

# Andrew Elia, MD, PhD



## Elia Laboratory

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In response to DNA damage from environmental or endogenous sources, cells evoke an elaborate signaling network known as the DNA damage response (DDR). This response functions to preserve genomic integrity, which is necessary for normal development and the prevention of cancer. **The Elia laboratory** studies the DNA damage response, with current projects focusing on DDR pathways regulated by ubiquitin-dependent signaling and DDR pathways that promote the stabilization and repair of stalled replication forks. We utilize innovative proteomic and genetic approaches to investigate these processes. Our ultimate goal is to understand how DDR disruption influences cancer progression and can be exploited to target tumors with specific DNA repair defects.

### DNA damage response

DNA within cells is under continual assault from metabolic and environmental sources. In response to the ensuing damage, cells activate a signaling network called the DNA damage response (DDR). Defects in this response can lead to numerous hereditary cancer syndromes and can underlie the genomic instability which is a hallmark of many sporadic cancers. The DDR promotes genomic integrity by targeting hundreds of factors in diverse pathways ranging from DNA replication and repair to cell-cycle arrest, senescence, and immune regulation. While much is known about these core pathways, the complex regulatory events coordinating them are less well understood. Our lab aims to elucidate biochemical and genetic relationships between DDR factors to understand how they are integrated and collectively regulated.

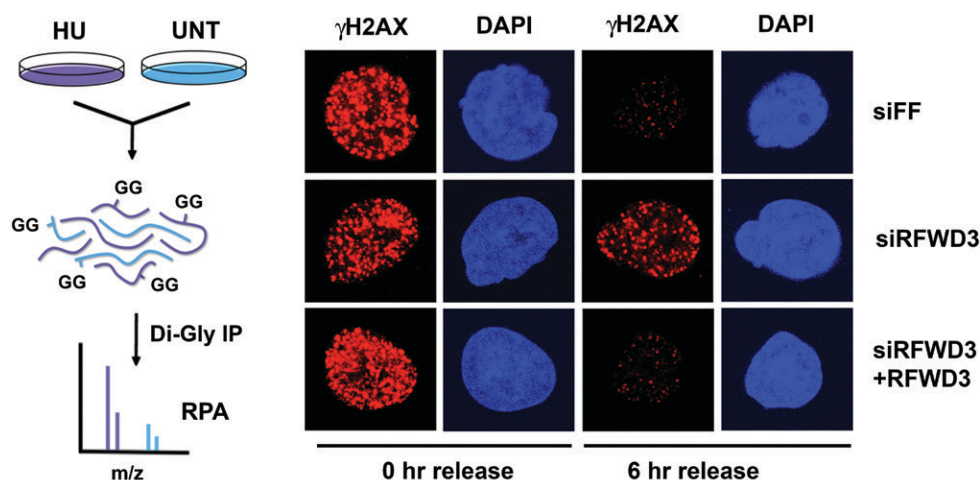
### Quantitative proteomics in ubiquitin signaling

Execution of the DDR relies upon a dynamic array of protein modifications, with phosphorylation playing a historically central role. It is now evident that the DDR also

depends on ubiquitin signaling. Numerous ubiquitin ligases have been implicated in the response, yet finding their substrates by simple binding techniques can be difficult due to weak substrate interactions. To circumvent this problem, we have pioneered a quantitative proteomic approach to globally profile ubiquitination. Initially, we used this approach to identify substrates of Cullin-RING ubiquitin ligases (*Cell* 2011), which are involved in numerous DNA repair processes. Subsequently, we used it to uncover novel ubiquitination events directly stimulated by DNA damage (*Mol Cell* 2015a), demonstrating the vast breadth of ubiquitin signaling in the DDR. We are continuing to use innovative proteomic approaches to characterize novel and poorly understood ubiquitin ligases in DNA damage signaling pathways.

### Replication stress and cancer

Replication fork collapse can induce chromosome instability and mutagenic events that cause cancer. Organisms have therefore evolved pathways to stabilize stalled replication forks and to repair collapsed forks through processes such as homologous recombination (HR). Multiple



(Left) Quantitative proteomics identifies RPA ubiquitination mediated by the ubiquitin ligase RFWD3, which is mutated in the cancer predisposition syndrome Fanconi anemia. (Right) Depletion of RFWD3 inhibits the repair of collapsed replication forks, as demonstrated by delayed resolution of  $\gamma$ H2AX foci six hours after release from hydroxyurea-induced replication fork stalling and collapse.

factors involved in HR and replication fork stabilization, such as BRCA1 and BRCA2, are mutated in hereditary cancer syndromes, highlighting the importance of these pathways. We have demonstrated that the ubiquitin ligase RFWD3, which is mutated in the cancer predisposition syndrome Fanconi anemia, ubiquitinates the single-stranded DNA binding protein RPA to promote homologous recombination at stalled replication forks and replication fork restart (*Mol Cell* 2015b). We are currently studying RFWD3 function in the replication stress response and elucidating novel mechanisms of replication fork stabilization and repair.

### Targeted cancer therapy

Defects in the DNA damage response can render tumors dependent upon specific DNA repair pathways for survival. Moreover, targeted modulation of the DDR can affect tumor sensitivity to genotoxic chemotherapy and radiation. Increased understanding of DNA repair pathways will lead to enhanced opportunities for developing therapies that target cancers with DNA repair defects, and for improving the efficacy of genotoxic

treatments. We are employing methods to translate our work to the development of such therapies.

### Selected Publications:

Duan H, Mansour S, Reed R, Gillis MK, Parent B, Liu B, Sztupinszki Z, Birkbak N, Szallasi Z, Elia AE, Garber JE, Pathania S. E3 ligase RFWD3 is a novel modulator of stalled fork stability in BRCA2-deficient cells. *J Cell Biol.* 2020; 219(6):e201908192.

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Elia AE, Wang DC, Willis NA, Boardman AP, Hajdu I, Adeyemi RO, Lowry E, Gygi SP, Scully R, Elledge SJ. RFWD3-Dependent Ubiquitination of RPA Regulates Repair at Stalled Replication Forks. *Molecular Cell.* 2015; 60(2):280-93.

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Emanuele MJ, Elia AE, Xu Q, Thoma CR, Izhar L, Leng Y, Guo A, Chen YN, Rush J, Hsu PW, Yen HC, Elledge SJ. Global identification of modular cullin-RING ligase substrates. *Cell.* 2011; 147(2):459-74.

Elia AE, Cantley LC, Yaffe MB. Proteomic screen finds pSer/pThr-binding domain localizing Plk1 to mitotic substrates. *Science.* 2003; 299:1228-31.

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