

Editorial

Understanding Hydrogen Sulfide in Inflammation: Opportunities and Challenges

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Abstract

Inflammation is an adaptive response to injury, but uncontrolled inflammation can lead to tissue damage and disease. Research in our laboratory (since confirmed in different laboratories worldwide) has shown that hydrogen sulfide (H₂S) acts as a mediator of inflammation in different disease conditions. Learning about a novel mediator of inflammation results in unique opportunities with which to approach inflammatory diseases. At the same time, the complexity of biological systems and translation of research from the bed to the bedside also presents challenges. This Editorial aims to discuss the opportunities and challenges in relation to the role of H₂S in inflammation, and the future prospects for this research.

Keywords: Hydrogen sulfide; inflammation; substance P; leukocytes.

Inflammation is a normal adaptive response to traumatic, infectious, post-ischemic, toxic or autoimmune injury. It is a highly orchestrated process characterized by redness, heat, pain, and swelling, and known to mankind for more than 2000 years. Uncontrolled inflammation, however, results in disease,

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and most of the diseases have been shown to have an inflammatory component. Inflammation is a challenging research problem in systems biology in the academia. Identifying biological molecules that mediate inflammation, and strategies that could control inflammation, therefore, can lead to novel therapeutic approaches for several diseases. In this scenario, inflammation also represents a multibillion dollar market for the pharmaceutical industry, in addition to being a key research question.

Hydrogen sulfide (H_2S), a toxic gas, has been known as an environmental and industrial pollutant for more than 300 years. In recent years, however, it has been shown to be produced in the mammalian (including human) body. We have shown that H_2S , synthesized by the activity of cystathionine- γ -lyase (CSE), acts as a mediator of inflammation in different disease conditions, including acute pancreatitis [1–3], sepsis [4–6], burn injuries [7] and joint inflammation [8]. In experimental models of disease, H_2S synthesis by CSE is increased resulting in aggravated inflammation. Blockage of the action of CSE – by pharmacological means using a selective inhibitor, gene silencing (by siRNA) and gene deletion (using gene knockout mice) results in protection against different inflammatory diseases. Depending on the disease model, evidence of such protection as a result of inhibition of H_2S synthesis includes attenuated increase in indicators of inflammation, protection against organ damage, and improvement in survival.

The discovery of H_2S as a novel mediator of inflammation led to studies aimed at trying to understand the mechanism by which it acts. In human monocyte cell line U937, treatment with H_2S results in significant increases in the production of the cytokines tumor necrosis factor (TNF)- α , interleukin (IL)- 1β , and IL-6. This effect is mediated via NF- κ B and extracellular signal related kinase (ERK) pathway [9]. Treatment of mouse macrophage RAW264.7 cells with lipopolysaccharide (LPS) results in increased production of proinflammatory cytokines and chemokines IL- 1β , IL-6, TNF- α , and monocyte chemoattractant protein (MCP)-1. Silencing of the CSE gene by small interference ribonucleic acid (siRNA) results in decreased levels of proinflammatory cytokines [10].

Recruitment of leukocytes (primarily neutrophils and monocytes/macrophages) at the site of inflammation is a key component of inflammation. Cytokines, chemokines, and adhesion molecules play a key role in leukocyte recruitment in inflammation. *In vivo* studies with models of inflammatory diseases, such as acute pancreatitis and sepsis, show that H_2S contributes to inflammation by promoting leukocyte recruitment through cytokines (IL- 1β ,

IL-6, TNF- α), chemokines (MCP-1, macrophage inflammatory protein (MIP)-1 α and MIP-2), and adhesion molecules (intercellular adhesion molecule (ICAM)-1, P-selectin, and E-selectin) [11, 12]. As observed in the *in vitro* studies described above, at the molecular level, the effect of H₂S was shown to be mediated *via* NF- κ B and the ERK pathway in experimental models of inflammatory disease. In these studies, leukocyte recruitment and the expression of cytokines, chemokines, and adhesion molecules was shown to be upregulated on disease induction. Pharmacological inhibition of CSE with D/L-propargylglycine, a selective inhibitor of the enzyme, resulted in protection against inflammation and reduction in leukocyte recruitment, and, in sepsis, administration of an H₂S donor further aggravated inflammation.

Neurogenic inflammation involves activation of transient receptor potential vanilloid type 1 (TRPV1) expressed by primary sensory neurons and release of substance P from these neurons. Substance P, acting via neurokinin (NK)-1 receptor, plays an important role in the pathogenesis of several inflammatory conditions, including acute pancreatitis, sepsis, burn injuries, and joint inflammation. Administration of H₂S to mice causes an increase in substance P levels, resulting in lung inflammation. A relationship between H₂S and substance P has been investigated in experimental acute pancreatitis and sepsis [13, 14]. Using a variety of different and complementary experimental approaches, including antagonism of TRPV1 and NK-1 receptor, gene deletion, and CSE inhibition, it has been shown that H₂S contributes to inflammation *via* TRPV1-mediated neurogenic inflammation involving substance P.

The liver is the richest source of CSE in the body. Liver sinusoidal endothelial cells (LSECs) line the low shear, sinusoidal capillary channels and are the most abundant non-parenchymal hepatic cell population. LSECs have unique vital physiological and immunological functions, including leukocyte recruitment. LSECs have been reported to influence the inflammatory disease process by mediating recruitment of leukocytes *via* chemokines and adhesion molecules. We have recently shown that sepsis is associated with the disruption of the LSECs and formation of gaps, which are large defects through the LSECs. Furthermore, mice genetically deficient in CSE are protected against this disruption of the LSECs following sepsis [6]. These results point to an important role of LSECs in the pro-inflammatory action of H₂S.

These studies show that H₂S contributes to inflammation *via* cytokines, chemokines, adhesion molecules (and leukocyte recruitment), and TRPV1 and substance P. Also, in sepsis, LSECs may play a significant role in the pro-inflammatory action of H₂S.

The studies, described above, have all been done in experimental animal models of disease or *in vitro* systems, and the evidence of clinical relevance of H₂S in inflammatory disease has only recently started emerging. In one study, H₂S levels were investigated in patients with arthritis. Rheumatoid arthritis (RA) and gout are both common forms of inflammatory arthritis. In one study, H₂S levels have been found to be elevated in the synovial fluids of patients with RA and gout. In RA, a clear correlation was also observed between disease activity and synovial fluid H₂S levels [15]. In another study, H₂S and substance P levels were determined in patients with sepsis. Results from this study showed that circulating levels of H₂S and substance P were significantly elevated in patients with sepsis [16]. The elevation in H₂S levels followed that in substance P levels, suggesting (as has been shown in animal models of disease) that H₂S acts upstream of substance P in the pathogenesis of inflammatory disease.

Based on the studies with experimental models of disease, which have been substantiated by recent clinical studies, we now have a good understanding of the contribution of H₂S to inflammation. Awareness of a new mediator of inflammation, which can be inhibited as a potential therapeutic approach for inflammatory disease, presents numerous opportunities. This is a new area of research with immense promise, both in terms of understanding the pathology of inflammation, and discovering new treatments for diseases which are major health problems.

At the same time, there are challenges that we must endeavor to overcome. Although significant progress has been made in understanding the mechanism by which H₂S acts, questions remain, particularly with regard to its interaction with substance P and LSECs. Furthermore, there is need to develop novel H₂S synthesis inhibitors, which are more selective and have a better safety profile than the ones currently available. Availability of such inhibitors will be a major step forward towards taking this knowledge to the clinic. And finally, now is the time to investigate the role of H₂S in inflammation in clinical disease, and build on the studies that have clearly been promising.

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References

- [1] Bhatia M, Wong FL, Fu D, Lau HY, Moochhala S, Moore PK. Role of hydrogen sulfide in acute pancreatitis and associated lung injury. *FASEB J*. 2005; 19: 623–625.
- [2] Ang AD, Rivers-Auty J, Hegde A, Ishii I, Bhatia M. The effect of CSE gene deletion in caerulein-induced acute pancreatitis in the mouse. *Am J Physiol Gastrointest Liver Physiol*. 2013; 305: G712–G721.
- [3] Badieli A, Chambers ST, Gaddam RR, Fraser R, Bhatia M. Cystathionine-gamma-lyase gene silencing with siRNA in monocytes/macrophages protects mice against acute pancreatitis. *Appl Microbiol Biotech*. 2016; 100: 337–346.
- [4] Zhang H, Zhi L, Moore PK, Bhatia M. The role of hydrogen sulfide in cecal ligation and puncture induced sepsis in the mouse. *Am J Physiol Lung Cell Mol Physiol*. 2006; 290: L1193–L1201.
- [5] Badieli A, Chambers ST, Gaddam RR, Bhatia M. Cystathionine-gamma-lyase gene silencing with siRNA in monocytes/macrophages attenuates inflammation in cecal ligation and puncture-induced sepsis in the mouse. *J Biosci*. 2016; 41: 87–95.
- [6] Gaddam RR, Fraser R, Badieli A, Chambers S, Cogger VC, Le Couteur DC, Ishii I, Bhatia M. Cystathionine-gamma-lyase gene deletion protects mice against inflammation and liver sieve injury following polymicrobial sepsis. *PLoS ONE*. 2016; 11: e0160521.
- [7] Zhang J, Sio SW, Moochhala S, Bhatia M. Role of hydrogen sulfide in severe burn injury-induced inflammation in the mouse. *Mol Med*. 2010; 16: 417–424.
- [8] Bhatia M, Sidhapuriwala J, Moochhala S, Moore PK. Hydrogen sulphide is a mediator of carrageenan-induced hindpaw oedema in the rat. *Br J Pharmacol*. 2005; 145:141–144.
- [9] Zhi L, Ang AD, Zhang H, Moore PK, Bhatia M. Hydrogen sulfide induces the synthesis of pro-inflammatory cytokines in human monocyte cell line U937 via ERK-NFκB pathway. *J Leuk Biol*. 2007; 81: 1322–1332.
- [10] Badieli A, Rivers-Auty J, Ang AD, Bhatia M. Inhibition of hydrogen sulfide production by gene silencing attenuates inflammatory activity of LPS-activated RAW 264.7 cells. *Appl Microbiol Biotech*. 2013; 97: 7845–7852.
- [11] Zhang H, Zhi L, Moochhala S, Moore PK, Bhatia M. Endogenous hydrogen sulfide regulates leukocyte trafficking in cecal ligation and puncture induced sepsis. *J Leuk Biol*. 2007; 82: 894–905.

- [12] Tamizhselvi R, Moore PK, Bhatia M. Inhibition of hydrogen sulfide synthesis attenuates chemokine production and protects mice against acute pancreatitis and associated lung injury. *Pancreas*. 2008; 36:e24-e31.
- [13] Bhatia M, Sidhapuriwala J, Ng SW, Tamizhselvi R, Moochhala S. Proinflammatory effects of hydrogen sulfide on substance P in caerulein-induced acute pancreatitis. *J Cell Mol Med*. 2008; 12: 580–590.
- [14] Ang SF, Moochhala S, Bhatia M. Hydrogen sulfide promotes transient receptor potential vanilloid 1-mediated neurogenic inflammation in polymicrobial sepsis. *Crit Care Med*. 2010; 38: 619–628.
- [15] Muniraj N, Stamp LK, Badiei A, Hegde A, Cameron V, Bhatia M. Hydrogen sulfide acts as a pro-inflammatory mediator in rheumatic disease. *Int J Rheum Dis*. 2017; 20: 182–189.
- [16] Gaddam RR, Chambers S, Murdoch D, Shaw G, Bhatia M. Circulating levels of hydrogen sulfide and substance P in patients with sepsis. *J Infect*. 2017; 75: 293–300.

Biography



Madhav Bhatia heads the Inflammation Research Group in the Department of Pathology and Biomedical Science at the University of Otago, Christchurch. Research in his laboratory has shown hydrogen sulfide and substance P as mediators of inflammation and potential therapeutic targets for inflammatory diseases such as acute pancreatitis, sepsis, burn injuries, and joint inflammation. He has received numerous grants, has authored more than 180 contributions to the peer-reviewed literature, given several invited presentations in different countries and is on Editorial Boards of 43 journals. He has mentored several postgraduate students and research staff, who are now independent investigators in their own right, leading research groups in different parts of the world. His publications have been cited more than 11000 times, and he has an “h”-index of 54.