Convergence of Nanotechnology and Cancer Prevention: Are We There Yet?

Ernest Hawk, MD, MPH
Vice President and Head
Division of Cancer Prevention and Population Sciences

Executive Director
Duncan Family Institute for Cancer Prevention and Risk Assessment
Principle #1 - Cancer Results From An Interplay of Inherited Factors & Exposures That Damage Cardinal Elements of Cellular/Tissue Growth Control & Identity

“Non-modifiable” Risk Factors
- Major defects in cancer-promoting/inhibiting genes
- Subtle differences in genetic coding or expression

“Modifiable” Risk Factors
- Tobacco
- Poor diet
- Physical inactivity
- Viruses
- Occupational exposures

Modified from Hanahan & Weinberg, Cell 100:57, 2000; Science 2006
Principle #1 - Cancer Results From An Interplay of Inherited Factors & Exposures That Damage Cardinal Elements of Cellular/Tissue Growth Control & Identity

"Non-modifiable" Risk Factors
- Major defects in cancer-promoting/inhibiting genes
- Subtle differences in genetic coding or expression

"Modifiable" Risk Factors
- Tobacco
- Poor diet
- Physical inactivity
- Viruses
- Occupational exposures

Questions critical to prevention:
- Timing
- Order
- Frequency/prevalence
- Reversibility
  in preinvasive neoplastic lesions, especially in "lesions of greatest concern"

Modified from Hanahan & Weinberg, Cell 100:57, 2000; Science 2006
Principle #2 – Carcinogenesis is a Chronic Disease – From Molecular Defect to Dysplasia to Cancer

Existing Focus of Screening & Surgical Care

Rational Focus of Molecularly-targeted Intervention

Adapted from Ilyas et al. Eur. J. Cancer 1999; 35:335-351
Principle #3 – Carcinogenesis is Typically Polychronotropic in Nature

Figure 5. Intraepithelial neoplasia develops at multiple sites which progress independently, in an epithelium subject to diffuse genomic instability (called field cancerization by Slaughter [37]).

Boone et al. 1997
Principle #4 - The Therapeutic Index Drives Preventive Applications

A function of:

- Agent’s intended & unintended effects
- Individual/cohorts’ susceptibilities to both the disease and the agent’s effects
- Disease of interest

Achieving balance is particularly critical in prevention
Major Challenges in Cancer Prevention & Control – 2010

Effective Delivery of What Already “Works”
• Increasing diversity – race/ethnicity, geography, SES
• Growing numbers of those “at-risk”
  – Aging
  – Unhealthy lifestyles
  – Screening
  – Cancer survivors
• Limited public understanding of cancer development, risk, and prevention
• Increased coverage/reimbursement for prevention in health care reform legislation

Research & Translation to Improve Insights & Options
• Advances in –omics, biospecimen-based risk assessments & prevention
  – Limited impact to date
  – Flood of biomarkers without clear criteria for validation/clinical application
  – Flood of potential targets for intervention
  – Concerns over risk:benefit balance
• Advances in molecular and clinical imaging
• Paucity of dedicated research & researchers
• Paucity of private investment
Approaches to the Development and Marketing Approval of Drugs That Prevent Cancer


National Cancer Institute, Division of Cancer Prevention and Control, Bethesda, Maryland 20892 [G. J. K., J. A. C., C. W. B., V. E. S., P. G.], and the Food and Drug Administration, Center for Drug Evaluation and Research, Rockville, Maryland 20857 [J. R. J., J. J. D. G., M. U. M., J. W. T., W. J. S., G. B., R. J. T.]

The examples given of chemopreventive approaches primarily involve intraepithelial neoplasia (dysplasia) since the majority of human cancers are epithelial in origin. However, in the future, the chemopreventive approach may be applied to dysplasia in mesenchymal tissue as well, such as in premalignant lesions of connective and hematopoietic tissues leading to the sarcomas, leukemias, and lymphomas. It must be emphasized that this is not a regulatory document but rather a summary of consensus views of the participants.
<table>
<thead>
<tr>
<th>Disease</th>
<th>Intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breast cancer</td>
<td>Tamoxifen</td>
</tr>
<tr>
<td></td>
<td>Raloxifene</td>
</tr>
<tr>
<td>Cervical intraepithelial neoplasia &amp; cancer</td>
<td>Human Papillomavirus vaccines</td>
</tr>
<tr>
<td>Esophageal dysplasia</td>
<td>Photofrin + PDT*</td>
</tr>
<tr>
<td>Colonic adenomas</td>
<td>Celecoxib</td>
</tr>
<tr>
<td>Bladder dysplasia</td>
<td>Bacillus Calmet Guerin</td>
</tr>
<tr>
<td></td>
<td>Valrubicin</td>
</tr>
<tr>
<td>Actinic keratosis</td>
<td>Fluorouracil</td>
</tr>
<tr>
<td></td>
<td>Diclofenac sodium</td>
</tr>
<tr>
<td></td>
<td>5-aminolevulinic acid + PDT*</td>
</tr>
<tr>
<td></td>
<td>Masoprocol</td>
</tr>
</tbody>
</table>

*PDT – Photodynamic therapy
NIH-Funded Cancer Nanotechnology Grants

(n=191)

Prevention 1%
Screening 2%
Therapy/Treatment 55%
Imaging/Detection 41%

### NIH Portfolio in Nanotechnology Relevant to Cancer Screening & Prevention

<table>
<thead>
<tr>
<th>Project Number</th>
<th>Project Title</th>
<th>PI</th>
<th>Awardee Organization</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PREVENTION</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5U54CA119343-05</td>
<td>CAROLINA CENTER OF CANCER NанOTECHNOLOGY EXCELLENCE</td>
<td>JULIANO, RUDOLPH</td>
<td>UNIVERSITY OF NORTH CAROLINA, CHAPEL HILL</td>
</tr>
<tr>
<td>5U54CA119342-05</td>
<td>AN INFORMATICS RESOURCE FOR TARGETED NANOPARTICLE THERAPEUTICS</td>
<td>SEPT, DAVID</td>
<td>WASHINGTON UNIVERSITY</td>
</tr>
<tr>
<td><strong>SCREENING</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5R41CA132256-02</td>
<td>ENDOSCOPICALLY-GUIDED OPTICAL COHERENCE TOMOGRAPHY FOR EARLY CANCER SCREENING</td>
<td>IFTIMIA, NICUSOR</td>
<td>PHYSICAL SCIENCES, INC</td>
</tr>
<tr>
<td>5R41CA135911-02</td>
<td>ENHANCED CONTRAST FOR SCREENING OF EARLY PANCREATIC CANCER</td>
<td>IFTIMIA, NICUSOR</td>
<td>PHYSICAL SCIENCES, INC</td>
</tr>
<tr>
<td>5R01CA119402-05</td>
<td>INTEGRATED SYSTEM FOR CANCER BIOMARKER DETECTION</td>
<td>MANALIS, SCOTT</td>
<td>M.I.T.</td>
</tr>
<tr>
<td>5R21EB008520-02</td>
<td>NANODEVICE FOR DIGITAL IMAGING OF PALPABLE STRUCTURE AT HUMAN-FINGER RESOLUTION</td>
<td>SARAF, RAVI</td>
<td>UNIVERSITY OF NEBRASKA, LINCOLN</td>
</tr>
</tbody>
</table>

Cancer Biomarkers & Screening – Potential for Nanodetection?

Integrated barcode chips for rapid, multiplexed analysis of proteins in microliter quantities of blood

Rong Fan1–3,5, Ophir Vermesh1–3,5, Alok Srivastava1,4, Brian K H Yen1–3, Lidong Qin1–3, Habib Ahmad1–3, Gabriel A Kwong1–3, Chao-Chao Liu1–3, Juliane Gould1–3, Leroy Hood1,4 & James R Heath1–3

As the tissue that contains the largest representation of the human proteome2, blood is the most important fluid for clinical diagnostics2–4. However, although changes of plasma protein profiles reflect physiological or pathological conditions associated with many human diseases, only a handful of plasma proteins are routinely used in clinical tests. Reasons for this include the intrinsic complexity of the plasma proteome1, the heterogeneity of human diseases and the rapid degradation of proteins in sampled blood2. We report an integrated microfluidic system, the integrated blood barcode chip that can sensitively sample a large panel of protein biomarkers over broad concentration ranges and within 10 min of sample collection. It enables on-chip blood separation and rapid measurement of a panel of plasma proteins from

• Potential for inexpensive, non-invasive, informative detection
• Rapid assays for multiple cancer biomarkers from a finger prick of blood

We first present an overview of the IBBC and then discuss control of assay sensitivity, extension of a single protein assay to an assay for a large panel of biomarkers and, finally, integration of plasma separation from whole blood, followed by the rapid measurement of a panel of protein biomarkers. Figure 1 shows the design of an IBBC for blood separation and in situ protein measurement. We designed a polydimethylsiloxane (PDMS)-on-glass chip to perform 8–12 separate multiprotein assays sequentially or in parallel, starting from whole blood.

The Zweifach-Fung effect describes highly polarized blood cell flow at branch points of small blood vessels14–16. A component of the IBBC, redesigned from a previous report14, exploits this hydrodynamic effect by flowing blood through a low-flow-resistance primary channel with high-resistance, centimeter-long channels that branch off it at right
Information is the most valuable commodity in a systems approach to medicine, so diagnostic tests will have to easily and accurately measure large numbers of biological molecules for a few cents or less per measurement. Extreme miniaturization allowed the authors and their colleagues to produce a prototype chip that can measure concentrations of a panel of cancer-associated proteins in a droplet of blood in 10 minutes, at a cost of five to 10 cents per protein.

Promise of Nanoscale Interventions – Relevance to Prevention?

Nanoparticle Advantages\textsuperscript{1}

- Carry large “payloads” to improve efficacy
  - Including novel effectors, such as siRNA
- Improve targeting to bolster efficacy and/or reduce toxicity
  - Passive – based on enhanced vascular permeability and retention, differential pH/temperatures, etc.
  - Active – antigens, cell surface receptors, vasculature targets
- Provide a combinatorial platform to improve efficacy
- Bypass efflux pump-based resistance

Prevention Relevance

- Yes\textsuperscript{2}
  - Demonstrated 10x dose advantage in prostate cancer model with EGCG
- Probably
  - Although relevance of differential vascular EPR and active targeting strategies would require confirmation in preinvasive neoplasia
- Probably
  - Strong rationale based on success of combinations elsewhere in prevention
- Possibly

DFMO + Sulindac Combination Markedly Reduces Colon Adenoma Recurrence

<table>
<thead>
<tr>
<th>Variable</th>
<th>Placebo N=129</th>
<th>Treatment N=138</th>
<th>Relative risk (Reduction)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total adenoma recurrence (%)</td>
<td>53 (41.1)</td>
<td>17 (12.3)</td>
<td>0.30 (70%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Advanced adenoma recurrence (%)</td>
<td>11 (8.5)</td>
<td>1 (0.7)</td>
<td>0.085 (92%)</td>
<td>0.001</td>
</tr>
<tr>
<td>Large (&gt;1cm) adenoma recurrence (%)</td>
<td>9 (7.0)</td>
<td>1 (0.7)</td>
<td>0.10 (90%)</td>
<td>0.004</td>
</tr>
<tr>
<td>Patients with &gt;1 adenoma recurrence (%)</td>
<td>17 (13.2)</td>
<td>1 (0.7)</td>
<td>0.055 (95%)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Currently unclear when abnormal vessel architecture & EPR occur in carcinogenesis...estimates = 0.8 – 1 mm vs. < 0.2mm

Nano-EGCG

- Epigallocatechin-3-gallate – a green tea polyphenol
- Chemopreventive potential in human breast, pancreatic, colon, esophageal & lung cancers
- Poor oral absorption (<1%) requiring high doses (~ 8-10c/d) for chemopreventive benefit
- Caffeine-related GI & neurological side effects

- EGCG in PLA-PEG nanoparticles to improve therapeutic density

- Efficient uptake by murine prostate cancer cells in vitro & in vivo

- At significantly lower doses than those required with typical EGCG
  - Reduced cellular penetration barriers
  - Induced apoptosis
  - Retained mechanistic identity
  - Inhibited angiogenesis
  - Decreased tumor volume
  - Inhibited serum PSA
Nano-EGCG – Effective at 10-fold Lower IP Doses Measuring Tumor Volume and PSA Secretion in a Murine Xenograft Model Using Androgen-responsive 22Rv1 Prostate Cancer Cells

Celecoxib

1998: first FDA-approved COX-2 inhibitor to treat signs/symptoms of osteo- & rheumatoid arthritis

1999: approved as adjunct treatment for FAP

After 1 & 3 years of Celebrex at 200 & 400 mg BID, or 400mg QD vs. placebo in adenoma patients:
- 2004: inc risk of serious CV events up to 2-3x
- 2006: 30-60% red’n in recurrent adenomas

2007: Celecoxib-loaded solid lipid nanoparticles delivered directly to arthritic joints in rats
- Articular concentrations of celecoxib 15x higher 24h post-injection
- Reduced drug clearance into systemic circulation, potentially mitigating CV risks
Phosphatidylcholine-associated NSAIDs

- Chronic inflammation is associated with CV, autoimmune & neurologic diseases, arthritis, diabetes and cancer
- NSAIDs are efficacious in reducing risk of these diseases, but can result in gastric erosions and CV complications
- Conjugating phosphatidylcholine (PC) to NSAIDs (NSAID-PC) could mitigate adverse effects without compromising protective properties

In early clinical studies, a NSAID-PC conjugate demonstrated improved GI safety profile compared to unmodified NSAID in older OA patients at risk for NSAID-induced GI injury
Current Challenges & Limitations of Nanotechnology in Cancer Prevention

- Debate over the merits of early detection of preinvasive neoplasia
- Insufficient sensitivity and specificity to detect preinvasive neoplasia or early-stage cancer
- Need to measure multiple markers for reliable detection at early stages
- Biodegradability of nano-delivery systems (carbon, silica)
- High cost
- Need for IV administration
- Human data severely limited

**Can Nanotechnology Improve Health More Broadly?**

**Maybe Yes...**

### Functional Foods

- Addition of nanocarriers to transport vitamins, minerals and phytochemicals
- Infusion with nanoenhancers to improve taste and health benefits while reducing salt and sugar content

### Food Packaging

- Nano-based, non-chlorine coating as a barrier to preserve freshness
- Coating fruits and vegetables to protect from humidity and O₂ and to allow for harvesting of crops when they are riper

### Food Storage & Preparation

- Silver nanoparticles imbedded in food storage containers, plastic bags, cutting boards and cooking utensils retards growth of bacteria, mold and fungus
- Cookware coated with “nanoglaze” to prevent gaseous/toxic odors and peeling of cooking surface onto foods

### Vitamins & Supplements

- Many vitamins and precursors are insoluble in water but when formulated as nanoparticles, bioavailability increases

*The Project on Emerging Nanotechnologies: [www.nanotechproject.org](http://www.nanotechproject.org)*
Can Nanotechnology Improve Health More Broadly? Maybe No...

• Public perception
  – “Nano-inside” (e.g., foods) appears less acceptable than “nano-outside” (e.g., packaging)

• Unknowns
  – Do nanoparticles migrate from packaging to food?
    – What are the downstream environmental impacts during disposal?

• Social trust
  – Acceptance of technology
  – Assurance of regulatory oversight
Are We There Yet?

Francis Collins, MD, PhD
Director, National Institutes of Health

CANCER IN AMERICA
The Cancer You Can Beat
by Dr. Francis S. Collins
published: 06/20/2010

The incidence of colorectal cancer fell by more than half between 1965 and 2005—mostly because more people are getting screened. Yet colorectal cancer remains the second-leading cause of cancer death, claiming the lives of nearly 50,000 Americans each year. In these cases, colorectal cancer can be detected and treated at an early stage when a person is more likely to be cured. Indeed, a third of colorectal cancers have never been screened for colorectal cancer.

Many experts consider a colonoscopy to be the most effective cancer-screening tool available. In the procedure, a long, flexible tube equipped with a tiny camera is inserted into the rectum. The camera enables the doctor to detect and remove polyps and even cancers.

6 Things You Can Do to Help Prevent Cancer

Another screening mainstay is the fecal occult blood test, in which stool samples are examined for microscopic specks of blood. (If you get this test, make sure it’s the more accurate version that uses...

Current imaging methods detect cancers once they are large enough to be visible. But imagine how great it would be if doctors could spot a single cancerous or even pre-cancerous cell. This may soon be possible through nanotechnology, the branch of engineering that deals with the manipulation of individual atoms and molecules.

Parade Magazine, June 20, 2010