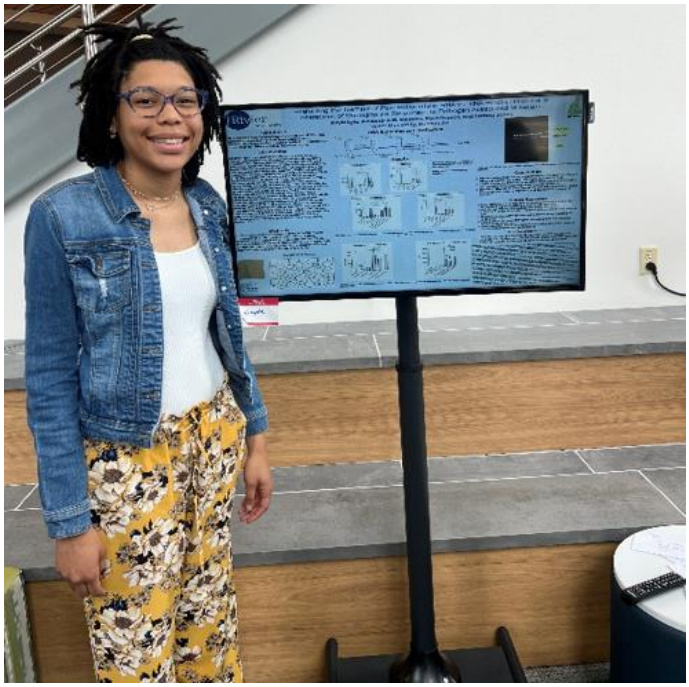
EVALUATING INVOLVEMENT OF PURE EXTRACELLULAR (ex)RNA vs. RNA BINDING PROTEINS IN ALTERATIONS IN MACROPHAGES RESPONSES TO PATHOGEN ASSOCIATED MOLECULES

Kayla Lyle[*], Alexandria M. Williams[†], and Tatiana Jones[‡]
Department of Biology, Division of Arts and Sciences, Rivier University
and
Pavel Ivanov[††]
Division of Rheumatology, Immunology and Allergy, Harvard University School of Medicine, Boston, MA



Kayla Lyle, a senior Biology major, graduating in December 2024, presents at the Transformative Learning Conference (TLC) in April 2024.

"… I find myself immersed in the boundless wonders of scientific inquiry. Even though I haven't nailed down my exact interest yet, I feel most at home in the lab. There's something magical about watching each experiment peel back another layer of the biological world. I'm all about paying close attention to detail and getting pumped about discovering new things in the lab.

As I stand on the brink of a new chapter, my future plans remain dynamic. While the path ahead may be uncertain, I embrace the journey with open arms, confident that each stride forward will unveil new opportunities for growth and exploration. When I'm not in the lab, you can find me cozying up with a knitting project or diving into a good book. Whether I'm stitching away or lost in the pages, these hobbies feed my curiosity and desire for new adventures."

Abstract

An expanding array of information highlights the role of extracellular ribonucleic acid (exRNA) in regulating inflammatory responses, possibly leading to autoimmunity or chronic inflammation. During inflammatory responses, RNA can be released from damaged during inflammation cells (self-exRNA), or from pathogens (non-self-exRNA). Regardless of the source, exRNA can be released as pure nucleic acid or in combination with RNA-associated proteins, also referred to as RNA-binding proteins (RBPs). Using RAW264.7 bone-marrow-derived macrophages cultured in the presence of 20 ng/mL of Granulocyte-Macrophage Colony Stimulating Factor (GM-

ISSN 1559-9388 (online version), ISSN 1559-9396 (CD-ROM version).

CSF), we previously showed that RNA extracted from macrophages (self-exRNA) downregulates macrophages' production of Tumor Necrosis Factor (TNF)α and Interleukin (IL)6 in response to Lipopolysaccharide (LPS), toll-like receptor (TLR)4 ligand. Thus, the question we have been trying to answer is the role of pure RNA vs. RBPs in the downregulation of macrophage inflammatory responses. To answer this question, we showed that RNA extracted from macrophages is comprised of ribosomal and transfer RNA, which both involve RBPs in their structure. After treating RNA extracted from macrophages with various RNases, with or without the addition of Proteinase K, we observed a gradual decrease in detectable RNA levels. Moreover, a further decrease was noted with the addition of Proteinase K. Subsequently, when the RNases-treated self-exRNA was added to LPS-activated macrophages, we observed a restoration of TNFα production but not IL6 production. This raises the question of whether there is a differential regulation of TNFα and IL6 production by self-exRNA.

Research was supported by NH-INBRE One-Year Pilot Project Grant P20GM103506. ■

---

* **KAYLA LYLE** '24, a senior Biology major at Rivier University, graduating in December 2024, was a presenter at the Transformative Learning Conference (TLC) in April 2024.

† **ALEXANDRIA M. WILLIAMS** '24, a senior Biology major at Rivier University, graduating in May 2024, was a presenter at the Transformative Learning Conference (TLC) in April 2024.

‡ **TATIANA JONES, M.D., Ph.D.**, Assistant Professor of Biology, obtained her M.D. at First Pavlov's State Medical Institute; she completed her clinical postdoctoral training in rheumatology and obtained her Ph.D. in clinical immunology at Military Medical Academy in Leningrad (St. Petersburg), Russia. She has 17 years of experience working as a practicing physician and rheumatologist. After moving to the United States in 2000, Dr. Jones worked at Harvard Medical School performing research in immunology. She joined Rivier University in 2020. Since 2021, she has been a Principal Investigator on NH-INBRE grants. Her research interest is the influence of extracellular (ex)RNA on the inflammatory profile of macrophages.

†† **Dr. PAVEL (PASHA) IVANOV** is an Associate Professor of Medicine at Harvard Medical School. Dr. Ivanov was trained as a chemist specializing in nucleic acid chemistry at Moscow State University where he completed his Ph.D. During postdoctoral training, Dr. Ivanov moved to EMBL (Heidelberg, Germany) to study human nonsense-mediated mRNA decay in the Molecular Medicine Partnership Unit, EMBL. His second postdoctoral training was at Harvard Medical School, where he studied mammalian RNA granules and stress-responsive non-coding RNAs. Dr. Ivanov is an Associate Professor of Medicine at BWH and HMS, an Associate Member of the Broad Institute of Harvard and M.I.T., and an Associate Member of the Harvard Institute of RNA Medicine.