## BIOGRAPHICAL SKETCH

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NAME: **Georgieva, Elka Radoslavova**

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

POSITION TITLE: Assistant Professor in Chemistry and Biochemistry, Adjunct Assistant Professor in Cellular Physiology and Molecular Biophysics

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 |
|---|---|---|---|
| Sofia University, Sofia, Bulgaria | M.S. | 06/1998 | Chemistry and Physics |
| Bulgarian Academy of Sciences, Sofia, Bulgaria | Ph.D. | 12/2005 | Chemistry |
| Stockholm University, Stockholm, Sweden | Postdoctoral | 01/2006-09/2007 | Biophysics/Biochemistry |
| Cornell University, Ithaca, NY, USA | Postdoctoral | 10/2007-10/2010 | Biophysics/Biochemistry |

### A. Personal Statement

I am Assistant Professor in the Department of Chemistry and Biochemistry at Texas Tech University (TTU), and Adjunct Assistant Professor in the Department of Cell Physiology and Molecular Biophysics at TTU Medica School (TTUHSC). I am leading a research program to study the structure, function and interaction with drugs of membrane proteins from human pathogens. My expertise encompasses biophysical, biochemical and molecular biology techniques and methods applied to physiologically important systems at the nanoscale.

I have unique comprehensive expertise in the state-of-the-art biological electron paramagnetic resonance (EPR) spectroscopy, pulse and continuous wave, and proficiency in the development of EPR-based methods to characterize in details the structure and function of complex biomolecules and biomolecular ensembles. I was trained and conducted research in world-leading EPR and biochemical laboratories, at the forefront of modern biomedical studies. I am also proficient in protein engineering and protein biochemistry, protein chromatography, mutagenesis, circular dichroism (CD) spectroscopy, dynamic light scattering (DLS), fluorescence spectroscopy, functional assays on membrane transporters, negative staining electron microscopy on membrane proteins, and study of protein-drug interactions.

In the last more than 10 years, I have been a PI (R03 AI137735, 2018-2020), co-Investigator (R01 GM123779-35, 2017-2020, and R01GM124310, 2018-2020), and Key Personnel (on the NIH grant supporting the National Center for Advanced ESR Technology, ACERT, at Cornell University, 2011-2016) on NIH research grants, which demonstrates my capability to successfully conduct independent and collaborative research. To all these NIH-funded projects, I made major contributions by conducting the conception, design, and implementation of original studies on protein structure, structural dynamics and function. I demonstrated my leadership abilities while at Cornell University by establishing within ACERT a biochemical laboratory for membrane protein production and spin labeling, which was based on my original idea and is still functioning; then I had my own NIH research grant and supervised research personnel; now I am the advisor of four Graduate Students who, since joining my lab, have made significant advancement of their research projects, acquired new skills, and gave presentations at international and local meetings. I also established very successful and fruitful collaborations.

Given my extensive experience and expertise, I am determinedly poised to successfully lead and carry out the proposed research.

Recently completed projects that I would like to highlight include:

R03 AI137735 Georgieva (PI) 06/11/2018 – 05/31/2021
*I stopped working on this project in August 2020 due to accepting a faculty position at TTU.*
**Production and functional characterization of the L-lysine exporters from bacterial pathogens**
NIH NIAID
The overall objective of this study was to elucidate the functional mechanism(s) of L-lysine membrane exporters (LysE) from *Mycobacterium tuberculosis* and other pathogenic bacteria.
Role: PI

R01 GM123779-35 Freed (PI) 07/01/2017 – 03/31/2021
*I completed the work on this project in August 2020 due to accepting a faculty position at TTU.*
**Electron Spin Relaxation in Model Membranes**
NIH NIGMS
This project was aimed at establishing a quantitative physical understanding of membrane and membrane proteins dynamics using modern ESR methods.
Role: Co-Investigator

**Citations:**

a. **Elka R. Georgieva*,** Peter P. Borbat, Christina Fanouraki, Jack H. Freed. (**2020**) High-yield production in *E. coli* and characterization of full-length functional p13II protein from human T-cell leukemia virus type 1. *Prot Expr Purif*, 173, 105659
b. **Elka R. Georgieva*,** Akram Bani Ahmad, Oluwatosin Adetuyi, Saman Majeed (**2022**) Production of recombinant *Mtb* membrane efflux pump for structural and functional studies to reveal mechanisms of drug resistance *FASEB J*, 36, S1, Abstract issue
c. Saman Majeed, Akram Bani Ahmad, Oluwatosin Adetuyi, Eric G. Evans, Stefan Stoll, **Elka R. Georgieva*** (**2022**) Protein engineering and biochemical/biophysical approaches for structural studies of small membrane proteins and their complexes: Application to viroporins, *FASEB J*, 36, S1, Abstract issue
d. **Elka R. Georgieva*,** Christina Fanouraki, Peter P. Borbat (2020) Expression, purification and initial characterization of LysE membrane exporter from *Mycobacterium tuberculosis*: Towards comprehensive functional and structural study *FASEB J*, 34, S1, Abstract Issue

## B. Positions, Scientific Appointments, and Honors

**Positions and Scientific Appointments**

2002 – 2005 Graduate Student, Institute of Catalysis, Bulgarian Academy of Sciences, Sofia, Bulgaria

2006 – 2007 Visiting Postdoctoral Scientists, Department of Biochemistry and Biophysics Stockholm University, Stockholm, Sweden

2007 – 2010 Postdoctoral Associate, Department of Chemistry and Chemical Biology and ACERT, Cornell University, Ithaca, NY

2010 – 2012 Volunteer (Visiting Postdoctoral/Research Associate), Department of Physiology and Biophysics, Weill Cornell Medical College, New York City, NY

2010 – 2015 Research Associate, Department of Chemistry and Chemical Biology and ACERT, Cornell University, Ithaca, NY

2015 – 2016, March 4th Sr. Research Associate, Department of Chemistry and Chemical Biology and ACERT, Cornell University, Ithaca, NY

2016, March – February 2018, Department of Physiology and Biophysics, Weill Cornell Medical College, New York, NY

March 2018 – August 2020, Sr. Research Associate, Department of Chemistry and Chemical Biology, Cornell University, Ithaca, NY

September 2020 – Present, Assistant Professor, Department of Chemistry and Biochemistry, Texas Tech University, Lubbock, TX

July 2021 – Present, Adjunct Assistant Professor in Cell Physiology and Molecular Biophysics, TTUHSC, Lubbock, TX

**Other Experience and Professional Memberships**
2022 – Member, Royal Society of Chemistry (after admission based on scientific merits and recommendations)
2022 – Member, Deputy Editorial Board of the Journal of Structural Biology and Journal of Structural Biology X.
2021 – Member, Topic Editorial Board of the journal Membranes (MDPI).
2015 – Reviewer for multiple high-impact scientific journals
2013 – Member, Biophysical Society

**Honors**
**2003 Ph.D. Pre-doctoral Fellowship from the Marie Curie Foundation, European Commission**
Laboratory for Molecular Magnetism, University of Florence, Italy with Professor Dante Gatteschi (SIX MONTHS).
**2004 Ph.D. Pre-doctoral Fellowship from the Marie Curie Foundation, European Commission**
Max Planck Institute for Polymer Research, Mainz, Germany with Professor Gunnar Jeschke (SIX MONTHS).
**2007 Post-doctoral Fellowship from the Carl Trygger Foundation, Stockholm University**, Sweden with Professor Astrid Gräslund (NINE MONTHS)
**2010 Young Investigator Travel Grant from The Protein Society**, Finn World Travel Award to attend the 24th Annual Symposium of The Protein Society.

## C. Contributions to Science

### 1. Mechanisms of function and inhibition of membrane transporters, with emphasis on exporters from *Mycobacterium tuberculosis* (*Mtb*).

One of the major directions of my research program at TTU is to elucidate the structure and function of *Mtb* membrane exporters linked to drug- and multidrug-resistance (DR and MDR). My particular effort is directed toward the EfpA exporter, which belongs to the MFS and transports variety of drugs out of the cell for the antiport of $H^+$. Learning the structure and function of this protein in detail will help to understand one of *Mtb* mechanisms (through drug export) for DR and MDR. My research group successfully expressed the EfpA protein in *E. coli* and purified it to significant level, for the first time to the best of our knowledge.[a] Currently, we are conducting EfpA-drug binding experiments and aim to characterize its conformational dynamic upon drug-binding.

Prior to TTU, I conducted an independent study on the L-Lysine exporters (LysE-s) from *Mtb*. I developed protocol for purification of LysE in lipodisqs made of native *E. coli* membranes and found that in lipid environment the protein forms oligomeric structure.[b] (Supported by the R03 AI137735 grant, E. Georgieva PI)

In a collaborative effort, I also studied the sodium-coupled aspartate transporter GltPh. The study established that the subunits of this homo-trimeric protein function independently and sample outward and inward facing conformation with almost equal probability regardless of the presence of ligands, which was priorly not known.[c]

a. **Elka R. Georgieva*,** Akram Bani Ahmad, Oluwatosin Adetuyi, Saman Majeed (2022) Production of recombinant Mtb membrane efflux pump for structural and functional studies to reveal mechanisms of drug resistance *FASEB J*, 36, S1, Abstract issue

b. **Elka. R. Georgieva***, C. Fanouraki, P.P. Borbat (**2020**). Expression, purification and initial characterization of LysE membrane exporter from Mycobacterium tuberculosis: Towards comprehensive functional and structural study. *The FASEB Journal* 34(S1):1-1 (Abstract from the Annual Meeting of American Society for Biochemistry and Molecular Biology).

c. **Elka R. Georgieva**, P.P. Borbat, C. Ginter, J.H. Freed, O. Boudker. (**2013**). Conformational ensemble of the sodium-coupled aspartate transporter. *Nat Struct Mol Biol* 20:215-221.

### 2. Mechanisms of function and inhibition of viral membrane proteins.

Another major direction of my research at TTU is to study membrane proteins encoded by human viruses. These proteins, however, are expressed in the host and function in the cellular membranes. Particularly, my focus is on elucidating the molecular mechanisms of viroporins. These are small viral proteins forming homo-oligomers

in the plasma, endo and/or mitochondrial membranes, and by doing so, they increase membrane permeability to ions, thus affecting ion homeostasis and controlling signaling events.

My research group succeeded in designing fusion constructs to express these highly hydrophobic proteins in soluble form in *E. coli*, and we identified membrane conditions, under which the proteins form stable oligomers visualized by negative staining electron microscopy.[a] We are currently working on HIV-1 Vpu and HVC p7 proteins.

Previously, I produced and conducted *in vitro* characterization of HTLV-1-encoded protein p13II,[b] a proposed viroporin.

I also developed a very successful project to elucidate the mechanism of assembly of the Influenza A-encoded M2 proton channel (also viroporin), revealing that this protein forms functional tetramers via dimer intermediates.[c] It was further found in my studies that binding of anti-influenza drug amantadine to M2 under acidic conditions (pH 5.5) stabilizes the tetrameric channel form of M2 and closes the C-terminal proton exit pore.[d]

a. Saman Majeed, Akram Bani Ahmad, Oluwatosin Adetuyi, Eric G. Evans, Stefan Stoll, **Elka R. Georgieva*** (**2022**) Protein engineering and biochemical/biophysical approaches for structural studies of small membrane proteins and their complexes: Application to viroporins, *FASEB J*, 36, S1, Abstract issue

b. **Elka R. Georgieva*,** Peter P. Borbat, C. Fanouraki, Jack H. Freed. (**2020**) High-yield production in *E. coli* and characterization of full-length functional p13II protein from human T-cell leukemia virus type 1. *Prot Expr Purif*, 173, 105659.

c. **Elka R. Georgieva***, Peter P. Borbat, Haley D. Norman, Jack H. Freed*. (**2015**). Mechanism of influenza A M2 transmembrane domain assembly in lipid membranes. *Sci Rep,* 5:11757.

d. **Elka R. Georgieva***, Peter P. Borbat, Kiril Grushin, Svetla Stoilova-McPhie, Nichita J. Kulkarni, Zhichun Liang, Jack H. Freed*. (**2016**). (2015). Conformational response of influenza A M2 transmembrane domain to amantadine drug binding at low pH (pH 5.5). *Front Physiol,* 7:317. doi: 10.3389/fphys.2016.00317.

**3. Structure and function of proteins involved in neurological disorders**.

During my PostDoc and then senior appointment at Cornell University, I conducted studies on the structure and function of proteins that play important physiological roles in nervous system. In collaboration with Dr. David Eliezer from Weill Cornell Medical College, I studied the structure and function of human alpha-Synuclein, a synaptic vesicle-associated protein, and tau, a microtubule-associated protein. Besides their very important physiological functions, both proteins are prone to misfolding and formation of insoluble aggregates linked to severe pathological conditions, such as Parkinson's disease, Alzheimer's disease and others. Under physiological conditions, these proteins interact with biological membranes. I elucidated the conformational dynamics upon transition from soluble to membrane-bound state of alpha-Synuclein and tau, and characterized for the first time their long-range structure in membrane-bound state.[a,b,c]

I also collaborated with Dr. Song-I Han from the University of Santa Barbara, CA to study the early stages of human tau protein aggregation. This is relevant to tau's role in neurodegenerative disorders. We studied two key fibril-forming regions, PHF6 and PHF6*, of tau protein. We found that both the PHF and PHF6* hexapeptide regions assume a comparably compact conformation in stable solution state that completely transforms into a fully extended conformation found in β-sheet fibrils, within minutes of initiating aggregation by the addition of heparin, i.e. well before any fibrillar species or aggregates are detectable.[d]

a. **Elka R. Georgieva**, Trudy F. Ramlall, Peter P. Borbat, Jack H. Freed, David Eliezer. (**2008**). Membrane-bound alpha-synuclein forms an extended helix: long-distance pulsed ESR measurements using vesicles, bicelles, and rodlike micelles. *J Am Chem Soc* 130:12856-12857.

b. **Elka R. Georgieva**, Trudy F. Ramlall, Peter P. Borbat, Jack H. Freed, David Eliezer. (**2010**). The lipid-binding domain of wild type and mutant alpha-synuclein: compactness and interconversion between the broken and extended helix forms. *J Biol Chem* 285:28261-28274.

c. **Elka R. Georgieva**, Shifeng Xiao, Peter P. Borbat, Jack H. Freed, David Eliezer. (**2014**). Tau binds to lipid membrane surfaces via short amphipathic helices located in its microtubule-binding repeats. *Biophys J* 107:1441-1452.

d. Neil A. Eschmann, **Elka R. Georgieva**, Pritam Ganguly, Peter P. Borbat, Maxime Rappaport, Yasar Akdogan, Jack H. Freed, Joan-Emma. Shea, Song-I Han. (**2017**). Signature of an aggregation-prone conformation of tau. *Sci Rep* 7:44739.

**4. Developing EPR based dosimeters to estimate exposure to high-energy radiation**.

During my training as Graduate Student in the laboratory of Professor Nicola D. Yordanov at the Institute of Catalysis, Bulgarian Academy of Sciences, I developed dosimeters, based on saccharide materials, which are sensitive to high energy-radiation. Upon exposure to high-energy particle emission, stable free radicals are formed in these saccharide materials. In the course of my research, I established and characterized in detail the correlation between EPR signal intensity of the radiation-induced radicals and absorbed radiation doses under laboratory conditions for solid saccharides treated with known doses of gamma-radiation. Thereafter, I developed self-calibrated dosimeters, based on sucrose/manganese(II) mixtures, with high sensitivity for emergency and routine dosimetry. Furthermore, I collaborated with and was trained by experts in high-field EPR spectroscopy to conduct a study aiming to better understand the nature of gamma radiation-generated radicals in sucrose. These efforts also contributed significantly to the understanding of the radiation chemistry of this compound.

a. **Elka R. Georgieva***, Luca Pardi, Gunnar Jeschke, Dante Gatteschi, Lorenzo Sorace, Nicola D. Yordanov. (**2006**). High–field/high-frequency EPR study on stable free radicals formed in sucrose by gamma-irradiation *Free Radical Res,* 40: 553-563. (**Funded by the Marie Curie, European Commission, fellowship to ERG**)

b. Nicola D. Yordanov, **Elka Georgieva**. (**2004**). EPR and UV spectral study of gamma-irradiated white and burned sugar, fructose and glucose *Spectrochim Acta Part A: Mol Biomol Spectroscopy*, 60: 1307-1314.

c. Nicola D. Yordanov, Veselka Gancheva, **Elka Georgieva**. (**2002**). EPR and UV spectroscopic study of table sugar as a high-dose dosimeter *Radiat Phys Chem*, 65: 269-276.

Complete list of published work in My Bibliography:
https://www.ncbi.nlm.nih.gov/myncbi/1Zq7b7-v9pxQz/bibliography/public/