EXTRACELLULAR (ex)RNA INFLUENCES THE PROINFLAMMATORY PROFILE OF MACROPHAGES STIMULATED WITH TLR2 AND TLR4 AGONISTS

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Alexandria M. Williams, a senior Biology major, graduating in May 2024, was a presenter at the Transformative Learning Conference (TLC) in April 2024.

"… I have always had a strong interest in science, yet I also love many other things, such as dancing, coffee, and traveling. I am graduating in May of 2024 and during my academic journey at Rivier University, I was determined to make the most of the opportunities available on campus. I have been fortunate to work with Dr. Tatiana Jones as a biomedical research assistant for two years.

This experience not only allowed me to acquire a wealth of knowledge but also helped me develop invaluable social and personal skills. I have had the opportunity to present a poster at the Annual NH-INBRE Conference in 2022 and 2023 and was awarded first place for poster presentation in 2023. As I delved deeper into the realm of research, I discovered a true passion for research, with a goal of becoming a pediatric oncologist.

In the next step of my journey, I have been selected to participate in The Summer Undergraduate Research Fellowship at the Geisel School of Medicine at Dartmouth. This incredible opportunity will enable me to further expand my understanding of research, bringing me one step closer to my goal of becoming a physician-scientist. I am excited about the journey ahead and the chance to make a meaningful contribution to the field of medicine."

Abstract

The proinflammatory profile of activated macrophages plays a crucial role in the outcome of inflammation. In our previous work, we have demonstrated that proinflammatory responses of cultured RAW264.7 macrophages activated through toll-like receptor (TLR)4 or TLR2/6 could be altered in a dose-dependent manner by extracellular (ex)RNA that derives from macrophages, (self)-exRNA. The addition of 5 μg/mL of self-exRNA to macrophages, activated by either TLR4 agonist lipopolysaccharide (LPS) or by TLR2/6 agonist Pam2SC4

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

resulted in reduced production of tumor necrosis factor (TNF)α and interleukin (IL)6, and in downregulation of major histocompatibility complex (MHC)II expression, without affecting nuclear factor kappa B (NFkB)1, suggesting alterations in proinflammatory responses. Yet, adding 5 μg/mL of exRNA extracted from bacteria to macrophages activated by either LPS or Pam2SC4 did not influence the production of TNFα and IL6 or the expression of MHCII. We also observed an increased production of TNFα and IL6 by macrophages co-cultured with mouse lung epithelial cells in response to bacterial exRNA; however, we found a reduced expression of MHCII and Interferon α and β Receptor (INFAR)1. Those results have prompted us to verify the purity of extracted RNA using the OD260/OD280 ratio and conduct a dose-response experiment using a broader range of exRNA doses. After establishing an adequate dose for self- and non-self exRNA and confirming RNA molecule's involvement in macrophages' profile alterations by pre-treating extracted RNA with RNases, we will develop an *in vitro* model co-culturing macrophages with lung epithelial cells to continue evaluation of self-exRNA influence on intracellular interactions.

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

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†† **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.