Abstract Information

First Name :	Amalia N
Last Name :	Awala
Email :	amalia.awala@uct.ac.za
Address :	University of Cape Town, South Africa
Participation :	symposium
Category :	Student
Thematic Area :	Neuroimmunology, Neuroinflammation, Neuroinfection
Title :	Silent sentinels: microglial (in)action during cryptococcal infection.
Co-Authors :	Awala AN1,2,3,4, Higgitt ER2,3,4, de Lange A1,2,3,4, Kauchali M2,3,4, Pato Y2,3,4, Raimondo JV1,2, Dangarembizi R2,3,4. 1Division of Cell Biology, Department of Human Biology, Faculty of Health Sciences; 2Neuroscience Institute, Faculty of Health Sciences, University of Cape Town, Groote Schuur Hospital; 3Division of Physiological Sciences, Department of Human Biology, Faculty of Health Sciences, 4CMM AFRICA Medical Mycology Research Unit, Institute of Infectious Disease and Molecular Medicine, Faculty of Health Sciences, 1,2,3,4 University of Cape Town, South Africa

Abstract :

Background: Cryptococcal meningitis (CM) is a fatal fungal infection of the brain and the leading cause of HIV-related mortality in the world. Brain injury in CM is thought to be of an inflammatory origin however substantial gaps exist in the neuropathogenesis of the disease. Particularly unknown is how microglia, the resident immune cells of the brain, respond to brain invasion by Cryptococcus neoformans, the causative fungus for CM. We therefore employed rodent, and human brain-based experimental models to characterize the interaction between microglia and C. neoformans in vitro.

Methods: We stimulated cultured rodent and human organotypic brain slices (OBSs) with a GFP/mCherry expressing reporter H99 strain of C. neoformans and characterized fungal-cell interactions using cell type-specific immunofluorescent markers for microglia (IBA1). Inflammatory activation of microglial cells within C. neoformans-treated OBS was measured by dual immunofluorescence staining with the inflammatory transcription factor nuclear factor for interleukin 6 (NF-IL6), a robust marker for tracking inflammation. We also performed single nuclei RNA-sequencing to determine inflammatory responses at the transcriptomic level. C. neoformans-treated slices were compared to untreated slices and slices treated with a known immunogen, lipopolysaccharide (LPS). To investigate potential neuroimmune modulation, we co-stimulated C. neoformans-treated slices with LPS. Inflammation was further confirmed by measuring proinflammatory cytokine release using a multiplex assay.

Results: C. neoformans was internalized by microglia but infected microglia were paradoxically inactive. There was minimal NF-IL6 activation and proinflammatory cytokine release in C. neoformans-treated slices and controls as was observed in LPS-treated slices. The slices co-stimulated with LPS and C. neoformans showed a significant reduction in NF-IL6 and cytokines suggesting that C. neoformans may possess immunomodulatory properties.

Conclusions

We conclude that C. neoformans engages in a unique immunomodulatory interaction with microglial cells, being internalized however failing to induce a robust proinflammatory response.