BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
Follow this format for each person. **DO NOT EXCEED FIVE PAGES.**

NAME: Liang Tong

eRA COMMONS USER NAME (credential, e.g., agency login): LIANGTONG

POSITION TITLE: William R. Kenan, Jr. Professor

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Peking University, Beijing, China	B. Sc.	07/1983	Chemistry
University of California, Berkeley, California	Ph. D.	07/1989	Structural Biology
Purdue University, West Lafayette, Indiana	Post-doc	07/1992	Structural Biology

A. Personal Statement

Research: I have been in the field of structural biology for more than 38 years, and have made major contributions to understanding the molecular mechanisms of several different biological systems. I have a background in chemistry as an undergraduate student and established a structure-based drug design laboratory in a pharmaceutical company before starting at Columbia. I still have strong interests in studying proteins involved in human diseases and understanding protein-ligand interactions. We started working on acetyl-CoA carboxylase (ACC) in 2002, which is an attractive drug discovery target for diabetes, cancer and other diseases. This evolved into studies on other members of the biotin-dependent carboxylase family as well as other metabolic enzymes, including PFK. I am also very interested in deciphering the molecular mechanisms of fundamental biological processes. We started working on mRNA 3'-end processing as a collaboration with Dr. James Manley, an expert in this field in our Department. I then expanded to related areas, such as RNA degradation and RNA polymerase II termination, which unexpectedly led us to the discovery of the mRNA 5'-end capping quality surveillance mechanism. Finally, I also have strong training in the theory and practice of protein crystallography, and have developed new methodologies and computer software that has been distributed to many laboratories around the world. Overall, I have the expertise, motivation and leadership necessary to successfully carry out the proposed project.

Ongoing and recently completed projects that I would like to highlight include:

R35 GM118093
Tong (PI)
05/01/16-04/30/26 (ongoing)
Structural and functional studies of mRNA processing, stability and quality control

R01 NS135070
Tong (co-I, subcontractor)
09/20/23-07/31/2028 (ongoing)
Probing the role of Integrator in neuronal function

Kintor Pharmaceuticals
Tong (PI)
01/01/16-12/31/23 (ongoing)
Structures of human androgen receptor ligand binding domain (AR)-VHL in complex with inhibitors

Nimbus Discovery
Tong (PI)
02/17/22-12/17/23 (ongoing)
Structural studies of human VPS4

Citations:

1. S. Xiang, A. Cooper-Morgan, X. Jiao, M. Kiledjian, J.L. Manley & **L. Tong**. (**2009**). Structure and function of the 5'→3' exoribonuclease Rat1 and its activating partner Rai1. ***Nature***, 458, 784-788. PMCID: PMC2739979.
2. J. Wei & **L. Tong**. (**2015**). Crystal structure of the 500-kDa yeast acetyl-CoA carboxylase holoenzyme dimer. ***Nature***, 526, 723-727. PMCID: PMC4838907.
3. J. Wei, S. Leit, J. Kuai, E. Therrien, S. Rafi, H.J. Harwood Jr, B. DeLaBarre & **L. Tong**. (**2019**). An allosteric mechanism for potent inhibition of human ATP-citrate lyase. ***Nature***, 568, 566-570.
4. Y. Sun,* Y. Zhang,* W.S. Aik, X.-C. Yang, W.F. Marzluff, T. Walz[$], Z. Dominski[$] & **L. Tong**.[$] (**2020**). Structure of an active human histone pre-mRNA 3′-end processing machinery. ***Science***, 367, 700-703. (**–*equal first authors, [$]–co-corresponding authors*) PMCID: PMC7008720.

B. Positions, Scientific Appointments, and Honors

Positions

2015-present William R. Kenan, Jr. Professor of Biological Sciences, Columbia University, NY
2013-2019 Chair, Department of Biological Sciences, Columbia University, NY
2004-2015 Professor of Biological Sciences, Columbia University, NY
1997-2004 Associate Professor of Biological Sciences, Columbia University, NY
1996-1997 Principal Scientist. Boehringer Ingelheim Pharmaceuticals, Inc. Ridgefield, CT
1992-1995 Senior Scientist. Boehringer Ingelheim Pharmaceuticals, Inc. Ridgefield, CT

2019-2022 Honorary Research Professor, Durban University of Technology, Durban, South Africa
2015-2017 Honorary Professor, Nanjing Tech University, Nanjing, China

Current Scientific Appointments

2020-present Overseas Evaluation Expert, Chinese Academy of Sciences
2020-present Independent non-executive director, Kintor Pharmaceuticals, Suzhou, China
2016-present Consultant, Nimbus Therapeutics, Cambridge, MA

Selected Service

Columbia University Standing Committee on the Conduct of Research, member, 2022-present
Chair, Singapore Ministry of Education Academic Research Fund Tier 2 Expert Panel 3, 2015-present
co-Editor, Third Edition of Volume F of International Tables of Crystallography, 2018-present
NIH ZRG1 F07A-F (20), Fellowships: Infectious Diseases and Immunology, 2022
Chair, Department of Biological Sciences, Columbia University, 2013-2019
Regional Editor, Protein and Peptide Letters, 2006-2017
Manuscript Reviews for *Nature, Science, NSMB, Nature Chemical Biology, Molecular Cell, Science Advances, PNAS, Nature Communications, Nucleic Acids Research, RNA, Structure, Journal of Molecular Biology, Biochemistry*, and others.

Honors

1989 Phi Beta Kappa
1996 The Vice President's Golden Achievement Award, Boehringer Ingelheim Pharmaceuticals, Inc.
1997 The first Boehringer Ingelheim Worldwide Research and Development Award
2009 American Association for the Advancement of Science (AAAS) Fellow
2021 American Crystallographic Association (ACA) Fellow

C. Contributions to Science

1. Eukaryotic pre-mRNAs undergo extensive co-transcriptional processing, including cleavage and polyadenylation at the 3'-end. A large protein machinery has been identified, but the molecular mechanism for its function is still poorly understood. Based on our structures of several of the protein factors in this machinery, we showed that CPSF73 is the long-sought endoribonuclease for the cleavage step of 3'-end processing, and that the cleavage stimulation factor (CstF) may be dimeric in the machinery. In addition, the RNA polymerase II (Pol II) C-terminal domain phosphatase Ssu72 recognizes the *cis* configuration of the pSer5-Pro6 peptide bond, in contrast to general understanding about the substrate preference of protein phosphatases. We have also produced the first structural information on the distinct 3'-end processing machinery for replication-dependent histone pre-mRNAs, which are cleaved but not polyadenylated. Over the past five years, using the cryo-EM technique, we have revealed for the first time how the polyadenylation signal AAUAAA is recognized and how the mammalian CPSF is organized. Most excitingly, we have most recently determined the structure of an active, fully recombinant human histone pre-mRNA 3'-end processing machinery, the first structure of any active processing machinery. I am the principal investigator on all the structural studies, and collaborated with Drs. James Manley (in our Department), William Marzluff and Zbigniew Dominski (University of North Carolina, Chapel Hill), and Yongsheng Shi (University of California, Irvine) on some of the biochemical and functional studies. For the EM studies, I collaborated with Dr. Thomas Walz (Rockefeller University) as we did not have the expertise or infrastructure at the time.
    a. Y. Sun,* Y. Zhang,* K. Hamilton, J.L. Manley,[$] Y. Shi, T. Walz[$] & L. Tong.[$] (**2018**). Molecular basis for the recognition of the human AAUAAA polyadenylation signal. ***Proc. Natl. Acad. Sci. USA***, **115**, E1419-E1428. (Epub 12/5/17) (**–equal first authors, [$]–co-corresponding authors*) PMCID: PMC5816196.
    b. K. Hamilton, Y. Sun & L. Tong. (**2019**). Biophysical characterizations of the recognition of the AAUAAA polyadenylation signal. ***RNA***, **25**, 1673-1680. PMCID: PMC6859858.
    c. Y. Zhang,* Y. Sun,* Y. Shi, T. Walz[$] & L. Tong.[$] (**2020**). Structural insights into the human pre-mRNA 3′-end processing machinery. ***Mol. Cell***, **77**, 800-809. (Epub 12/3/19) (**–equal first authors, [$]–co-corresponding authors*) PMCID: PMC7036032.
    d. Y. Sun,* Y. Zhang,* W.S. Aik, X.-C. Yang, W.F. Marzluff, T. Walz[$], Z. Dominski[$] & L. Tong.[$] (**2020**). Structure of an active human histone pre-mRNA 3′-end processing machinery. ***Science***, **367**, 700-703. (**–equal first authors, [$]–co-corresponding authors*) PMCID: PMC7008720.

2. Our structure of the 5'-3' exoribonuclease Rat1 in complex with its binding partner Rai1 revealed that Rai1 contains an active site. In collaboration with Drs. Megerditch Kiledjian (Rutgers University) and James Manley (in our Department), we showed that Rai1 possesses RNA 5'-end pyrophosphohydrolase (PPH) activity and can also remove unmethylated caps. We hypothesized that Rai1 may be a central player in a novel mRNA 5'-end capping quality surveillance mechanism, and functional studies in Dr. Kiledjian's laboratory have demonstrated the existence of this mechanism in yeast and mammalian cells. We also discovered that the mammalian homolog of Rai1 (Dom3Z, now renamed as DXO) has distributive 5'-3' exoribonuclease activities. These studies overturn the current belief that mRNA capping always proceeds to completion and therefore no quality surveillance is necessary, and have opened up a new area of research. Over the past five years, we have shown that the DXO/Rai1 enzymes can also remove other non-canonical caps on RNAs, such as NAD (deNADding), FAD (deFADding), and dephospho-CoA (deCoAping). Moreover, we have found that Nudix family protein Nudt12 also has deNADding activity.
    a. X. Jiao, S. Doamekpor, J.G. Bird, B.E. Nickels, L. Tong, R.P. Hart & M. Kiledjian. (**2017**). 5′ end nicotinamide adenine dinucleotide cap in human cells promotes RNA decay through DXO-mediated deNADding. ***Cell***, **168**, 1015-1027. PMCID: PMC5371429.
    b. E. Grudzien-Nogalska,* Y. Wu,* X. Jiao, H. Cui, M.K. Mateyak, R.P. Hart, L. Tong[$] & M. Kiledjian.[$] (**2019**). Structural and mechanistic basis of mammalian Nudt12 deNADding. ***Nat. Chem. Biol.*** **15**, 575-582. (**–equal first authors, [$]–co-corresponding authors*) PMCID: PMC6527130.
    c. S.K. Doamekpor,* E. Grudzien-Nogalska,* A. Mlynarska-Cieslak, J. Kowalska, M. Kiledjian[$] & L. Tong.[$] (**2020**). DXO/Rai1 enzymes remove 5′-end FAD and dephospho-CoA caps on RNAs. ***Nucl. Acids Res***. **48**, 6136-6148. (**–equal first authors, [$]–co-corresponding authors*) PMCID: PMC7293010.

d. S. Sharma, E. Grudzien-Nogalska,* K. Hamilton,* X. Jiao, J. Yang, L. Tong & M. Kiledjian. (**2020**). Mammalian Nudix proteins cleave nucleotide metabolite caps on RNAs. ***Nucl. Acids Res***. **48**, 6788-6798. (**–equal second authors*) PMCID: PMC7337524.

3. Acyl-CoA carboxylases have central roles in metabolism and defective mutations in several of these enzymes are linked to serious diseases, especially in infants. However, the molecular basis for their function is still poorly understood. We have determined the crystal structures of the catalytic domains of acetyl-CoA carboxylase (ACC) and the 750 kD holoenzymes of propionyl-CoA carboxylase (PCC), 3-methylcrotonyl-CoA carboxylase (MCC) and a novel long-chain acyl-CoA carboxylase (LCC). The structures provide a basis for understanding the catalysis by these enzymes and the disease-causing mutations. The structures also reveal striking differences in the overall architectures of the holoenzymes despite the fact that they are composed of domains with substantial amino acid sequence identity, which also has general implications for the relationship between sequence conservation and structural similarity. This has been a long-term project in my laboratory at Columbia, and I am the principal investigator on all of these studies. The electron microscopy studies were carried out by Drs. Z. Hong Zhou (UCLA) and Thomas Walz (Harvard Medical School and Rockefeller University).
    a. C.S. Huang,* K. Sadre-Bazzaz,* Y. Shen, B. Deng, Z.H. Zhou & L. Tong. (**2010**). Crystal structure of the $\alpha_6\beta_6$ holoenzyme of propionyl-coenzyme A carboxylase. ***Nature***, **466**, 1001-1005. (**–equal first authors*) PMCID: PMC2925307.
    b. C.S. Huang, P. Ge, Z.H. Zhou & L. Tong. (**2012**). An unanticipated architecture of the 750-kDa $\alpha_6\beta_6$ holoenzyme of 3-methylcrotonyl-CoA carboxylase. ***Nature***, **481**, 219-223. PMCID: PMC3271731.
    c. T.H. Tran, Y.-S. Hsiao, J. Jo, C.-Y. Chou, L. Dietrich, T. Walz & L. Tong. (**2015**). Structure and function of a single-chain, multi-domain long-chain acyl-CoA carboxylase. ***Nature***, **518**, 120-124. PMCID: PMC4319993.
    d. J. Wei & L. Tong. (**2015**). Crystal structure of the 500-kDa yeast acetyl-CoA carboxylase holoenzyme dimer. ***Nature***, **526**, 723-727. PMCID: PMC4838907.

4. Pyruvate carboxylase (PC) is another important metabolic enzyme, and defective mutations are linked to serious diseases. In addition, this enzyme in the human pathogen *Listeria monocytogenes* (LmPC) was found to be regulated by the bacterial second messenger cyclic-di-AMP (c-di-AMP) in the laboratory of our collaborator, Dr. Joshua Woodward at the University of Washington. We have determined the structures of this enzyme, including that of LmPC in complex with c-di-AMP, which confirms that c-di-AMP is an allosteric regulator. Based on the structures, we hypothesize that c-di-AMP inhibits LmPC by freezing the enzyme into a single conformation, which is incompatible with catalysis. Besides PC, we also produced the first structures of several other metabolic enzymes, including carnitine acetyltransferase, the heterotrimer core of SNF1, which is the yeast homolog of AMP-activated protein kinase (AMPK), and human ATP-citrate lyase (ACLY) by cryo-EM. ACLY produces acetyl-CoA in the cytoplasm and is the upstream enzyme of ACC. It is a target for drug discovery against cancer and hypercholesterolemia. I am the principal investigator on all the structural studies.
    a. K. Sureka,* P.H. Choi,* M. Precit, M. Delince, D.A. Pensinger, T.N. Huynh, A.R. Jurado, Y.A. Goo, M. Sadilek, A.T. Iavarone, J.-D. Sauer, L. Tong[$] & J.J. Woodward.[$] (**2014**). The cyclic dinucleotide c-di-AMP is an allosteric regulator of metabolic enzyme function. ***Cell***, **158**, 1389-1401. (**–equal first authors, [$]–co-corresponding authors*) PMCID: 4166403.
    b. G. Jogl & L. Tong. (**2003**). Crystal structure of carnitine acetyltransferase and implications for the catalytic mechanism and fatty acid transport. ***Cell***, **112**, 113-122.
    c. G.A. Amodeo,* M.J. Rudolph* & L. Tong. (**2007**). Crystal structure of the heterotrimer core of the *Saccharomyces cerevisiae* AMPK homologue SNF1. ***Nature***, **449**, 492-495. (**–equal first authors*)
    d. J. Wei, S. Leit, J. Kuai, E. Therrien, S. Rafi, H.J. Harwood Jr, B. DeLaBarre & L. Tong. (**2019**). An allosteric mechanism for potent inhibition of human ATP-citrate lyase. ***Nature***, **568**, 566-570.

5. I have made important contributions to several other areas of modern biology. While at Boehringer Ingelheim Pharmaceuticals, I determined the structure of p38 MAP kinase in complex with a highly selective inhibitor, revealing for the first time how one can achieve strong selectivity with compounds that target the ATP binding site of protein kinases. With another series of compounds, we revealed a major conformational change in the active site region of the kinase that was necessary for inhibitor binding. I coined the term ‘DFG-out conformation’ for this new structure, which has been adopted by the field. During

my first years at Columbia, we studied the molecular mechanism of signal transduction by the Toll-like receptors, which have important functions in innate immunity. We determined the first structure of the Toll/interleukin-1 receptor (TIR) domain, and I coined the term 'BB loop' as the name of a loop with important roles in protein-protein interaction for signaling. This term has also been adopted by scientists in the field. We discovered serendipitously the protocol of *in situ* partial proteolysis for crystallization, as did several other labs, and this protocol is now in common use in the field. In addition, I have developed new methodologies (locked translation function, combined molecular replacement) and computer software for protein crystallography (GLRF and COMO), which has been distributed to many laboratories around the world.

a. C. Pargellis,* L. Tong,* L. Churchill, P.F. Cirillo, T. Gilmore, A.G. Graham, P.M. Grob, E.R. Hickey, N. Moss, S. Pav & J. Regan. (**2002**). Inhibition of p38 MAP kinase by utilizing a novel allosteric binding site. ***Nature Struct. Biol.*** **9**, 268-272. (**–co-corresponding authors*)
b. Y. Xu, X. Tao, B. Shen, T. Horng, R. Medzhitov, J.L. Manley & L. Tong. (**2000**). Structural basis for signal transduction by the Toll/Interleukin-1 receptor (TIR) domains. ***Nature,*** **408**, 111-115.
c. C.R. Mandel, D. Gebauer, H. Zhang & L. Tong. (**2006**). A serendipitous discovery that *in situ* partial proteolysis is essential for the crystallization of yeast CPSF-100 (Ydh1p). ***Acta Cryst.*** **F62**, 1041-1045.
d. G. Jogl, X. Tao, Y. Xu & L. Tong. (**2001**). COMO: A program for combined molecular replacement. ***Acta Cryst.*** **D57**, 1127-1134.

Complete List of Published Work: (316 total, 268 research papers, 48 reviews/book chapters)
http://www.ncbi.nlm.nih.gov/sites/myncbi/liang.tong.1/bibliography/41159164/public/?sort=date&direction=ascending