Disclosures

1. Dr. Smith is a co-inventor on 8 patents - the following patents related to pancreatic cancer:
   • a patent for OGF for therapy in pancreatic cancer.
   • a patent for nanoliposomes targeting the CCK receptor.
   • A patent for a monoclonal antibody to the CCK-C receptor

2. Dr. Smith is the Director of Clinical & Translation Research, LLC, a research consulting company’
   • Consult for Immune Therapeutics, Cytocom, and Cato Research
OBJECTIVES

- Understand how an idea is taken from the research lab to patient care.
- Learn the steps in conducting clinical trials
- Comprehend some of the obstacles to overcome in drug development?
- Examples of my translational projects
- Pitfalls and the Prize
Dreams

Nelson Mandela: “There is no easy walk to freedom anywhere, and many of us will have to pass through the valley of the shadow of death again and again before we reach the mountaintop of our desires.”

Edmond Hillary: "Mount Everest, you have defeated me. But I will return. And I will Defeat you. Because-you can't get any bigger but I can!"

Martin Luther King Jr: “I have a dream that one day this nation will rise up and live out the true meaning of its creed.. We hold these truths to be self-evident: that all men are created equal.”
Research & Drug Development

Preclinical research

Bottleneck of Drug development
You need an Idea

What is the Problem at hand?
What needs to be done to solve the issue?
How can your research change the problem?

Hypothesis

Passion!!
Phases of Clinical Trials

Most trials that involve new drugs go through a series of steps:

1. Experiments in the laboratory
   - In cells, \textit{in vitro}
   - In animals, \textit{in vivo}

2. Once deemed safe in animals, go through 1-4 phases
   - Phase 1: Safety, toxicity, dose
   - Phase 2: efficacy
   - Phase 3: compared to standard treatment
   - Phase 4: post-marketing
Types of Clinical Trials

• Treatment trials
• Prevention trials
• Early-detection trials/screening trials
• Diagnostic trials
• Quality-of-life studies/clinical benefit studies
• Genetic trials
Phase 1

- 15-30 people
- Determines
  - what dose is safe?
  - How the treatment should be given?
  - Pharmacokinetics?
  - How the treatment affects the body?
  - Safety & toxicity
Phase 2: Efficacy

• Less than 100 people
• Must have a primary endpoint
• Usually unbiased (blinded)
• Determines
  ➢ Does it work?
  ➢ Is it more effective than a placebo?
  ➢ Does not compare with other treatments
Phase 3

• From 100 to thousands of people
• Equal chance to be assigned to one of two or more groups
• Determines
  ➢ How the new treatment compares with the current standard
  ➢ Or how it compares with placebo
  ➢ Superiority or non-inferiority trials
Phase 4

• From hundreds to thousands of people
• Usually takes place after drug is approved to provide additional information on the drug’s risks, benefits and optimal use
• Called ‘Post-marketing” or
Or post-approval trials
Randomized Clinical Trials

• Equal chance to be assigned to one of two or more groups
  • One group gets the most widely accepted treatment (standard treatment) or placebo
  • The other gets the new treatment being tested
• All groups are as similar as possible
• Provides the best way to prove the effectiveness of a new agent or intervention
Pilot Study

• A small study that helps develop a bigger study
• A first venture into a particular area
• Used to iron out possible difficulties, and help with design of the bigger, more pivotal study.
• Helps provide ‘tentative response rate’ to estimate the sample size needed in a Phase 2 trial to reach significance over control
How Are Patients’ Rights Protected?

• Ethical and legal codes that govern medical practice also apply to clinical trials
• Informed consent
• Review boards
  ➢ Scientific review
  ➢ Institutional review boards (IRBs)
  ➢ Data safety and monitoring boards

Genetic testing Add to consent
How Do You Do It?

Examples from my experience
Pancreatic Cancer

- Fourth leading cause of cancer-related deaths in the United States; about 43,000/yr
- The median survival from diagnosis is 3-6 months.
- Five year survival is approximately 5%.
- Most cases are not diagnosed in the early stages.
- There is no effective treatment for nonsurgical cases.
Deaths from Pancreatic Cancer is Increasing

Pancreatic cancer is projected to become one of top two killers of cancer by 2020.
Treatment of advanced pancreatic cancer is poor

Survival 11.1 mos

Survival 8.5 months
Over 70 different Chemotherapeutic Regimens tried in pancreatic cancer
Pancreatic cancer is resistant to chemotherapy

Congress named pancreatic cancer one of the recalcitrant malignancies in 2012 and asked NIH to fund more research on pancreatic cancer.
You cannot expect to cure pancreatic cancer if you keep doing the same thing, i.e., we need to change strategy.
PROBLEMS

• Survival from pancreatic cancer has not improved in over 50 years in spite of all our technology and research. WHY?
Pancreatic Cancer: Reasons for Poor Prognosis

- No methods for early detection
- No screening tests for high risk subjects
- Resistant to chemotherapy and chemotherapy is toxic and kills normal cells
- We do not understand the biology of this cancer
Endogenous Opioid Peptides

- Play a role in neurotransmission
- Serve as potent regulators of growth
- Influence cells undergoing cellular repair
- Augment the immune system
- Induce feeling of overall well-being (i.e. euphoria, runner’s high)
Opioid Peptides and Receptors

Endogenous Opioids
- Enkephalin
- $[\text{Met}^5]\text{-enkephalin} = \text{OGF}$
- Endorphin
  - Euphoria
  - Runner’s high

Synthetic Opiates
- Morphine
- Demerol
- Codeine
- Darvon
- Fentanyl
- Methadone +/-
  - Pain
  - Sedation
  - Diarrhea
Once inside the nucleus, OGF activates the Rb pathway by upregulating p16 and p21 which are cyclin-dependent inhibitory kinases, and thereby retards transition from G1 to S phases in the cell cycle, with delayed cell replication and ultimate cell number resulting.

**Hypothesis**

- OGF inhibits growth of pancreatic cancer through the OGF receptor.

**Receptor Binding studies**

- $K_d = 1.2 \pm 0.3$ nM
- $B_{max} = 36.4 \pm 4.1$ fmol/mg protein
Mice were treated with 5mg/kg OGF three times a day to shrink tumors.
So how much do you give a 70 kg man?
Pancreatic Cancer

• What’s the problem?
  ➢ Pancreatic cancer has a dismal prognosis, 3-6 month survival, and no effective therapy

• What’s the hypothesis?
  ➢ OGF inhibits growth in pancreatic cancer cells and in nude mice so it should be effective in humans

• Where to start?
  ➢ A Phase 1 trial in patients
Before you start (Drug trial)

• Need approval from FDA
  • Apply for and IND# (investigational new drug#)
  • 1571 and 1572

• Write a protocol- study design with outcomes
• Write a consent form
• Obtain IRB approval
• Find a Sponsor - Get Funding support
• Responsibilities of the Principal Investigator
• Research Nurse /Study coordinator
• Registration of clinical trial on www.clinicaltrials.gov
FDA 1571 and 1572 forms, info about sponsor & drug

What are you Submitting or requesting In this report

Must be submitted with every communication to FDA
Aims of Phase 1 Study
First in Humans Trial

Study Objectives:

- Study the safety and toxicity of OGF in humans
- Determine the Maximum-Tolerated Dose (MTD)
- Study the biological kinetics and metabolism of OGF (Pharmacokinetics)
- Study the route of administration
Dose-escalation study
Classic 3x3 design

Determine at onset
What grade of side effect is acceptable

Started at 25 µg/kg

50 µg/kg
75 µg/kg
100 µg/kg
150 µg/kg
200 µg/kg
250 µg/kg

MTD
OGF Blood levels after Pharmacokinetic Studies
OGF: Phase 2 Clinical Study

- **Purpose:** efficacy: Does it work?
- **Study Design:** Open-labeled study
- **Sample size:** based on Phase 1 data
  - Treatment OGF 250μg/kg weekly IV (used the MTD discovered in Phase 1)
  - 50 subjects total / 166 controls

- **Eligible patients:**
  - Unresectable pancreatic cancer
  - Failed or refused standard therapy*
  - Controls: refused therapy and went to hospice
Phase 2 clinical trial

Outcomes:

• Survival
• Time to progression
• Tumor response (efficacy) - RECIST criteria (response eval. Criteria in solid tumors)
• Clinical Benefit
• Quality of Life (PRO-patient reported outcomes)
Results: CT Scan before & after OGF
RESULTS: PHASE 2 OGF Trial

Problems with study: FDA would only allow patients to enroll who had failed standard therapy for pancreatic cancer.

***p<0.001)

Kaplan Meier survival curve

***p<0.001)
Cholecystokinin Receptors and Pancreatic cancer

- GPCR: G-protein coupled receptors
- 7-trans-membrane domains
- Ligands: CCK and gastrin
Cholecystokinin Receptors:

- **CCK-A**: alimentary tract, gallbladder, mouse pancreas. Binds CCK > Gastrin (1,000:1)
- **CCK-B**: brain, stomach, human pancreas. Binds CCK = Gastrin (1:1)
- **CCK-C**: pancreatic cancer, splice variant of CCK-B; Only found in human cancer, not rodents. Binds Gastrin > CCK (10:1)
**CCK-B receptors are over-expressed in pancreatic cancer**

Smith et al, Am J Physiol 1994; 266: R 277

<table>
<thead>
<tr>
<th>Tissue /Cell Line</th>
<th>Binding Affinity</th>
<th>Receptor number</th>
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<tbody>
<tr>
<td></td>
<td>Kd, (nM)</td>
<td>Bmax (fmol/mg protein)</td>
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<tr>
<td>PANC-1 cells</td>
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<tr>
<td>MDA-Panc-28</td>
<td>3.6 ± 0.1</td>
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<tr>
<td>MDA-Amp-7</td>
<td>2.0 ± 0.4</td>
<td>211 ± 54</td>
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<td>MIA PaCa-2</td>
<td>3.0 ± 0.7</td>
<td>151 ± 12 *</td>
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<tr>
<td>Capan-1</td>
<td>2.7 ± 1.3</td>
<td>149 ± 83</td>
</tr>
<tr>
<td>BxPC-3</td>
<td>3.4 ± 0.1</td>
<td>125 ± 44</td>
</tr>
<tr>
<td>Fresh cancer from surgery</td>
<td>2.3 ± 0.8</td>
<td>285 ± 36</td>
</tr>
<tr>
<td>Normal human pancreas</td>
<td>1.8 ± 0.7</td>
<td>68 ± 7.2</td>
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</tbody>
</table>

CCK or gastrin stimulate growth of pancreatic cancer

![Graph showing the effect of CCK on cell count in SW1990 cells](image)
Gastrin and the Pancreas

*Gastrin is present in fetal pancreas where it aids in growth and differentiation but its expression is shut off at week 14.

*Gastrin is not present in the normal adult pancreas.

Direct Relationship Between Endogenous Gastrin Production & Cancer Growth

Gastrin is re-expressed in pancreatic cancer

Cancer growth is directly proportional to the amount of gastrin expressed

Growth rates of tumors

Antisense gastrin knock-down decreases gastrin mRNA expression and *in vivo* subcutaneous tumor formation.

**Matters. Pancreas** 38: e151-161, 2009
Novel Therapy for Pancreas Cancer Delivery Vehicles for Therapy

Nanoliposomes

1,2-Dioleoyl-3-Trimethyl- ammonium-propane (DOTAP, MW 774.19),
1,2-distearoyl-sn-glycero-3-phosphethanolamine-N-[methoxy
(polyethylene glycol)-2000 (PEG, MW 2,805.54), and 1,2-Dioleoyl-sn-
Glycero-3-Phosphethanolamine (DOPE, MW 744.04)

Calcium PO₄ Nanoparticles

SiRNA / Drug (purple)

Targeting protein (blue)

Fluorescent Probe (green)
Delivery of gastrin siRNA in nanoliposomes attenuates growth of pancreatic tumors \textit{in vivo}

Growth of an established pancreatic cancer is decreased by treatment of mice with gastrin siRNA laden cationic nanoliposomes

Pancreas 38: e151-161, 2009
Target-specific nanoparticles to pancreatic tumor cells *in vivo*: Using the CCK receptor as a target

A. Mice injected with untargeted, ICG loaded CPNP at 7 hrs (left) or 24 hrs (right) post-injection

B. Mice injected with gastrin-targeted CPNP at 7 hrs (left) or 24 hrs (right) post-injection showing enhanced CPNP uptake into the orthotopic tumor

C. Excised pancreatic tumor 24 h post-injection

Nude mice bearing orthotopic human pancreatic cancer cell tumors

Barth, ACS Nano, VOL. 4 • NO. 3 • 1279–1287 • 2010
Vaccine Against Gastrin

Patients with advanced pancreatic cancer were vaccinated either to gastrin or given a placebo vaccination.


Cancer Advances
Pancreatic cancer, gastrin/ CCK and gastrin/CCK receptors.
Alternative mRNA splicing in cancer cells creates a novel, CCK-C Receptor

Occurs in ~35% patients with pancreatic cancer and predicts risk  $p = 7.5 \times 10^{-8}$

CCK-B Receptor Gene

SNP Intron 4 + 32 (C->A) rs1800843

Human vs animal research

CCK-C receptor

<table>
<thead>
<tr>
<th>Exon</th>
<th>1</th>
<th>2</th>
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<th>4</th>
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<td>252</td>
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<tr>
<td>Mouse</td>
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<td>252</td>
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<td>176</td>
<td>194</td>
<td>533 bp</td>
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<tr>
<td>Rat</td>
<td>151</td>
<td>252</td>
<td>250</td>
<td>173</td>
<td>194</td>
<td>533 bp</td>
</tr>
</tbody>
</table>

Exon 4

Human: CAA GGC GGG CTG CCA G... gtt ggg gct gga cca cgt gag caa aat
GlIn Gly Gly Leu Pro Gly Gly Ala Gly Pro Arg Glu GlIn Asn

Mouse: GGG GCA GCA GCA CCA G... gtt ggg tga aaa aca cga ggg ggc ggg
Gly Ala Ala Ala Pro Gly Gly Stop

Rat: GGT GGG GCA GCA CCA G... gtt ggg tga aaa aca cga ggg gac aca ggg
Gly Gly Ala Ala Ala Pro Gly Gly Stop
The CCK-C receptor: SNP 4 +32 (C>A) rs1800843; found in 40% with pancreatic cancer
A. C- genotype

- SRp55

B. A- genotype

- SF2/ASF
- SF2/ASF (IgM-BRCA1)
- SC35
- SRp40
- SRp55

C. CCK-B Receptor pre-mRNA

C- allele pre-mRNA binds SRp55 - Normal splicing of IV4

A- allele pre-mRNA does not bind SRp55 - No splicing of IV4

CCKB Receptor mRNA

CCKC Receptor mRNA
Intellectual Property

• Submit an invention disclosure and provisional patent before you present the research results publically (including abstracts).

• The patent belongs to whomever you worked for when you made the discovery. If your employer does not want to file a patent have them assign the rights to you.

• Find a company

• License the patent when it issues.
Obstacles with Translational Research Today

1. $$$$$ Is the problem a lack of funds, misuse of funds, or disparity of funds?
2. Clinicians do not get protected time to do translational research.
3. Chiasm between industry and NIH /academia
4. Problems with patient accrual into research studies.
5. No more –one man bands, we need team science. PhDs must work with MDs
To cure pancreatic cancer

We need to think outside of the box
Don’t Be Afraid to take some Risks
Bottom Line: Does the research have Clinical relevance to Help people?

Bobbie (with permission) 
Pancreatic cancer patient

Vickie (with permission) 
Pancreatic cancer patient
Don’t Give Up!

“Strivers achieve what dreamers believe.” Usher

“I stand for freedom of expression, doing what you believe in, and going after your dreams.” Madonna Ciccone

“A dream doesn't become reality through magic; it takes sweat, determination and hard work.” Colin Powell

If you don’t believe in yourself and your dreams, no one else will either. Believe in yourself, have faith in your work and DON’T GIVE UP!

Jill Smith, MD
Thanks from the Smith lab

Funding NIH /NCI
Questions?