BIOGRAPHICAL SKETCH

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NAME: Brunk, Elizabeth C

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

POSITION TITLE: Assistant Professor of Chemistry and Pharmacology

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)*

INSTITUTION AND LOCATION	DEGREE	Completion	FIELD OF STUDY
University of Michigan	BS	05/2007	Biochemistry
Ecole Normale Supérieure, France	MS	06/2008	Chemistry
Ecole Polytechnique Fédérale de Lausanne	PHD	07/2013	Biochemistry
University of California, Berkeley	Visiting Scholar	05/2014	Genomics
University of California, San Diego	Postdoc	05/2017	Genomics
Moores Cancer Center, San Diego	Postdoc	07/2020	Genomics

A. Personal Statement

Research

My laboratory investigates how structural DNA variations, particularly extrachromosomal DNA (ecDNA), contribute to genetic heterogeneity and adaptability within cell populations. Our primary focus is on double minute chromosomes (DMs), a subtype of ecDNA known for their uneven inheritance during cell division and their role in driving extreme gene copy number variance within multicellular systems. Despite their importance across species, ecDNA remains challenging to study due to their small size, dynamic behavior, and lack of methods to directly quantify or manipulate them in live cells. Since founding my lab in 2021, we have addressed these challenges by developing new approaches that integrate computational tools, high-resolution imaging, and single-cell techniques.

I have a proven track record of diverse, productive, and high-impact research, as evidenced by a Google Scholar H-index of 22, 39 publications, and over 3,400 citations. My laboratory has published five independent papers, with three additional manuscripts currently under review and available in bioRxiv.

One of my research team's most significant contributions is a study currently under consideration at Nature. We created an approach to sort and isolate live cells based on ecDNA dosage, allowing us to enrich for distinct subpopulations and, for the first time, directly compare their functional behavior. We developed a proxy for ecDNA, enabling FACS-based enrichment of "high" and "low" ecDNA without disrupting living cells. We found that ecDNA dosage directly scales with molecular phenotypes, shaping transcriptional, epigenetic, and cell state differences across individual cells. Using a multimodal approach, we captured how ecDNA drives continuous, graded shifts from DNA to RNA to chromatin to protein levels, and altered growth and cell cycle behavior in single cells.

More surprising, when we allowed these isogenic ecDNA-variant subpopulations to continue growing, they spontaneously returned to the original heterogeneous ecDNA distribution seen before sorting. This recovery happened within just 2-3 cell divisions. It is reproducible, striking, and fundamentally challenges the long-held assumption that ecDNA divides in an entirely random way. Through stochastic modeling, simulations revealed that such rapid redistribution of ecDNA variation could only be explained if ecDNA inheritance was biased and if division, (not death), is a dominant force shaping population recovery.

This work opens the door to a new space of questions that could not be asked before and this proposal will allow us to pursue them. We will test whether ecDNA inheritance is truly random or biased, investigate how ecDNA dosage is linked to cell division, and identify the molecular factors that allow populations to restore ecDNA heterogeneity so quickly. Our preliminary work has already nominated 25 candidate genes that may play a role in maintaining ecDNA variation, and we are now ready to test them mechanistically.

At a broader level, my long term research goal is to determine whether cell populations use ecDNA as a bet-hedging strategy, actively maintaining large-range variation in dosage to enable adaptability in uncertain environments. If so, this work could change how we think about genome regulation and evolutionary dynamics in multicellular systems. As mounting evidence emerges that ecDNA is more widespread across species than previously thought, our studies will have implications far beyond a single system or model.

We are well-positioned to carry this work forward. As PI, I bring a hybrid background in computation and experimental biology. I trained in AI and multi omics and gained hands-on experience in molecular biology, imaging, and sequencing during my F32 postdoctoral fellowship. This dual perspective defines how our lab operates. We design and execute all core experiments in-house, working closely with core facilities and co-investigators who bring deep expertise in single-cell sequencing, flow cytometry, and cell cycle dynamics. We have already piloted many of the experiments outlined in this proposal and built a team with the skills and infrastructure needed to deliver on this next phase of discovery.

Teaching, Mentoring and Outreach

Through my teaching, mentoring and outreach activities, I demonstrate a deep commitment to fostering the development of coding and data literacy skills in the next generation of scientists to handle the “big data” of biology. Over the past three years, I have introduced more than 3,000 undergraduate students to data literacy in my large-enrollment biochemistry course. The curriculum and teaching materials I developed for this course features Jupyter Lab notebooks and access to the NSF-funded Cyverse supercomputer. Integrating “big data” skills into this undergraduate biochemistry course allows over 500 students each semester to gain vital computational skills for careers in biomedical and biological sciences. This work has been recently published in the ACS *Journal of Chemical Education*. Our data shows significant disparities in comfort, confidence, and knowledge of coding at the beginning of the class in students from all backgrounds; after completing the course, these gaps were entirely closed. We also see significant grade improvements after introducing these data skills along with a highly structured curriculum, highlighting the transformative power of integrating coding and data literacy into biological and chemical science education.

Beyond the classroom, my lab engages in multiple mentorship activities, collaborating with programs such as the Carolina Summer Fellows, WINSPIRE, EDGE Genomics, and highschools, such as NC School of Science and Math, lowering the barrier of entry to data science and genomics research to students of all backgrounds.

As a mentor, I emphasize technical expertise alongside the development of independence, scientific rigor, and effective communication. I see mentoring as equally important to other faculty-level activities and seek to improve my skills by attending yearly mentorship training through the Office of Graduate Education at the University of North Carolina at Chapel Hill. Currently, I serve as the primary mentor to four graduate students, three post-bacs and eight undergraduates. Additionally, I mentor undergraduate students within the AI@UNC club, a student-run organization that matches computer science students with faculty-led projects so that they can apply their knowledge to real-world problems. Lastly, I value all backgrounds and experience levels by creating inclusive research and publishing practices. Undergraduate students in my lab have completed honors theses, earning highest honors, and also receiving the prestigious Brantley Research Award. I was selected along with fifty-seven UNC Chapel Hill faculty, to be honored by Morehead-Cain undergraduate seniors for excellence in teaching and mentorship in 2024. In the past three years, more than six undergraduates have co-authored manuscripts with my lab (four as co-first authors).

B. Positions, Scientific Appointments, and Honors

Professional Experience:

2021– Present	**Assistant Professor**, Departments of Chemistry and Pharmacology, Integrative Program for Biological and Genome Sciences, Lineberger Comprehensive Cancer Center, Computational Medicine Program, Carolina Applied Math Program, University of North Carolina at Chapel Hill School of Medicine and College of Arts & Sciences
2019 – Present	**Adjunct Faculty**, Altman Translational Clinical Research Institute, University of California San Diego
2020 – 2021	**Research Data Analyst**, Moores Cancer Center, University of California San Diego
2018 – 2020	**Postdoctoral Fellow**, Moores Cancer Center, University of California, San Diego
2016 – 2018	**Industry Consultant**, Celgene (now Bristol Myers Squibb), San Diego

2015 – 2016 **Industry Consultant**, Center for Biosustainability, Coppenhagen, Denmark
2014 – 2017 **Postdoctoral Fellow**, Department of Bioengineering, University of California San Diego
2013 – 2014 **Visiting Scholar**, Joint Bioenergy Institute, University of California, Berkeley

Selected Honors, Awards and Service:

2025 – 2026 National Synthesis Center for Emergence in the Molecular and Cellular Sciences Award
2025 – 2026 PhRMA Drug Discovery Early Faculty Starter Award, PhRMA Foundation
2024 – 2025 Computational Medicine Pilot Award, UNC Chapel Hill
2023 – 2024 IBM Junior Faculty Development Award, Office of the Provost, UNC Chapel Hill
2021 – 2022 Office of Vice Chancellor of Research IDEA Grant, UNC Chapel Hill
2018 – 2020 Kirschstein-NRSA F32 postdoctoral fellowship, UCSD
2013 – 2015 Early Postdoc Mobility Award, Swiss National Science Foundation, UC Berkeley
2013 Swiss Academy of Sciences (SCNAT), Platform Chemistry Travel Award, EPFL
2013 Global Young Scientist Summit Award, National Research Foundation Singapore
2007 – 2008 Erasmus Mundus Atosim Masters Scholarship, ENS France
2005 Undergraduate Research Opportunities Fellowship, University of Michigan

Grant Advisory Committees:

2025 Early Career Reviewer for Special Emphasis Panel on Human Genomics and Genetics, NIH RO1 & RO3 Grants
2022 Ad Hoc Reviewer for Department of Defense Breast Cancer Breakthrough Award

C. Contributions to Science

A full list of my publications at **NCBI**: https://www.ncbi.nlm.nih.gov/myncbi/1j3l47nY-e2knN/bibliography/public/
ORCID ID: 0000-0001-8578-865

1. Identifying and Deciphering Impacts of Extrachromosomal DNA (ecDNA) in Single Cells

Over the past several years, my lab has developed a comprehensive hybrid computational-experimental toolkit to detect, characterize, and understand the functional impact of extrachromosomal DNA (ecDNA). We developed a sample preparation protocol for Scanning Electron Microscopy (SEM) and Correlative Light Electron Microscopy (CLEM) that preserves the fragile structure of ecDNA during intense fixation [1] to resolve ecDNA ultrastructure. We developed a computational convolutional neural networks-based approach that outperforms standard tools in detecting ecDNA in metaphase FISH images achieving (~92% accuracy) [2]. We developed CytoCellDB, the largest experimentally-anchored ecDNA dataset in human cells to date, that links genomic and karyotype data from large-scale resources like DepMap and CCLE [3]. Most recently, we developed a live-cell sorting strategy that isolates isogenic subpopulations with high vs low ecDNA levels, enabling the first multi omic dissection of how ecDNA dosage shapes cell state and adaptive behavior. [4].

(1) Madren JA, Chen J, Dennis WC, Ford C, White K, **Brunk E**
A Standardized Protocol for Sample Preparation for Scanning Electron Microscopy (SEM) to Visualize Extrachromosomal DNA (ecDNA).
Biotechniques (2024)

(2) Gobel K*, Mehta A*, Guilbaud D, Fessler J, Chen J, Cope O, Nenad W, Cheng D, Dennis W, Gurumurthy N, Wang Y, Shukla K, **Brunk E.**
AI for Drug Response: Automated Detection and Quantification of Extrachromosomal DNA in Fluorescent Microscopy Images Using Convolutional Neural Networks
Frontiers in Systems Pharmacology (2024)

(3) Fessler J*, Ting S*, Yi H, Haase S, Chen J, Gulec S, Wang Y, Smyers N, Goble K, Cannon D, Mehta A, Ford, C, **Brunk, E**
CytoCellDB: a comprehensive resource for exploring extrachromosomal DNA in cancer cell lines
NAR Cancer (2024)

(4) Wang Y, Cope O, Chen J, Mehta A, Fleifel D, Ford CG, Benhamie P, Haase S, Gulec S, Elston T, Spanheimer P, Tomblin CA, Rojas AM, Tate T, Purvis J, Wang J, Dahl JM, Cook JG, **Brunk E**
Extrachromosomal DNA Gives Cancer a New Evolutionary Pathway
bioRxiv **[Preprint]**. (2025); *In review at* ***Nature***

2. AI-Driven Integration of Genomic and Structural Data to Predict Variant Impacts on Cell States

My lab has developed integrative AI frameworks to decipher how genetic variation shapes cell state by bridging genome, transcriptome, and protein structural data. We combined WGS and RNA-seq with 3D structural data from crystallography, NMR, and AlphaFold to predict how specific variants (especially SNPs) alter transcriptional regulatory networks. Using this approach, we identified variants linked to changes in key regulators like MYC and NRF2 [5], ESR1 and EZH2 [6]. We applied single-cell transcriptomics across large-scale cell lines and human population datasets to uncover transcriptional heterogeneity and its predictive power for drug response [7]. We have also mapped variant effects on metabolic states by integrating protein structure with genome-scale metabolic models, revealing critical residues that control cellular metabolic resilience [8]. This approach has been highly influential, published in *Nature Biotechnology* with **over 680 citations**.

(5) Shukla K, Idanwekhai K, Naradikian M, Ting S, Schoenberger SP, **Brunk E** (2023)
Machine learning of three-dimensional protein structures to predict the functional impacts of genome variation.
ACS J. Chem. Inf and Mod (2024)

(6) Shukla K, Wang Y, Spanheimer P, **Brunk E**
AI-driven Variant Annotation for Precision Oncology in Breast Cancer
bioRxiv **[Preprint]**. (2025); *In review at* ***Clinical Translational Medicine***

(7) Wang Y, Cope O, Chen J, Mehta A, Fleifel D, Ford CG, Benhamie P, Haase S, Gulec S, Elston T, Spanheimer P, Tomblin CA, Rojas AM, Tate T, Purvis J, Wang J, Dahl JM, Cook JG, **Brunk E**
A Multimodal Framework to Uncover Drug-Responsive Subpopulations in Triple-Negative Breast Cancer
bioRxiv **[Preprint]**. (2025); In review at ***iScience***

(8) **Brunk E**, Sahoo S, Zielinski D, Altunkaya A, Drager A, Mih N, Gonzales GAP, Aurich MA, Prlic A, Sastry A, Danielsdottir AD, Heinken A, Noronha A, Rose PW, Burley SK, Fleming RMT, Nielsen J, Thiele I, Palsson BO
Recon 3D enables a three-dimensional view of gene variation in human metabolism.
Nature Biotechnology (2018)

3. Mechanistic Studies of Genome-Wide Biology and Cellular Adaptation

My postdoctoral work laid the foundation for understanding how cellular phenotypes emerge from genome-scale interactions, particularly in microbial systems. We developed multi-omics frameworks that revealed how post-translational modifications tune metabolic networks in response to environmental changes [9], and how genomic and transcriptional variation among engineered *E. coli* strains influences cellular performance [10]. In a complementary study, we helped construct iML1515, a comprehensive computational model of *E. coli* integrating genome annotation with experimental data to predict whole-cell behavior [11]. These studies reflect a long-standing commitment to decoding how diverse regulatory layers, DNA, RNA, protein, and growth, interact to shape cell state, adaptation, and function. This systems-level perspective continues to inform my lab's current focus on genetic variation and phenotypic plasticity in more complex, multicellular contexts.

(9) **Brunk E**, Chang RL, Xia J, Hefzi H, Yurkovich JT, Kim D, Buckmiller E, Wang HH, Cho BK, Yang C, Palsson BO, Church GM, Lewis NE
Characterizing post-translational modifications in prokaryote metabolism using a multi-scale workflow.
PNAS (2018)

(10) **Brunk E***, George KJ*, Alonso-Gutierrez J, Thompson M, Baidoo E, Wang G, Petzold PJ, McCloskey D, Monk J, Yang L, O'Brien EJ, Batth TS, …, Adams PD, Keasling JD, Palsson BO, Lee TS
Characterizing Strain Variation in Engineered E. coli Using a Multi-omics Based Workflow.
Cell Systems (2016)

(11) Monk J*, Llyod C*, **Brunk E***, Mih N, Sastry A, King Z, Takeuchi R, Nomura W, …, Palsson BO
iML1515; a computable knowledge base for E coli.
Nature Biotechnology (2017)

4. Digital Upskilling: Introducing 3,000 Undergraduates to Data Science and Data Literacy Skills

Many undergraduates never get exposed to coding or data literacy, even though these skills are essential in modern science. To change that, I redesigned a large-enrollment biochemistry course to integrate coding exercises into the curriculum; an effort now published [12] and adopted by other instructors for other undergraduate and graduate-level courses. With over 500 students per term, we leveraged NSF's Cyverse supercomputing platform to provide equitable access to cloud-based coding tools across 4 years of teaching. The class is now Cyverse's largest recurring educational workshop, helping scale data skills across the life sciences.

(12) Brunk, R, Shukla, K, Hutson, B, Wang, Y, Verber, M, Gutierrez Ford, C, Dennis, W, Mehta, A, Hogan, B, Swetnam, T, **Brunk, E**.
Data science for Chemists: Integrating and evaluating the use of interactive digital python notebooks in a large enrollment undergraduate biochemistry course
ACS Journal of Chemical Education (2024)

5. Constraint-Based Perspectives on Gene Expression and Cellular Adaptation

My prior work laid the foundation for modeling how physical and structural constraints, such as protein folding, solubility, and ribosome occupancy, shape gene expression and cellular behavior.

We developed foundational frameworks for integrating multi-omic, structural, and machine learning data to reveal hidden constraints on protein expression, solubility, and cellular growth. In collaboration with the Human Protein Atlas, we used AI models to predict high-throughput protein expression outcomes across the human proteome [13]. We also advanced systems-level integration of protein structure into genome-scale models, uncovering how translational pausing and folding constraints shape growth dynamics and regulatory responses[14-15].

These insights into how molecular constraints shape emergent cell behavior laid the groundwork for our current efforts to decode how DNA-level variation (including ecDNA) propagates through molecular networks to drive adaptive states. ecDNA, as modeled in our stochastic models, imposes a unique burden on cellular systems due to its extreme gene dosage and non-chromosomal inheritance. By applying constraints and multi-omic integration, we can systematically decode how ecDNA alters resource allocation, transcriptional dynamics, and protein expression at scale. This perspective positions ecDNA not just as an amplification event, but as a genome-embedded perturbation whose systemic effects can now be mechanistically understood.

(13) Sastry A, Monk J, Tegel H, Uhlen M, Palsson BO, Rockberg J, **Brunk E**.
Computational Biology to Accelerate High-throughput protein expression.
Bioinformatics. (2017)

(14) **Brunk E***, Mih N*, Monk J, Chen K, Zhang Z, Bliven S, O'Brien E, Chang RL, Bourne PE, Palsson BO
Systems Biology of the Structural Proteome
BMC systems biology (2016)

(15) Ebrahim A*, **Brunk E***; Tan J*, O'Brien EJ, Donghyuk K, Lerman J, Lechner A, Sastry A, Bordbar A, Feist A, Palsson BO
Multi-omic data integration enables discovery of hidden biological regularities.
Nature Communications (2016)