 May;28(5):613-20. PubMed PMID: 15105649.
40. **Omary MB, Lugea A, Lowe AW, Pandol SJ**. The pancreatic stellate cell: a star on the rise in pancreatic diseases. *J Clin Invest*. 2007 Jan;117(1):50-9. Review. PubMed PMID: 17200706; PubMed Central PMCID: PMC1716214.
41. **Pandol S, Edderkaoui M, Gukovsky I, Lugea A, Gukovskaya A**. Desmoplasia of pancreatic ductal adenocarcinoma. *Clin Gastroenterol Hepatol*. 2009 Nov;7(11 Suppl):S44-7. Review. PubMed PMID: 19896098.
42. **Esposito I, Menicagli M, Funel N, Bergmann F, Boggi U, Mosca F, Bevilacqua G, Campani D**. Inflammatory cells contribute to the generation of an angiogenic phenotype in pancreatic ductal adenocarcinoma. *J Clin Pathol*. 2004 Jun;57(6):630-6. PubMed PMID: 15166270; PubMed Central PMCID: PMC1770337.
43. **Kindler HL, Niedzwiecki D, Hollis D, Sutherland S, Schrag D, Hurwitz H, Innocenti F, Mulcahy MF, O'Reilly E, Wozniak TF, Picus J, Bhargava P, Mayer RJ, Schilsky RL, Goldberg RM**. Gemcitabine plus bevacizumab compared with gemcitabine plus placebo in patients with advanced pancreatic cancer: phase III trial of the Cancer and Leukemia Group B (CALGB 80303). *J Clin Oncol*. 2010 Aug 1;28(22):3617-22. Epub 2010 Jul 6. PubMed PMID: 20606091; PubMed Central PMCID:PMC2917317.
44. **Gonçalves A, Gilabert M, François E, Dahan L, Perrier H, Lamy R, Re D, Largillier R, Gasmi M, Tchiknavorian X, Esterni B, Genre D, Moureau-Zabotto L, Giovannini M, Seitz JF, Delpero JR, Turrini O, Viens P, Raoul JL**. BAYPAN study: a double-blind phase III randomized trial comparing gemcitabine plus sorafenib and

gemcitabine plus placebo in patients with advanced pancreatic cancer. *Ann Oncol*. 2012 Nov;23(11):2799-805. doi: 10.1093/annonc/mds135. Epub 2012 Jul 5. PubMed PMID: 22771827.

45. **Nakano Y, Tanno S, Koizumi K, Nishikawa T, Nakamura K, Minoguchi M, Izawa T, Mizukami Y, Okumura T, Kohgo Y**. Gemcitabine chemoresistance and molecular markers associated with gemcitabine transport and metabolism in human pancreatic cancer cells. *Br J Cancer*. 2007 Feb 12;96(3):457-63. Epub 2007 Jan 16. PubMed PMID:17224927; PubMed Central PMCID: PMC2360025.
46. **Fujita H, Ohuchida K, Mizumoto K, Itaba S, Ito T, Nakata K, Yu J, Kayashima T, Souzaki R, Tajiri T, Manabe T, Ohtsuka T, Tanaka M**. Gene expression levels as predictive markers of outcome in pancreatic cancer after gemcitabine-based adjuvant chemotherapy. *Neoplasia*. 2010 Oct;12(10):807-17. PubMed PMID: 20927319; PubMed Central PMCID: PMC2950330.
47. **Richards NG, Rittenhouse DW, Freydin B, Cozzitorto JA, Grenda D, Rui H, Gonye G, Kennedy EP, Yeo CJ, Brody JR, Witkiewicz AK**. HuR status is a powerful marker for prognosis and response to gemcitabine-based chemotherapy for resected pancreatic ductal adenocarcinoma patients. *Ann Surg*. 2010 Sep;252(3):499-505; discussion 505-6. PubMed PMID: 20739850.
48. **Costantino CL, Witkiewicz AK, Kuwano Y, Cozzitorto JA, Kennedy EP, Dasgupta A, Keen JC, Yeo CJ, Gorospe M, Brody JR**. The role of HuR in gemcitabine efficacy in pancreatic cancer: HuR Up-regulates the expression of the gemcitabine metabolizing enzyme deoxycytidine kinase. *Cancer Res*. 2009 Jun 1;69(11):4567-72. PubMed PMID: 19487279; PubMed Central PMCID: PMC2744447.
49. **Heinemann V, Vehling-Kaiser U, Waldschmidt D, Kettner E, Märten A, Winkelmann C, Klein S, Kojouharoff G, Gauler TC, Fischer von Weikersthal L, Clemens MR, Geissler M, Greten TF, Hegewisch-Becker S, Rubanov O, Baake G, Höhler T, Ko YD, Jung A, Neugebauer S, Boeck S**. Gemcitabine plus erlotinib followed by capecitabine versus capecitabine plus erlotinib followed by gemcitabine in advanced pancreatic cancer: final results of a randomised phase 3 trial of the 'Arbeitsgemeinschaft Internistische Onkologie' (AIO-PK0104). *Gut*. 2012 Jul 7. [Epub ahead of print] PubMed PMID: 22773551.

*Submitted on August 23, 2012*