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The Alarmin Properties of DNA and DNA-Associated Nuclear Proteins

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ABSTRACT

Purpose: The communication of cell injury and death is a critical element in host defense. Although immune cells can serve this function by elaborating cytokines and chemokines, somatic cells can repurpose nuclear macromolecules to function as damage-associated molecular patterns (DAMPs) or alarmins to exert similar activity. Among these molecules, DNA, high-mobility group box-1, and histone proteins can all act as DAMPs once they are in an extracellular location. This review describes current information on the role of the nuclear DAMPs, their translocation to the outside of cells, and pathways of activation after uptake into the inside of immune cells.

Methods: MEDLINE and PubMed databases were searched for citations (1990–2016) in English related to the following terms: DAMPs, high-mobility group box-1, DNA, histones, cell death, danger, and immune activation. Selected articles with the most relevant studies were included for a more detailed consideration.

Findings: Although nuclear molecules have important structural and genetic regulatory roles inside the cell nucleus, when released into the extracellular space during cell death, these molecules can acquire immune activity and serve as alarmins or DAMPs. Although apoptosis is generally considered the source of extracellular nuclear material, other cell death pathways such as necroptosis, NETosis, and pyroptosis can contribute to the release of nuclear molecules. Importantly, the release of nuclear DAMPs occurs with both soluble and particulate forms of these molecules. The activity of nuclear molecules may depend on post-translational modifications, redox changes, and the binding of other molecules. Once in an extracellular location, nuclear DAMPs can engage the same pattern recognition receptors as do pathogen-associated molecular patterns. These interactions can activate immune cells and lead to cytokine and chemokine production. Among these receptors, internal receptors for DNA are key to the response to this molecule; the likely function of these internal sensors is the recognition of DNA from intracellular infection by bacteria or viruses. Activation of these receptors requires translocation of extracellular DNA into specialized compartments. In addition to nuclear DNA, mitochondrial DNA can also serve as a DAMP.

Implications: The communication of cell injury and death is a critical element in host defense and involves the repurposing of nuclear molecules as immune triggers. As such, the presence of extracellular nuclear material can serve as novel biomarkers for conditions involving cell injury and death. Targeting of these molecules may also represent an important new approach to therapy. (*Clin Ther*. 2016;▮:▮▮▮–▮▮▮) Published by Elsevier HS Journals, Inc.

Key words: cell death, DNA, histones, high-mobility group box-1, inflammation.

INTRODUCTION

The mammalian organism is under constant threat from the environment and prone to harm from danger.[1] Danger represents challenges (physical, chemical, or infectious) that can injure or kill cells and lead to tissue loss and organ dysfunction. Because every cell in the body is subject to danger, the organism requires a universal alarm system to combat these hazards. Immune cells, which can elaborate cytokines and chemokines, can orchestrate

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the response, but somatic cells are key to engaging the immune system and communicating the presence of danger. Alarmins, released by both somatic and immune cells, are the basis of this communication.[2,3] As such, this concept enlarges the immune system to encompass essentially all cells in the body.

The communication of danger by somatic cells involves "repurposing" of cellular constituents, utilizing endogenous molecules that can be repurposed as immune signals. Such molecules can display dual function: their ordinary physiologic function and the function as alarmins. Thus, upon release from cells during infection and injury, endogenous molecules can acquire immunologic activity and transition from their ordinary function to danger signals or alarmins. Nuclear molecules, including DNA, histones, and nonhistone proteins such as high-mobility group box-1 (HMGB1), can all be repurposed as immunologic mediators, a striking finding in view of their essential physiologic role.

By analogy with the exogenous pathogen-associated molecular patterns (PAMPs), the endogenous molecules with similar immunologic roles are called damage-associated molecular patterns (DAMPs).[4] The term alarmin can be used for those DAMPs that can induce chemotaxis and promote the induction of immune responses. Characterizing a molecule as a DAMP or an alarmin, however, may be a matter of the survey of immune activity rather than a fundamental difference in biologic activity. In the present article, the terms alarmin and DAMP are used interchangeably for the nuclear macromolecules of this class. To find information on these issues, MEDLINE and PubMed databases were searched for citations (1990–2016) in English related to the following terms: DAMPs, alarmin, HMGB1, DNA, histones, cell death, danger, and immune activation. Selected articles with the most relevant studies are included for a more detailed consideration.

THE GENERATION OF EXTRACELLULAR NUCLEAR DAMPS

For DAMPs to induce responses, they need to first leave their ordinary intracellular location to provide a source of extracellular DAMPs.[4,5] In some instances, a DAMP can act directly to stimulate responses in an unmodified form, although some DAMPs acquire immunologic activity after posttranslational modification, degradation, or binding to another molecule to form a complex.[6,7] DAMPs can trigger the same receptor systems as PAMPs. These receptor systems are called pattern recognition receptors (PRRs) and may be located on the inside or outside of cells. The Toll-like receptors (TLRs) are the classic PRRs.[8,9]

Because some PRRs are intracellular, the action of a DAMP may require translocation to the cell interior. This uptake may reflect the high extracellular concentration of the DAMP or phagocytosis of the DAMP in the context of cell debris. In some instances, the action of the DAMP may require a binding molecule to facilitate uptake. The nature of binding molecules likely varies depending on the DAMP and the clinical setting. LL37 is an example of a molecule that binds to DNA to mediate its activity.[10]

The role of nuclear molecules in stimulating immune responses has attracted great interest because, for many years, these molecules were considered immunologically inert, with the induction of responses exclusive to autoimmunity, especially systemic lupus erythematosus (SLE).[11] This disease is characterized by the expression of antibodies to nuclear molecules (antinuclear antibodies). Because antinuclear antibodies occur primarily in lupus and related diseases, the immune activity of nuclear molecules seemed essentially unique to autoimmunity. The recognition that nuclear molecules can act as immune mediators is thus a radical change in thinking.

The nuclear DAMPs can enter the extracellular space as a consequence of cell death or activation. Although these 2 settings may appear different, in the immune system at least, activation can lead to cell death. Such activation-induced cell death may be a strategy to curtail excessive cellular proliferation in an antigen-specific response.[12] In the case of immune cells activated by intracellular infection, death may terminate the infection by depriving the bacterium or virus of a site for replication. Because cytokine induction can occur concomitantly with death, the outcome of intracellular infection can be the amplification of a response and recruitment of other cell populations to assure the eradication of infection.[13,14]

The distinction between activation and death in vitro may be operational and depend on the model system studied, the analytes measured, and the treatment. As shown in studies on the responses of murine