BIOGRAPHICAL SKETCH

NAME: **Laising Yen**

POSITION TITLE: **Assistant Professor (***currently under review for tenured Associate Professorship)*

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE	Completion Date	FIELD OF STUDY
National Taiwan University, Taipei, Taiwan	B.A.	05/1984	Zoology
Yale University, New Haven	Ph.D.	05/1994	Biology
Yale School of Medicine, New Haven	Post-doc	08/1999	Neurobiology
Harvard Medical School, Boston	Instructor	07/2007	Molecular Medicine

A. Personal Statement

My expertise is in gene regulation and RNA mutations during development and in human cancers. I received my Ph.D. degree from Yale University with a focus on the activity-mediated gene regulation of the NMDA receptor during central nervous system development. After graduation, I continued my training as a post-doctoral fellow at Yale School of Medicine and Harvard Medical School. I was the first to show that small molecules can be used to control the function of catalytic RNAs in mammalian cells. By using high-throughput screening of compound libraries, I identified novel inhibitors of cis-acting catalytic RNAs that could be used to control the riboswitch-based gene regulation. Since establishing my independent lab at Baylor College of Medicine and receiving the Duncan Scholar Award from the Dan L. Duncan Cancer Center, my team and I started to investigate RNA and genomic mutations in ovarian and prostate cancer. These studies led to the identification of important cancer-specific fusion genes, chimeric RNAs, circular RNAs with functional and clinical significance. Here I highlight a few publications in which I am either the first or last author:

1. **Yen, L**., J. Svendsen, J-S. Lee, J.T. Gray, M. Magnier, T. Baba, R.J. D'Amato and R.C. Mulligan. Exogenous control of mammalian gene expression via modulation of RNA self-cleavage. ***Nature*** 431:471-476 (2004). PMID: 15386015.
2. Kannan, K., L. Wang, J. Wang, M. Ittmann, W. Li, and **L. Yen**. Recurrent chimeric RNAs enriched in human prostate cancer identified by deep-sequencing. ***Proc Natl Acad Sci USA*** 108:9172-9177 (2011). PMID: 21571633.
3. Kannan, K, C. Coarfa, K. Rajapakshe, S. Hawkins, M. Matzuk, A. Milosavljevic, and **L. Yen**. CDKN2D-WDFY2 is a cancer-specific fusion gene recurrent in high-grade serous ovarian carcinoma. ***PLoS Genetics*** 10:e1004216 (2014). PMID: 24675677.
4. Kannan, K, C. Coarfa, P-W Chao, L. Luo, Y. Wang, A. E. Brinegar, S. Hawkins, A. Milosavljevic, M. Matzuk, and **L. Yen**. A recurrent BCAM-AKT2 fusion gene leads to a constitutively activated AKT2 fusion kinase in high-grade serous ovarian carcinoma. ***Proc Natl Acad Sci USA*** 112:E1272-7 (2015). PMID: 25733895.

B. Positions and Honors

Positions and Employment

1986 – 1993 Ph.D. Program, Department of Biology, Yale University, New Haven; thesis research supervised by Dr. Martha Constantine-Paton

1994 – 1999 Postdoctoral Associate, Department of Neurology, Yale University, School of Medicine, New Haven; Laboratory of Dr. Robert G. Kalb

1999 – 2007 Instructor in Pediatrics, Division of Molecular Medicine, Children's Hospital, Harvard Medical School, Boston, associated with the Laboratory of Dr. Richard Mulligan
2007 – Pres. Assistant Professor, Department of Pathology & Immunology, Baylor College of Medicine

Honors

1986 – 1989 Yale Graduate Fellowship
1996 Brown-Coxe Post-doctoral Fellowship, Yale University (declined)
1997 Hereditary Disease Foundation, Post-doctoral Fellowship Grant
1999 Astra Pharmaceuticals, L.P. Travel Scholarship
2007-2010 Duncan Scholar, Dan L. Duncan Cancer Center, Baylor College of Medicine
2010 NuGEN Innovation Challenge Award

Other Experience and Professional Memberships

1986-2000 Member, The Society for Neuroscience
1999-present Member, RNA Society
2008-present Member, American Association for Cancer Research
2014-present Member, American Society of Human Genetics
2014-present Visiting Assistant Professor, Institute of Molecular Medicine, Cheng Kung University, Taiwan
2014-present Guest Editor, *International Journal of Genomics*
2007-present Reviewer, *PLoS Genetics, Genome Research, PLoS One, Human Reproduction Update Molecular Vision, J Biotech, Investigative Ophthalmology & Visual Science, J Healthcare Engineering*

C. Contribution to Science

1. Activity-dependent plasticity of the central nervous system. During my Ph.D. thesis work at Yale University under the mentorship of Dr. Martha Constantine-Paton (now at MIT), and my first post-doc training at Yale School of Medicine with Dr. Bob Kalb (now at Univ. of Pennsylvania), I studied how the activity of neurons refines the connections of the central nervous system. One of the fundamental and unanswered questions at that time was the role played by the NMDA subtype of glutamate receptors in the refinement of synaptic connections. To address this question, I developed a statistical sampling assay to quantify the amount and the distribution of synaptic contacts within single axonal arbors. The results of my study were among the first to demonstrate that the activation of the NMDA receptor significantly reduced synaptic contacts made by each single axon. Furthermore, the remaining synapses within each axon were locally clustered, which suggested that the activation of NMDA receptor stabilized co-active synapses. Later, it became clear to me that many of the important questions concerning the refinement of neuronal connections might be answered if the expression of a particular glutamate receptor subtype could be regulated during development. This was before the era of siRNA and few gene regulation tools were available at that time. I therefore developed a small guide RNA sequence, which directs the endonulcease RNase P to cleave the NMDA mRNA in cells. Meanwhile, I realized that this catalytic nucleotide approach might have broader applications, perhaps as a therapeutic instrument. The gene coding for Huntington's disease had already been cloned at that time, and some evidence suggested that the mutant Huntington protein might be involved in NMDA receptor-mediated excitotoxicity in the death of neurons. With that in mind, I developed catalytic DNA enzymes that specifically cleave Huntington mRNA in cells. These early studies demonstrated the power of using simple catalytic nucleotide approaches in understanding biological questions and the potential in combating diseases.

a. **Yen, L**., J. Sibley and M. Constantine-Paton. Fine-structural alterations and clustering of developing synapses after chronic treatment with low levels of NMDA. ***J. Neurosci***. 13:4949-4960 (1993). PMID: 8229207.
b. **Yen, L**., J. Sibley and M. Constantine-Paton. Analysis of synaptic distribution within single retinal axonal arbors after NMDA treatment. ***J. Neurosci***. 15:4712-4725 (1995). PMID: 7540683.

c. **Yen, L.,** S.M. Strittmater and R.G. Kalb. Sequence-specific cleavage of Huntington mRNA by catalytic DNA. ***Annals of Neurology***, 46:366-373 (1999). PMID: 10482267.
d. **Yen, L.** Gonzalez-Zulueta M, Feldman A, Yuan Y, Fryer H, Dawson T, Dawson V, and R.G. Kalb. Reduction of functional NMDA Receptor in neurons by RNase P-mediated cleavage of the NR1 mRNA. ***J. Neurochem***. 76:1386-94 (2001). PMID: 11238723.

2. Controlling gene expression by RNA-small molecule interactions in mammalian cells. The experience of working with these catalytic nucleotides fostered a strong interest in gene regulation within me. I then joined Dr. Richard Mulligan, a gene therapy pioneer at Harvard, to focus on one of the fundamental goals in that field: the ability to control the expression of a transgene safely and with precision in clinical settings. The ability to control gene expression has always been indispensable in order to elucidate the function of a specific gene product or to generate therapeutic proteins within a safe range. One major limitation of the widely used tetracycline inducible system is the requirement of a protein transactivator that could cause serious host immune response in clinical applications. In addition, the system is leaky and lacks promoter flexibility. I set out to develop a new gene regulation system to overcome these limitations. The strategy was to design an extremely efficient self-cleaving ribozyme that when embedded in the mammalian mRNA, it would destroy the mRNA and result in no gene expression. If the cleavage of the ribozyme is blocked specifically by a small molecule, then it would lead to the generation of intact mRNA and therefore induced gene expression. This strategy of a 'drug-inducible riboswitch' was successful. I was able to provide an important proof-of-principle for this RNA-only gene regulation system and published a *Nature* article. To identify more small molecules that can serve as inducers for this gene regulation system, I performed a high-throughput screen of chemical libraries containing 58,076 compounds, and identified several new compounds capable of inhibiting ribozyme self-cleavage in human cells. These studies, which received many news and editorial comments, are significant because they provide a striking example of how engineered RNAs can be efficiently harnessed to control gene expression in human cells and regulated by drug-like molecules, which could overcome a major safty issue in clinical applications.

a. **Yen, L**., J. Svendsen, J-S. Lee, J.T. Gray, M. Magnier, T. Baba, R.J. D'Amato and R.C. Mulligan. Exogenous control of mammalian gene expression via modulation of RNA self-cleavage. ***Nature***, 431:471-476 (2004). PMID: 15386015
   News and Editorial Comments on this paper:
   - Eggleston AK. Tailor-made riboswitches. *Nature*, 431:409 (2004).
   - Sullenger, BA. Riboswitches- To kill or save the messenger. *New England Journal of Medicine*, 351: 2759-2760 (2004).
   - Eisenstein, M. Leave it to the cleaver. *Nature Methods*, 1:95 (2004).
   - Jaffe, S. Ten technologies in five years. *Scientist*, 18:29-31 (2004).
   - Breaker, RR. Gene expression control- Harnessing RNA switches. *Gene Therapy*, 12:725-726 (2005).
   - Baker M. On again, off again- a gene comes with a handy switch. *Technology Review*, Jan: 83 (2005).
b. **Yen, L**., M. Magnier, R. Weissleder, B.R. Stockwell, and R.C. Mulligan. Identification of inhibitors of ribozyme self-cleavage in mammalian cells via high-throughput screening of chemical libraries. ***RNA***, 12:797-806 (2006). PMID: 16556935
c. Link K.H., L. Guo, T. Ames, **L. Yen**, R.C. Mulligan, and R.R. Breaker. Engineering high speed allosteric hammerhead ribozymes. ***Biological Chemistry***, 388:779-786 (2007). PMID: 17655496
d. **Yen, L**., B.R. Stockwell, and R.C. Mulligan. A mammalian cell-based assay for screening inhibitors of RNA cleavage. ***Methods in Molecular Biology***, 540:335 (2009). PMID: 19381571

3. Chimeric RNAs and fusion genes in cancer. As an Assistant Professor at Baylor College of Medicine, my group is using our knowledge in RNA/gene regulation to investigate the RNA mutational events in ovarian and prostate cancer. This includes transcription-induced chimeric RNAs enriched in cancer, as well as fusion RNAs derived from fusion genes as the result of chromosomal arrangements. Before our studies, transcription-induced chimeric RNAs were thought to be very rare, and little was known regarding their presence and roles in human cancers. By using high-throughput mRNA-seq of prostate tumors, we were the

first to demonstrate that transcription-induced chimeric RNAs are widespread and far more prevalent in human cells than previously thought. Furthermore, they occur in significantly higher frequency in cancer than in matched benign tissues, suggesting that in cancer cells the limited number of human genes could encode a substantially larger number of RNAs and proteins forming an additional layer of cellular complexity. In addition to chimeric RNAs, my group identified important cancer-specific fusion genes. This includes CDKN2D-WDFY2 that occurs at a frequency of 20% among high-grade serous ovarian cancer, and BCAM-AKT2 that occurs at a frequency of 7%. We went on to show that the expression of CDKN2D-WDFY2 markedly alters the PI3K/AKT pathway, whereas the expression of BCAM-AKT2 leads to an in-frame fusion protein that is membrane-associated, constitutively phosphorylated, and escapes regulation from external stimuli. These studies represent significant advances in ovarian cancer. Given that there are very few novel proteins that can serve as drug targets in ovarian cancer, the cancer-specific fusion proteins that alter the PI3K pathway represent excellent drug targets, and could allow us to develop novel therapeutic compounds for tailored therapies.

a. Kannan, K., L. Wang, J. Wang, M. Ittmann, W. Li, and **L. Yen**. Recurrent chimeric RNAs enriched in human prostate cancer identified by deep-sequencing. ***Proc Natl Acad Sci USA*** 108:9172-9177 (2011). PMID: 21571633.
b. Zhang H, W. Lin, K. Kannan, L. Luo, J. Li, P-W Chao, Y. Wang, Y-P Chen, J. Gu, and **L. Yen**. Aberrant chimeric RNA *GOLM1-MAK10* encoding a secreted fusion protein as a molecular signature for human esophageal squamous cell carcinoma. ***Oncotarget*** 4:2135-43 (2013). PMID: 24243830.
c. Kannan, K, C. Coarfa, K. Rajapakshe, S. Hawkins, M. Matzuk, A. Milosavljevic, and **L. Yen**. *CDKN2D-WDFY2* is a cancer-specific fusion gene recurrent in high-grade serous ovarian carcinoma. ***PLoS Genetics*** 10:e1004216 (2014). PMID: 24675677.
d. Kannan, K, C. Coarfa, P-W Chao, L. Luo, Y. Wang, A. E. Brinegar, S. Hawkins, A. Milosavljevic, M. Matzuk, and **L. Yen**. A recurrent *BCAM-AKT2* fusion gene leads to a constitutively activated AKT2 fusion kinase in high-grade serous ovarian carcinoma. ***Proc Natl Acad Sci USA***. 112:E1272-7 (2015). PMID: 25733895.

Complete List of Published Work in MyBibliography:
http://www.ncbi.nlm.nih.gov/sites/myncbi/laising.yen.1/bibliography/46169731/public/?sort=date&direction=descending

D. Research Support

Ongoing Research Support

Biogen/IDEC Sponsored Research Agreement (PI: Yen) 08/01/2016-7/31/2019
Exogenous control of gene expression in human cells via modulation of RNA-mediated cleavage
The Sponsored Research Agreement aims to harness the power of RNA-based switches to develop the next generation gene regulation systems that can be turned on or off by FDA-approved small molecules.
Role: PI

CPRIT High Impact High Risk Award (PI: Yen) 06/01/16-05/31/18
Cancer Prevention and Research Institute of Texas
A "Pap smear" for ovarian cancer
This project aims to leverage the existing cervical Pap smear screening strategy with the specificity of cancer fusion genes identified in ovarian cancer to create a "Pap smear" test for ovarian cancer.
Role: PI

CPRIT post-doctoral fellowship (PI: Gupta, Yen lab) 03/01/16-02/28/18
Cancer Prevention and Research Institute of Texas
RNA-Driven Gene Fusion In Human Cancer

This project investigates a novel RNA-driven mechanism which provides a new framework for understanding the causation of cancer fusion genes.
Role: Mentor

Dan L. Duncan Cancer Center Pilot Grant (PI: Yen) 07/01/15 – 10/30/16
Targeting BCAM-AKT2 Fusion Kinase by Small Molecules to Combat Ovarian Cancer
The goal of this grant is to use DNA-encoded chemistry library technology, a discovery technology that is rapid and cost-effective, to identify lead compounds with drug-like properties that inhibit BCAM-AKT2 kinase activity for ovarian cancer therapy.
Role: PI

Completed Research Support

American Cancer Society IRG Seeds Fund (PI: Yen) 11/01/07 – 04/30/09
New approach to identify "secretory" cancer-specific splice variants
The goal of this pilot project is to develop a new experimental strategy to obtain a comprehensive profile of protein isoforms differentially secreted by tumors.
Role: PI

DOD- PCRP Idea Dev. Award (PI: Yen) 05/01/10 -04/31/14
Identifying novel secreted proteins in prostate cancer as biomarkers
This application aims to harness the power of modern genomics technology to identify new secreted biomarkers useful for improved diagnosis and treatment response monitoring in prostate cancer.
Role: PI

NIH 1U01 HD060496-01 (PI: Matzuk) 02/01/09 – 01/31/14
Identification of Small Molecule Contraceptives that Target the Male Germline
The Specific Aims of these proposed U01 studies are 1) Use small molecule microarrays and 2-hybrid screening assays to identify small molecules that bind GASZ, VASA, TEX14, or STYX and/or block key protein:protein interactions; and 2) Perform in vitro, in vivo, and computational screens to identify the most promising male contraceptives.
Role: Collaborator

Ovarian Cancer-PPD/BCM/01.12 (Project 2 Leader: Yen) 01/01/12 – 12/31/15
Ovarian Cancer Research Fund
Diagnostic Strategies for Detection of Ovarian Cancer
The overall goal of this OCRF grant is to define the novel and known proteins and metabolites involved in ovarian cancers and develop sensitive and specific assays for early detection of ovarian cancer.
Role: Co-PI

PCF Young Investigator Award (PI: Kannan, Yen lab) 03/01/12 – 4/30/15
Prostate Cancer Foundation
Investigating the biological significance of novel recurrent chimeric RNAs in Prostate Cancer
This application aims to study the functional significance and clinical utilities of recurrent chimeric RNAs found in prostate cancer.
Role: Mentor

NIH NIBIB R01 (PI: Yen) 07/01/11-06/30/16
Harnessing the power of RNA sensors for imaging molecular signatures in vivo
This application aims to utilize RNA-based sensors to enable sensitive detection of specific molecular signatures in living cells. The RNA sensor described offers enhanced capacities for imaging molecular signatures *in vivo* than is currently possible.
Role: PI

“**Building designer RNA nanostructures in human cells**”

By Laising Yen, Ph.D.

Abstract:

Unlike proteins, RNAs are highly programmable polymers due to their ability to form specific Watson-Crick base pairing, a property that can be exploited to create well-defined 2D and 3D structures. These structures are thermodynamically stable, and formed via spontaneous self-assembly, a process that requires no catalytic co-factors. In contrast to DNA, single-stranded RNA can be efficiently expressed at high levels in live cells, thus offers the opportunity to program cells to assemble designer nanostructures. We designed and constructed short single-stranded RNA monomers that self-assembled into higher order architectures, reaching a size of micrometers. When these RNA monomers were expressed in human cells, they appeared to form stable higher order structures. The results point to the possibility that RNA can be used to program mammalian cells to assemble designer nanostructures for specific cellular functions useful for synthetic biology applications.