BIOGRAPHICAL SKETCH

NAME: **DIMITRIOS ANASTASAKIS**

eRA COMMONS USERNAME (credential, e.g., agency login): **anastasakisdg**

POSITION TITLE: **Postdoc Visiting Fellow (VF)**

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE	Start Date	Completion Date	FIELD OF STUDY
University of Thessaly, Larissa, Greece	**B.Sc.**	09/2004	02/2008	Biochemistry and Biotechnology
Yale University New Haven, CT, USA	(Postgrad. Associate)	4/2008	5/2009	Reproductive Science
University of Patras, Patras, Greece	**M.Sc**	09/2009	09/2011	Biomedical Sciences
University of Patras, Patras, Greece	**PhD**	12/2011	03/2016	Biomedical Sciences
NIAMS, National Institute of Health, Bethesda, MD, USA	(Postdoctoral Fellow)	12/2016	present	RNA Molecular Biology

A. Personal Statement

I am in my final years of my Postdoctoral training and I aspire to gain independence within the next 2 years. My research interests relate to how RNA-binding proteins impact post-transcriptional gene regulation (PTGR) in health and disease. Since my bachelor studies I was interested in mechanisms of PTGR. Thus, I joined a team to pursue related studies for my undergraduate thesis. During my postgraduate training at Yale University, I studied the role of FSHR variants in infertility and I got interested in the molecular mechanisms of human disease. I pursued a MSc and PhD on RNA biology and finished my PhD characterizing a novel deadenylase. After my PhD I was determined to follow a career in RNA biology and PTGR networks under normal cellular conditions but also in the contexts of development, immune response and cancer. Thus, I applied to and joined the RNA Molecular Biology team lead by Markus Hafner at the NIAMS/NIH.

During my first years at NIH, I trained in high-throughput methodologies, I became independent in computational analysis and developed my own pipelines to facilitate analysis of complicated experimental datasets. I developed new experimental protocols and optimized already established ones. I successfully collaborated with four different research groups and published thirteen scientific papers. I mentored post-bachelor and summer students through their projects. Finally, during the current pandemic, I conceived and initiated a Covid-19 project that was granted exempt research importance by the NIH. Recently I have been pursuing studies on role of nuclear PKM2 in PTGR and metastasis, a promising project in the field of molecular cancer. Thus, I have decided to continue my postdoctoral training focusing on getting acquainted on experimental systems reflecting carcinogenesis and disease outcome. I aim to collaborate with experts to get trained on structural and biophysical characterization of RNA binding proteins and on methodologies and protocols where I can measure the impact of a molecular mechanism in oncogenesis and metastasis both *in vitro* and *in vivo*. Gaining experience in these topics will provide me with all the necessary tools to lead a group that will perform state of art high impact multidisciplinary research, integrating basic science in the fields of biochemistry, RNA biology and computational biology with cancer-targeted research.

B. Positions, Scientific Appointments and Honors

9/2006-7/2007	B.Sc project Department of Biochemistry and Biotechnology, University of Thessaly, Greece.
7/2007-8/2007	Summer practice Department of Respiratory Medicine, University of Thessaly
4/2008 -5/2009	Postgraduate associate Laboratory research. Department of Obstetrics, Gynecology and Reproductive Science, School of Medicine, Yale University, USA.
10/2009-10/2011	M.Sc. Thesis Department of Biochemistry, School of Medicine, University of Patras, Greece.
5/2010-10/2010	Military service, Laboratory assistance 411 General Military Hospital, Tripoli, Greece.
1/2011-12/2013	Laboratory staff Diagnosis services at the Unit for Special Biochemical tests. Department of Biochemistry, School of Medicine, University of Patras, Greece
11/2011-02/2016	PhD Thesis, Department of Biochemistry, School of Medicine, University of Patras, Greece.
12/2016-Present	Postdoctoral fellow, National Institute of Arthritis and Musculoskeletal and Skin Diseases, National Institutes of Health, USA

Scholarship awarded to attend summer schools - forums:

1. The Onassis Lecture Series: The 2012 Lectures in Biology: A WORLD OF RNAs, Heraklion Crete, July 9-13
2. World Hellenic Biomedical Association, 2013, 2nd Summer School in Biomedical Research & Management, May 26 –June 4, 2013–Monemvasia & Sparta, Greece
3. 14th FEBS Young Scientists' Forum (YSF). Paris 27-30 August 2014

C. Contributions to Science

1. **Early Career:** Since my undergraduate studies I was interested in RNA processing and I characterized and evaluated natural and synthetic deadenylase inhibitors that can modulate deadenylation.
   Selected publications
   Balatsos N. A. A., **Anastasakis D**. and Stathopoulos C. (2009) Inhibition of human poly(A)- specific ribonuclease (PARN) by purine nucleotides: kinetic analysis. J. Enz. Inh. Med. Chem. 24: 516-523.
   Balatsos N. A. A., Vlachakis D., Maragozidis P., Manta S., **Anastasakis D**., Kyritsis A., Vlassi M., Komiotis D. and Stathopoulos C. (2009) Competitive inhibition of human poly(A)- specific ribonuclease (PARN) by synthetic fluoro-pyranosyl nucleosides. ***Biochemistry*** 48: 6044-6051.
   Balatsos N. A. A., Maragozidis P., **Anastasakis D**. and Stathopoulos C. (2012) Modulation of poly(A)-specific ribonuclease (PARN): current knowledge and perspectives. ***Current Med. Chem.*** 94: 214-221.'

2. **Graduate Career:** As a postgraduate associate at Yale University, I found that FSHR variant in heterozygous form found in women with low response to FSH impairs the function of the normal receptor. During my PhD, I characterized a novel deadenylase called PNLDC1. After I completed the biochemical characterization of PNLDC1, I found that it is the only exonuclease characterized so far that is epigenetically regulated and expressed only in early development and meiotic spermatocytes. I also showed that reduction of this enzyme in stem cells disrupts normal cell cycle and chromosome integrity surveillance-related genes. I was also involved in a research project where we successfully sequenced tRNA molecules and tRNA fragments from lung cancer tissues in order to identify deferential expression patterns with a potential use as molecular biomarkers.
   Selected publications:

Anastasakis D., Skeparnias I., Shaukat A-N., Grafanaki K., Kanellou A., Taraviras S., Papachristou D., Papakyriakou A. and Stathopoulos C. (2016) Mammalian PNLDC1 is a novel poly(A) specific exonuclease with discrete expression during early development. ***Nucleic Acids Res***. 44: 8908-892

Skeparnias*, I., **Anastasakis*, D**., Grafanaki, K., Kyriakopoulos, G., Alexopoulos, P., Dougenis, D., Scorilas, A., Kontos, C.K. and Stathopoulos, C. (2020) Contribution of miRNAs, tRNAs and tRFs to Aberrant Signaling and Translation Deregulation in Lung Cancer. ***Cancers*** (Basel), 12.

* equal contribution

3. **Postdoctoral Career:**

A. Technology development to study the dynamics of RNA processing

One of my most important contributions is the development and improvement of techniques for accurate mapping of Protein-RNA interactions in cells and capturing dynamic events of early RNA processing such as sequencing of intron lariats. I was able to significantly improve the sensitivity of the well-established method PAR-CLIP that is superior for the study of RNA binding proteins and established protocols for the accurate mapping of RNPs at any stage and cellular compartment with great accuracy.

Selected publications:

Benhalevy, D., **Anastasakis, D.G.,** and Hafner, M. (2018). Proximity-CLIP provides a snapshot of protein-occupied RNA elements in subcellular compartments. ***Nat Methods*** 15, 1074–1082

Anastasakis, D.G., Jacob, A., Konstantinidou, P., Meguro, K., Claypool, D., Cekan, P., Haase, A.D. and Hafner, M. (2021) A non-radioactive, improved PAR-CLIP and small RNA cDNA library preparation protocol. ***Nucleic Acids Res***, 27:gkab011

Wan Y*, **Anastasakis D.G***, Rodriguez J, Palangat M, Gudla P, Zaki G, Tandon M, Pegoraro G, Chow C.C, Hafner M, Larson D.R. Dynamic imaging of nascent RNA reveals general principles of transcription dynamics and stochastic splice site selection. ***Cell*** (in press)

doi: https://doi.org/10.1016/j.cell.2021.04.012

* equal contribution

B. Impact of RNA binding proteins in cancer

Having improved the techniques for global mapping of RBP binding sites in any cellular compartment, I was able to study RNA binding abnormalities that are involved in cancer development. Most importantly I was able to reveal an unconventional role of the splicing factor U2AF1 in translational regulation. Using protocols that I developed, I demonstrated that U2AF1 retention on cryptic splicing sites near the start codon in the cytoplasm inhibits translation of the associated mRNAs. This mechanism is impaired in cancer cells with mutations in U2AF1. This discovery has great impact in the research of molecular mechanisms implicated in myeloid malignancies.

Selected publications:

Palangat, M., **Anastasakis, D.G**., Fei, D.L., Lindblad, K.E., Bradley, R., Hourigan, C.S., Hafner, M., and Larson, D.R. (2019). The splicing factor U2AF1 contributes to cancer progression through a noncanonical role in translation regulation. ***Genes Dev***. 1;33(9-10):482-497

Muys, B.R., **Anastasakis, D.G**., Claypool, D., Pongor, L., Li, X.L., Grammatikakis, I., Liu, M., Wang, X., Prasanth, K.V., Aladjem, M.I. *et al.* (2021) The p53-induced RNA-binding protein ZMAT3 is a splicing regulator that inhibits the splicing of oncogenic CD44 variants in colorectal carcinoma. ***Genes Dev***, **35**, 102-116.

Complete List of Published Work in My Bibliography:

https://www.ncbi.nlm.nih.gov/myncbi/1foLnQxMXHAEhs/bibliography/public/