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Introduction

Our immune system is a highly versatile and dynamic defense system evolved to protect animals from the invading pathogens and cancer. The immune system provides human body a high level of protection from invading pathogens, in a robust, self-organized and distributed manner. The immune system specifically discriminates between foreign molecules and the body's own cells and proteins. Immunity has both a less specific and more specific component. The less specific component - innate immunity provides the first line of defense against infection. Innate immunity employs disease-resistance mechanisms that are not specific to a particular pathogen and recognize classes of molecules peculiar to frequently encountered pathogens. Contrary to innate immunity, the specific component - adaptive immunity acts in response to foreign antigens with a high degree of antigenic specificity, diversity and immunologic memory.

Humans routinely survive attacks from millions of pathogens and damage-causing agents by harnessing the power of our innate immune system. Innate immunity employs germline-encoded pattern recognition receptors (PRRs) for recognition of "non-self" from "self". Innate immune signaling empowers effective immune response against specific damage- and pathogen associated molecular patterns (DAMPs/PAMPs) with the action of highly conserved innate immune PRRs, including Toll-like receptors, RIG-I-like receptors, NOD-like receptors, and C-type lectin receptors. PPRs sense danger signals, such as PAMPs and DAMPs in form of conserved molecular signatures to trigger immune responses (Figure 1.1). Activation of PRRs leads to the activation of major inflammatory and innate immune-associated pathways, and subsequent release of inflammatory cytokines and chemokines [Harton, Linhoff *et al.*, 2002]. Innate immune signaling directs host responses to damage and infection by functional coordination of immune-regulatory and tissue repair mechanisms. Innate immunity plays a central role in the pathogenesis of several human infectious and inflammation associated diseases.

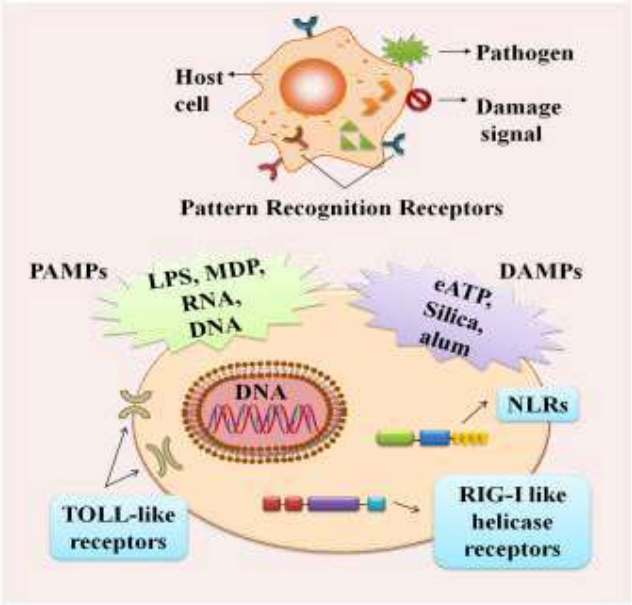

Figure 1.1 : *Innate immune sensing is mediated via germline-encoded pattern recognition receptors. Innate immunity forms the first line of host defense mechanisms in response to endogenous or exogenous foreign stimuli. Innate immune signaling empowers effective immune response against specific damage- and pathogen associated molecular patterns (DAMPs/PAMPs) with the action of highly conserved innate immune pattern recognition receptors (PRRs), that include Toll-like receptors, RIG-I-like receptors, NOD-like receptors, and C-type lectin receptors. Activation of PRRs leads to a signaling cascade of innate immune responses involving major inflammation-associated pathway activation, directing neighboring immune cells to the target site and other immunological responses controlling cellular damage and repair mechanisms.*

1.1 PURPOSE OF THE STUDY

NLRs (nucleotide-binding domain, leucine-rich repeat containing receptors) are evolutionarily conserved cytoplasmic pattern recognition receptors that recognize PAMPs and DAMPs to regulate inflammation and immunity.. Dysregulated NLR function is associated with several diseases including cancer, metabolic diseases, autoimmune disorders and autoinflammatory syndromes [Jha and Ting, 2009]. NLRs perform important pro-inflammatory and anti-inflammatory roles in cancer, however the role of NLRs in glioma remain unknown [Zhu and Cao, 2017]. The aim of my dissertation was 1) to elucidate if amorphous nanosilica act as DAMPs, to induce inflammation and NLR-mediated innate immune signaling leading to subsequent cytotoxic responses, and 2) to characterize NLR gene expression profiles and test our hypothesis that NLR regulated inflammation could play important role in molecular stratification of low grade glioma and glioblastoma.

1.2 BRIEF RESULTS, SCOPE AND FUTURE PROSPECTS OF THE WORK

This study is aimed to investigate the role of NLRs in inflammation and cell death-associated mechanisms in response to amorphous nanosilica particles. Previous studies have shown how crystalline silica, acts as DAMP to induce NLRP3 inflammasome activation, and subsequent inflammation and cytotoxic responses both *in vitro* and *in vivo* [Cassel, Eisenbarth *et al.*, 2008; Kuroda, Ishii *et al.*, 2011]. However, studies demonstrating the inflammatory and cytotoxic effects of amorphous nanosilica have been scarce. Our study shows cells exposed to amorphous nanosilica, undergo specific morphological and cytotoxic

changes with increasing particle dose and exposure time. Interestingly, Apoptosis – associated speck-like protein containing a caspase recruitment domain (ASC/PYCARD), mediates amorphous nanosilica induced inflammation and cell death. Nanosilica induced cytotoxicity and cell death mechanisms were specific for each particle size and cell type. Above findings illustrate toxic effects of different-sized amorphous nanosilica in human lung cell population.

The study also aims to characterize the expression profile of NLRs and NLR-associated genes in low grade glioma (LGG) and glioblastoma (GBM), to determine the relative contribution of NLR family members in glioma pathology. Current literature shows critical role of NLRs in carcinogenesis and anti-cancer immune responses [Zitvogel, Kepp *et al.*, 2012; Janowski, Kolb *et al.*, 2015]. The newly emerging dual tumor-promoting and -inhibitory roles of NLRs have been observed in inflammation-induced colorectal cancer [Allen, TeKippe *et al.*, 2010; Hu, Elinav *et al.*, 2010]. The link between NLRs and glioma pathology has not been identified so far, and this prompted us to investigate the regulation of NLRs in different grades of glioma. We found differential expression and methylation levels of NLRs and NLR-associated genes in GBM. The differential regulation of NLRs also strongly correlated with the glioma patient survival. NLRP12 specifically emerged as innate immune gene with strong prognostic value for GBM.

The preliminary results show differential regulation of NLRs in glioma, promoting future research investigations determining the functional significance of NLRs related to cancer cell migration, proliferation and metastasis in LGG and GBM. Overall, the purpose of the study was to provide a basic understanding of NLR regulation in inflammation, cell death and glioma, for future development of novel therapeutic approaches targeted at NLR-associated pathways for selective inhibition of tumor promoting mechanisms in different grades of glioma.