Published in final edited form as:

Curr Opin Allergy Clin Immunol. 2020 February ; 20(1): 14–22. doi:10.1097/ACI.0000000000000595.

Differential Expression and Role of S100 Proteins in Chronic Rhinosinusitis.

Jorgen S. Sumsion^{a,*}, Abigail Pulsipher^{a,*}, Jeremiah A. Alt^a

^aDepartment of Surgery, Division of Otolaryngology–Head and Neck Surgery, University of Utah School of Medicine, Salt Lake City, Utah, USA

Abstract

Purpose of Review: Immune system modulators have been under investigation to help elucidate the underlying pathophysiologies of chronic rhinosinusitis (CRS). Psoriasin (S100A7) and calgranulins (S100A8, S100A9, and S100A12) are S100 proteins that have been studied for their immune-mediating responses to pathogens within the context of CRS. This review highlights the expression patterns and proposed roles of S100 proteins in CRS with (CRSwNP) and without (CRSSNP) nasal polyps.

Recent Findings: Elevated levels of S100A7 and S100A12 were measured in the sinonasal tissues of patients with CRSSNP compared to CRSwNP and controls. S100A12 expression in CRSSNP was significantly correlated to disease severity. Contrastingly, increased S100A8, S100A9, and S100A8/A9 levels were demonstrated in the nasal polyp tissues of patients with CRSwNP compared to those in inferior turbinate and uncinate tissues of patients with CRSSNP and controls.

Summary: The reported differential expression patterns and activities of psoriasin and calgranulins suggest that S100 proteins exert unique and concerted roles in mediating immunity in different subtypes of CRS. These studies will enable further investigations focused on understanding the immune-modulating mechanisms of S100 proteins in different inflammatory signaling pathways and disease phenotypes of CRS towards the pursuit of identifying new biomarkers and targets for improved outcomes.

Keywords

Chronic Rhinosinusitis; Psoriasin; Calgranulins; S100 proteins; Innate and Adaptive Immunity

Correspondence to Jeremiah A. Alt, MD, PhD, FACS, University of Utah School of Medicine, Division of Otolaryngology – Head and Neck Surgery, 50 North Medical Drive, Room 3C120, Salt Lake City, UT 84132, USA. Tel: (801) 585-7143; Fax: (801) 585-5744; jeremiah.alt@hsc.utah.edu.

*Equal authorship contribution

CONFLICTS OF INTEREST Jeremiah A. Alt is a consultant for Medtronic, Inc. (Jacksonville, FL). Jeremiah A. Alt and Abigail Pulsipher are affiliated with GlycoMira Therapeutics, Inc. (Salt Lake City, UT). None of these companies are affiliated with this review.

INTRODUCTION

Chronic Rhinosinusitis (CRS) is an inflammatory condition of the nose and paranasal sinuses symptomatically characterized by nasal obstruction, mucopurulent nasal drainage, facial pain, and decreased or loss of sense of smell.[1] Although an etiologically and pathologically heterogeneous disease, studies have suggested that maladaptive innate immune responses with associated epithelial barrier breakdown may be an essential etiology of the disease. CRS is broadly classified into two phenotypes: CRS with nasal polyps (CRSwNP) and CRS without nasal polyps (CRSSNP)[2]. The observed inflammatory mechanisms in CRSwNP are commonly driven by allergic and eosinophilic T-helper cell type 2 (Th2)-mediated responses, characterized by CD4+ lymphocyte activation and the secretion of pro-inflammatory Th2-associated cytokines[3–6]. Th2 inflammation can also be non-allergic, induced by pollutants or pathogens that initiate innate immune responses, whereas CRSSNP pathology is predominated by neutrophilic Th1-associated cytokine-mediated signaling pathways and CD8+ lymphocyte activation. Increasing investigations, therefore, have focused on elucidating the biological roles of innate immune modulators in CRS to better understand the underlying pathologies caused by these complex inflammatory profiles.

Pro-inflammatory innate immune responses in CRS are triggered by interactions of pattern recognition receptors (PRRs) with their ligands, pathogen- and damage-associated molecular patterns (PAMPs and DAMPs, respectively). PRRs, such as the Toll-like receptors (TLRs) [7–10] and the receptor for advanced glycation end products (RAGE)[11], serve critical roles in maintaining airway homeostasis and in early inflammatory signaling. These functions are initiated by PRR response to endogenous molecules (alarmins) released by effector cells upon tissue damage, as well as to exogenous PAMPs derived from pathogens during infection and DAMPs secreted by activated immune and necrotic cells[12]. PRR-ligand interactions stimulate the production and release of pro-inflammatory cytokines, chemokines, and DAMPs, promoting the chemotaxis and activation of innate immune antigen-presenting cells and initiating adaptive immune and tissue repair processes. Abnormal PRR-ligand-mediated signaling can lead to maladaptive immune responses that may contribute to the pathogenesis of disease[12]. Numerous immune modulating DAMPs, therefore, have been studied for their roles in activating these pathways within the context of CRS, including psoriasin and calgranulins.

Psoriasin and calgranulins are calcium-binding S100 proteins that are involved in innate and adaptive immunity. Psoriasin (S100A7), secreted by primary keratinocytes[13,14], is present in the brain during infection[15] and contributes to tissue barrier defense mechanisms[16–18]. Although psoriasin was originally identified as an immune system modulator in the inflammatory condition psoriasis,[13] S100A7 has been implicated in breast cancer[19] and epithelial squamous cell carcinomas of the skin and bladder[20,21]. Calgranulins A, B, and C (S100A8, S100A9, and S100A12, respectively) are secreted by granulocytes, monocytes, and necrotic cells[22–25] and have been identified as emerging clinical biomarkers of inflammation[22,26–29]. Calgranulins have also demonstrated immune system modulating-properties in rheumatoid and juvenile idiopathic arthritis, inflammatory bowel disease, and cystic fibrosis[30,31]. In addition to their independent functions, S100A8 and S100A9 have

been reported to dimerize, forming calprotectin, which has unique activity in different biological contexts[32]. Psoriasin and calgranulins elicit pro-inflammatory and proliferative responses through activating RAGE[14,15,33–35] and TLR4[22,36–38] in numerous inflammatory conditions, including chronic asthma[39], and also exert chemotactic properties. S100A7 is a potent chemoattractant for CD4+ lymphocytes and neutrophils,[40] whereas S100A8, S100A9, and S100A8/A9 attract neutrophils[41,42], and S100A12 leads to the influx of monocytes and mast cells[28,43–45].

Within the last decade, investigations of psoriasin and calgranulins investigations have been extended to CRS, due to increasing reports underscoring the importance of S100 proteins as clinical markers of inflammation and in innate immunity. This review highlights reports that have characterized the gene and protein expression of psoriasin, calgranulins, and calprotectin in serum, nasal lavage fluid (NLF), and different sinonasal tissue regions and subsites with respect to CRS subtypes[46–52]. The proposed roles of S100 proteins in epithelial barrier function and innate immune-mediating response to pathogens will additionally be discussed.

DIFFERENTIAL EXPRESSION OF PSORIASIN AND CALGRANULINS IN SINONASAL TISSUES AND SUBTYPES OF CRS

The differential expression patterns of psoriasin and calgranulins are summarized in Table 1. Psoriasin is expressed in the epithelium and seromucous glands of inferior turbinate[50] and uncinate[47] tissues in healthy controls, with increased S100A7 signal observed in the inferior turbinate. Tieu *et al.* also detected S100A7 expression in the epithelium, glands, and subepithelial stroma of uncinate tissues in patients with CRSsNP and CRSwNP, although the S100A7 signal was decreased in both groups compared to controls.[47] Another study reported the downregulation of S100A7 mRNA levels in inferior turbinate and uncinate tissue homogenates in both CRS phenotypes when compared to healthy controls.[51] Protein levels of psoriasin were also found to be significantly lower in the NLF in both CRS subtypes when compared to controls[47]. A separate report corroborated this result, showing decreased levels of S100A7 in NLF obtained from patients with allergic rhinosinusitis in comparison to controls.[46] Similarly, serum psoriasin expression was shown to decrease in both CRSwNP and CRSsNP compared to controls, although not significantly.[47]

The expression patterns of calgranulins A and B and calprotectin have also been investigated in CRS with respect to sinonasal tissue location of sampling and subsite. Van Crombruggen *et al.* demonstrated the negligible expression of S100A8, S100A9, and S100A8/A9 in the inferior turbinate tissues of controls and increased expression of these proteins in the basement membrane, blood vessels, and extracellular matrix (ECM) of inferior turbinate and nasal polyp tissues of patients with CRSwNP.[52] This study additionally measured no changes in the gene expression of S100A8 and S100A9 in the nasal polyp and inferior turbinate tissues of CRSwNP compared to controls.[52] In contrast, Richer *et al.* reported decreased levels of S100A8 and S100A9 mRNA in the inferior turbinate and uncinate tissue homogenates of patients with CRSwNP compared to controls.[51] Moreover, Tieu *et al.* demonstrated that S100A8/A9 expression was significantly increased in nasal polyp tissue

compared to uncinate and inferior turbinate tissues of controls and of patients with CRSwNP and CRSsNP. This study additionally measured significant reductions in the levels of calprotectin in NLF and serum obtained from patients with CRSwNP in comparison to CRSsNP and controls. These results suggest that sinonasal tissue location and subsite, as well as underlying pathology associated with phenotype, are important when considering the potential biological implications of calgranulins and calprotectin in CRS.

Pulsipher *et al.* recently characterized the expression patterns of S100A12 in CRS[49]. In this study, the protein levels of S100A12 were found to be significantly elevated in the anterior ethmoid homogenates of patients with CRSsNP compared to CRSwNP and controls. S100A12 expression levels were not statistically different between patients with CRSwNP and controls. The authors further measured the expression of human neutrophil elastase (HNE) in anterior ethmoid tissues, reporting a significant increase in HNE expression in patients with CRSsNP compared controls and CRSwNP. Although not significant, a positive correlation between S100A12 and HNE expression in patients with CRSsNP was reported, whereas a significantly negative correlation between these two proteins was observed in patients with CRSwNP. In addition to this study, Van Crombruggen *et al.* examined the correlations between S100 protein expression and CRS-specific disease severity, reporting no significant correlations between clinical parameters of CRS and levels of S100A8, S100A9, and S100A8/A9. In contrast, Pulsipher *et al.* showed that disease severity (computed tomography scores) in patients with CRSsNP was significantly correlated to S100A12 tissue expression. Further characterization is required for determining whether S100A12 may be considered a biomarker of inflammation associated with Th1-biased signaling in CRSsNP.

Taken together, the expression patterns of psoriasin and calgranulins vary according to disease subtype, location of tissues sampling, and tissue subsite, suggesting that the manifestation of inflammation and signaling pathways also vary in different regions of the sinuses. Furthermore, most studies compare biomarker results measured in nasal polyp tissues from patients with CRSwNP to those from uncinate or inferior turbinate tissues of controls and patients with CRSsNP, which has a different tissue structure. While no nasal polyp analog exists in control patients, biomarker detection in similar tissue sampling regions should be conducted for intra-patient comparisons to fully characterize the pathophysiology of the nasal mucosa.

S100 PROTEINS AND IMMUNITY

Regarding S100A7 and innate immunity, Tieu *et al.* reported the differential expression of psoriasin in subtypes of CRS, which may be resultant from the differing etiologies contributing to the disease states observed in CRSwNP and CRSsNP. As psoriasin has been shown to exert antimicrobial effects by permeabilizing bacterial cell membranes in skin disorders,[16,18] it may also exhibit similar innate immune responses against invading pathogens in CRS.[53] Psoriasin production is known to be repressed by Th2-associated cytokine interleukin 4 (IL-4),[50] which supports the upregulation of S100A7 in CRSsNP compared to CRSwNP pathologies. Moreover, psoriasin is a potent chemoattractant for CD4⁺ lymphocytes and neutrophils,[40] suggesting that S100A7 may be helping to

modulate a positive feedback mechanism that maintains immune system homeostasis in CRSsNP.

Increasing evidence proposes the involvement of PRR-ligand-mediated interactions in the initiation of inflammatory signaling in CRS. PRRs, such as RAGE, serve as immune rheostats in stimulating immune responses at the mucosal-air interface in CRS.[8–10] With the exception of the upper and lower respiratory tract, RAGE expression is undetectable in most cell types and tissues under normal physiology.[11] Several investigations have implicated RAGE and calgranulins in CRS, hypothesizing that multiple mechanisms may be activated to influence CRS pathology. One possible mechanism is direct ligation with membrane-bound RAGE (mRAGE). Psoriasin is a known ligand of RAGE[35], and S100A12 was one of the first discovered ligands of RAGE, termed EN-RAGE.[38] RAGE ligation by Psoriasin and S100A12 can lead to pro-inflammatory responses through the activation of mitogen-activated protein kinases (MAPK) and nuclear factor kappa-light-chain-enhancer of activated B cells (NF- κ B), leading to the accumulation of reactive oxygen species (ROS) and cell death.[11,35,54,55] Activation of RAGE leads to increased secretion of tumor necrosis factor alpha (TNF α) and interleukin 1 beta (IL-1 β) by monocytes and endothelial cells[38], resulting in the activation of Th1-mediated responses.[43] Thus, the increased levels of Psoriasin and S100A12 observed in the sinonasal mucosa of patients with CRSsNP compared to CRSwNP and controls, suggest potential roles as key pro-inflammatory ligands that drive the inflammatory milieu seen in Th1-driven profiles in CRS[56,57].

Another mechanism for RAGE-mediated activation in CRS considers the interaction of S100A12 with the soluble forms of RAGE, collectively known as sRAGE. sRAGE lacks a transmembrane and cytoplasmic domain and has two isoforms: endogenous (1) secretory RAGE (esRAGE), derived from alternative mRNA splicing and (2) cleaved RAGE (cRAGE), derived from the proteolytic cleavage of mRAGE by metalloproteinases.[11,58] sRAGE is largely considered an anti-inflammatory decoy receptor for mRAGE ligands, but has also been associated with inflammatory signaling pathways.[59] Previous studies by Van Crombruggen *et al.* have reported higher levels of sRAGE in the inferior turbinate tissues from patients with CRSsNP in comparison to controls; sRAGE expression was also shown to be decreased in CRSwNP. The authors attributed the observed sRAGE increase in CRSsNP to elevated levels of ECM scaffolding proteins, as the cleaved form of sRAGE binds these proteins, compared to the disordered ECM environments found in CRSwNP.

CRSwNP is predominantly characterized by a Th2-biased cytokine profile and eosinophil infiltration[56,57]. S100A8 and S100A9 levels were found to be elevated in CRSwNP compared to controls and in CRSsNP, similar to observed in other conditions associated with a Th2-mediated pathophysiology[60,61]. In addition to psoriasin, S100A8/A9 exhibit antimicrobial effects in different biological contexts.[62,63] S100A8 and S100A9 are potent ligands of TLR4, and TLR4-mediated pathway activation can result in the production of cytokines that favor a Th2-biased adaptive immunity.[7,52] Although generally secreted as calprotectin by neutrophils, pro-inflammatory effectors commonly associated with CRSwNP, including *Staphylococcus aureus*, can promote the cleavage of this molecule,

leading to the activation of TLR4 by S100A8 and S100A9 and the initiation of host defense mechanisms[52].

Interestingly, S100A12 has also been shown to block the activity of S100A9. In a recent report, the binding domains of RAGE were examined. The authors reported that S100A12 can dimerize with S100A9, in turn restricting S100A9 from interacting with the RAGE V domain, which is essential for signal propagation[64]. S100A9 has been implicated in tumor growth and proliferation through binding to various innate immune receptors[65]. Although the role of RAGE compared to TLR4 may be minor in CRSwNP, it may contribute to CRSwNP pathology. While levels of S100A12 are elevated in CRSsNP and S100A9 in CRSwNP, perhaps the upregulation of S100A12 observed in CRSsNP is indicative of a more balanced immune environment, reducing the mechanisms that favor polyp growth.

CONCLUSIONS

This review highlights key findings demonstrating the differential expression patterns and proposed roles of innate immune-modulating s100 proteins in subtypes of CRS. Although only one study has been published on this topic within the last 18 months[49], increasing reports have focused on identifying new potential biomarkers of inflammation and innate immunity to further elucidate the complex inflammatory mechanisms contributing to CRS pathology. The literature described over the last decade illustrate such inflammatory pathway complexity in CRS. Differences in expression patterns of psoriasin (S100A7), calgranulins A (S100A8), B (S100A9), and C (S100A12), and calprotectin (S100A8/A9) were demonstrated in different sinonasal regions (*e.g.*, uncinate, inferior turbinate, anterior ethmoid, nasal polyp, blood, and NLF), tissue subsites (*e.g.*, epithelium, basement membrane, seromucous glands, and subepithelial stroma), and CRS phenotypes. Overall, these findings suggest inflammation may manifest differently with respect to sinus location and that similar tissue sampling regions should be analyzed for conducting intra- and interpatient comparisons of inflammatory profiling to fully characterize the pathophysiology in CRS.

ACKNOWLEDGEMENTS

DISCLOSURE OF FUNDING. Jeremiah A. Alt is supported by a grant from the Flight Attendant Medical Research Institute (CIA160008). Jeremiah A. Alt and Abigail Pulsipher are supported by a grant from the National Institute of Allergy and Infectious Diseases (R44AI126987). Jorgen S. Sumsion is supported by an award from the University of Utah Medical Student Research Program and the National Heart, Lung, and Blood Institute.

REFERENCES

1. Orlandi RR, Kingdom TT, Hwang PH, et al.: International Consensus Statement on Allergy and Rhinology: Rhinosinusitis. *Int Forum Allergy Rhinol* 2016, 6 Suppl 1:S22–209. [PubMed: 26889651]
2. Rosenfeld RM, Piccirillo JF, Chandrasekhar SS, et al.: Clinical practice guideline (update): adult sinusitis. *Otolaryngol Head Neck Surg* 2015, 152:S1–S39.
3. Hariri BM, Cohen NA: New insights into upper airway innate immunity. *Am J Rhinol Allergy* 2016, 30:319–323. [PubMed: 27657896]
4. Schleimer RP: Immunopathogenesis of Chronic Rhinosinusitis and Nasal Polyposis. *Annu Rev Pathol* 2016.

5. Lane AP, Truong-Tran QA, Schleimer RP: Altered expression of genes associated with innate immunity and inflammation in recalcitrant rhinosinusitis with polyps. *Am J Rhinol* 2006, 20:138–144. [PubMed: 16686375]
6. Detwiler KY, Smith TL, Alt JA, et al.: Differential expression of innate immunity genes in chronic rhinosinusitis. *Am J Rhinol Allergy* 2014, 28:374–377. [PubMed: 25198021]
7. Vogl T, Tenbrock K, Ludwig S, et al.: Mrp8 and Mrp14 are endogenous activators of Toll-like receptor 4, promoting lethal, endotoxin-induced shock. *Nat Med* 2007, 13:1042–1049. [PubMed: 17767165]
8. Wang X, Ji W, Xu Y, et al.: [Expression and significance of Toll like receptor 2 and Toll like receptor 4 in chronic rhinosinusitis]. *Lin Chung Er Bi Yan Hou Tou Jing Wai Ke Za Zhi* 2014, 28:1285–1288. [PubMed: 25522559]
9. Hirschberg A, Kiss M, Kadocska E, et al.: Different activations of toll-like receptors and antimicrobial peptides in chronic rhinosinusitis with or without nasal polyposis. *Eur Arch Otorhinolaryngol* 2016, 273:1779–1788. [PubMed: 26518209]
10. Shin SH, Ye MK, Kim YH, Kim JK: Role of TLRs in the production of chemical mediators in nasal polyp fibroblasts by fungi. *Auris Nasus Larynx* 2016, 43:166–170. [PubMed: 26254958]
11. Van Crombruggen K, Holtappels G, De Ruyck N, et al.: RAGE processing in chronic airway conditions: involvement of *Staphylococcus aureus* and ECP. *J Allergy Clin Immunol* 2012, 129:1515–1521 e1518. [PubMed: 22460069]
12. Van Crombruggen K, Jacob F, Zhang N, Bachert C: Damage-associated molecular patterns and their receptors in upper airway pathologies. *Cell Mol Life Sci* 2013, 70:4307–4321. [PubMed: 23673984]
13. Madsen P, Rasmussen HH, Leffers H, et al.: Molecular cloning, occurrence, and expression of a novel partially secreted protein “psoriasin” that is highly up-regulated in psoriatic skin. *J Invest Dermatol* 1991, 97:701–712. [PubMed: 1940442]
14. Vegfors J, Ekman AK, Stoll SW, et al.: Psoriasin (S100A7) promotes stress-induced angiogenesis. *Br J Dermatol* 2016, 175:1263–1273. [PubMed: 27155199]
15. Jansen S, Podschun R, Leib SL, et al.: Expression and function of psoriasin (S100A7) and koebnerisin (S100A15) in the brain. *Infect Immun* 2013, 81:1788–1797. [PubMed: 23478321]
16. Glaser R, Harder J, Lange H, et al.: Antimicrobial psoriasin (S100A7) protects human skin from *Escherichia coli* infection. *Nat Immunol* 2005, 6:57–64. [PubMed: 15568027]
17. Li X, de Leeuw E, Lu W: Total chemical synthesis of human psoriasin by native chemical ligation. *Biochemistry* 2005, 44:14688–14694. [PubMed: 16262268]
18. Michalek M, Gelhaus C, Hecht O, et al.: The human antimicrobial protein psoriasin acts by permeabilization of bacterial membranes. *Dev Comp Immunol* 2009, 33:740–746. [PubMed: 19162067]
19. Enerback C, Porter DA, Seth P, et al.: Psoriasin expression in mammary epithelial cells in vitro and in vivo. *Cancer Res* 2002, 62:43–47. [PubMed: 11782356]
20. Alowami S, Qing G, Emberley E, et al.: Psoriasin (S100A7) expression is altered during skin tumorigenesis. *BMC Dermatol* 2003, 3:1.
21. Ostergaard M, Wolf H, Orntoft TF, Celis JE: Psoriasin (S100A7): a putative urinary marker for the follow-up of patients with bladder squamous cell carcinomas. *Electrophoresis* 1999, 20:349–354. [PubMed: 10197442]
22. Foell D, Frosch M, Sorg C, Roth J: Phagocyte-specific calcium-binding S100 proteins as clinical laboratory markers of inflammation. *Clin Chim Acta* 2004, 344:37–51. [PubMed: 15149869]
23. Kessel C, Holzinger D, Foell D: Phagocyte-derived S100 proteins in autoinflammation: putative role in pathogenesis and usefulness as biomarkers. *Clin Immunol* 2013, 147:229–241. [PubMed: 23269200]
24. Rammes A, Roth J, Goebeler M, et al.: Myeloid-related protein (MRP) 8 and MRP14, calcium-binding proteins of the S100 family, are secreted by activated monocytes via a novel, tubulin-dependent pathway. *J Biol Chem* 1997, 272:9496–9502. [PubMed: 9083090]
25. Urban CF, Ermert D, Schmid M, et al.: Neutrophil extracellular traps contain calprotectin, a cytosolic protein complex involved in host defense against *Candida albicans*. *PLoS Pathog* 2009, 5:e1000639. [PubMed: 19876394]

26. Leach ST, Yang Z, Messina I, et al.: Serum and mucosal S100 proteins, calprotectin (S100A8/S100A9) and S100A12, are elevated at diagnosis in children with inflammatory bowel disease. *Scand J Gastroenterol* 2007, 42:1321–1331. [PubMed: 17852869]
27. Sidler MA, Leach ST, Day AS: Fecal S100A12 and fecal calprotectin as noninvasive markers for inflammatory bowel disease in children. *Inflamm Bowel Dis* 2008, 14:359–366. [PubMed: 18050298]
28. Oesterle A, Bowman MA: S100A12 and the S100/Calgranulins: Emerging Biomarkers for Atherosclerosis and Possibly Therapeutic Targets. *Arterioscler Thromb Vasc Biol* 2015, 35:2496–2507. [PubMed: 26515415]
29. Yamaoka M, Maeda N, Nakamura S, et al.: Gene expression levels of S100 protein family in blood cells are associated with insulin resistance and inflammation (Peripheral blood S100 mRNAs and metabolic syndrome). *Biochem Biophys Res Commun* 2013, 433:450–455. [PubMed: 23501102]
30. Ehrchen JM, Sunderkotter C, Foell D, et al.: The endogenous Toll-like receptor 4 agonist S100A8/S100A9 (calprotectin) as innate amplifier of infection, autoimmunity, and cancer. *J Leukoc Biol* 2009, 86:557–566. [PubMed: 19451397]
31. Chan JK, Roth J, Oppenheim JJ, et al.: Alarmins: awaiting a clinical response. *J Clin Invest* 2012, 122:2711–2719. [PubMed: 22850880]
32. Striz I, Trebichavsky I: Calprotectin - a pleiotropic molecule in acute and chronic inflammation. *Physiol Res* 2004, 53:245–253. [PubMed: 15209531]
33. Buhimschi CS, Baumbusch MA, Dulay AT, et al.: Characterization of RAGE, HMGB1, and S100beta in inflammation-induced preterm birth and fetal tissue injury. *Am J Pathol* 2009, 175:958–975. [PubMed: 19679874]
34. Jung ES, Chung W, Kim AJ, et al.: Associations between Soluble Receptor for Advanced Glycation End Products (sRAGE) and S100A12 (EN-RAGE) with Mortality in Long-term Hemodialysis Patients. *J Korean Med Sci* 2017, 32:54–59. [PubMed: 27914132]
35. Shubbar E, Vegfors J, Carlstrom M, et al.: Psoriasin (S100A7) increases the expression of ROS and VEGF and acts through RAGE to promote endothelial cell proliferation. *Breast Cancer Res Treat* 2012, 134:71–80. [PubMed: 22189627]
36. Rauvala H, Rouhiainen A: RAGE as a receptor of HMGB1 (Amphoterin): roles in health and disease. *Curr Mol Med* 2007, 7:725–734. [PubMed: 18331230]
37. Rouhiainen A, Kuja-Panula J, Tumova S, Rauvala H: RAGE-mediated cell signaling. *Methods Mol Biol* 2013, 963:239–263. [PubMed: 23296615]
38. Hofmann MA, Drury S, Fu C, et al.: RAGE mediates a novel proinflammatory axis: a central cell surface receptor for S100/calgranulin polypeptides. *Cell* 1999, 97:889–901. [PubMed: 10399917]
39. Cheng Z, Dai LL, Cao DF, et al.: [Changes of HMGB1 and RAGE in induced sputum from patients with bronchial asthma]. *Zhonghua Yi Xue Za Zhi* 2011, 91:1538–1542. [PubMed: 21914366]
40. Jinquan T, Vorum H, Larsen CG, et al.: Psoriasin: a novel chemotactic protein. *J Invest Dermatol* 1996, 107:5–10. [PubMed: 8752830]
41. Ryckman C, McColl SR, Vandal K, et al.: Role of S100A8 and S100A9 in neutrophil recruitment in response to monosodium urate monohydrate crystals in the air-pouch model of acute gouty arthritis. *Arthritis Rheum* 2003, 48:2310–2320. [PubMed: 12905486]
42. Ryckman C, Vandal K, Rouleau P, et al.: Proinflammatory activities of S100: proteins S100A8, S100A9, and S100A8/A9 induce neutrophil chemotaxis and adhesion. *J Immunol* 2003, 170:3233–3242. [PubMed: 12626582]
43. Yang Z, Yan WX, Cai H, et al.: S100A12 provokes mast cell activation: a potential amplification pathway in asthma and innate immunity. *J Allergy Clin Immunol* 2007, 119:106–114. [PubMed: 17208591]
44. Yan WX, Armishaw C, Goyette J, et al.: Mast cell and monocyte recruitment by S100A12 and its hinge domain. *J Biol Chem* 2008, 283:13035–13043. [PubMed: 18292089]
45. Yang Z, Tao T, Raftery MJ, et al.: Proinflammatory properties of the human S100 protein S100A12. *J Leukoc Biol* 2001, 69:986–994. [PubMed: 11404386]

46. Bryborn M, Adner M, Cardell LO: Psoriasin, one of several new proteins identified in nasal lavage fluid from allergic and non-allergic individuals using 2-dimensional gel electrophoresis and mass spectrometry. *Respir Res* 2005, 6:118. [PubMed: 16236163]
47. Tieu DD, Peters AT, Carter RG, et al.: Evidence for diminished levels of epithelial psoriasin and calprotectin in chronic rhinosinusitis. *J Allergy Clin Immunol* 2010, 125:667–675. [PubMed: 20226301]
48. Cho SH, Kim DW, Lee SH, et al.: Age-related increased prevalence of asthma and nasal polyps in chronic rhinosinusitis and its association with altered IL-6 trans-signaling. *Am J Respir Cell Mol Biol* 2015, 53:601–606. [PubMed: 26266960]
- *49. Pulsipher A, Davis BM, Smith KA, et al.: Calgranulin C (S100A12) Is Differentially Expressed in Subtypes of Chronic Rhinosinusitis. *Am J Rhinol Allergy* 2018, 32:380–387. [PubMed: 29943619] This study demonstrated the increased protein levels of calgranulin C in CRSsNP compared to CRSwNP, and the correlation of calgranulin C protein levels with disease severity in patients with CRSsNP.
50. Kvarnhammar AM, Rydberg C, Jarnkrants M, et al.: Diminished levels of nasal S100A7 (psoriasin) in seasonal allergic rhinitis: an effect mediated by Th2 cytokines. *Respir Res* 2012, 13:2. [PubMed: 22230654]
51. Richer SL, Truong-Tran AQ, Conley DB, et al.: Epithelial genes in chronic rhinosinusitis with and without nasal polyps. *American journal of rhinology* 2008, 22:228–234. [PubMed: 18588753]
52. Van Crombruggen K, Vogl T, Perez-Novo C, et al.: Differential release and deposition of S100A8/A9 proteins in inflamed upper airway tissue. *Eur Respir J* 2016, 47:264–274. [PubMed: 26493794]
53. Benninger MS, Ferguson BJ, Hadley JA, et al.: Adult chronic rhinosinusitis: definitions, diagnosis, epidemiology, and pathophysiology. *Otolaryngol Head Neck Surg* 2003, 129:S1–32. [PubMed: 12958561]
54. Bellussi LM, Chen L, Chen D, et al.: The role of High Mobility Group Box 1 chromosomal protein in the pathogenesis of chronic sinusitis and nasal polyposis. *Acta Otorhinolaryngol Ital* 2012, 32:386–392. [PubMed: 23349558]
55. Tardif MR, Chapeton-Montes JA, Posvanzic A, et al.: Secretion of S100A8, S100A9, and S100A12 by Neutrophils Involves Reactive Oxygen Species and Potassium Efflux. *J Immunol Res* 2015, 2015:296149. [PubMed: 27057553]
56. Van Zele T, Claeys S, Gevaert P, et al.: Differentiation of chronic sinus diseases by measurement of inflammatory mediators. *Allergy* 2006, 61:1280–1289. [PubMed: 17002703]
57. Tomassen P, Vandeplas G, Van Zele T, et al.: Inflammatory endotypes of chronic rhinosinusitis based on cluster analysis of biomarkers. *J Allergy Clin Immunol* 2016, 137:1449–1456 e1444. [PubMed: 26949058]
58. Ramasamy R, Yan SF, Schmidt AM: RAGE: therapeutic target and biomarker of the inflammatory response--the evidence mounts. *J Leukoc Biol* 2009, 86:505–512. [PubMed: 19477910]
59. Wittkowski H, Hirono K, Ichida F, et al.: Acute Kawasaki disease is associated with reverse regulation of soluble receptor for advance glycation end products and its proinflammatory ligand S100A12. *Arthritis Rheum* 2007, 56:4174–4181. [PubMed: 18050248]
60. Pham DL, Yoon MG, Ban GY, et al.: Serum S100A8 and S100A9 Enhance Innate Immune Responses in the Pathogenesis of Baker's Asthma. *Int Arch Allergy Immunol* 2015, 168:138–146. [PubMed: 26745257]
61. Gittler JK, Shemer A, Suarez-Farinas M, et al.: Progressive activation of T(H)2/T(H)22 cytokines and selective epidermal proteins characterizes acute and chronic atopic dermatitis. *J Allergy Clin Immunol* 2012, 130:1344–1354. [PubMed: 22951056]
62. Kehl-Fie TE, Chitayat S, Hood MI, et al.: Nutrient metal sequestration by calprotectin inhibits bacterial superoxide defense, enhancing neutrophil killing of *Staphylococcus aureus*. *Cell Host Microbe* 2011, 10:158–164. [PubMed: 21843872]
63. Sohnle PG, Hunter MJ, Hahn B, Chazin WJ: Zinc-reversible antimicrobial activity of recombinant calprotectin (migration inhibitory factor-related proteins 8 and 14). *J Infect Dis* 2000, 182:1272–1275. [PubMed: 10979933]

- *64. Katte R, Yu C: Blocking the interaction between S100A9 protein and RAGE V domain using S100A12 protein. *PLoS One* 2018, 13:e0198767. [PubMed: 29902210] This study investigated the binding of calgranulin B and calgranulin C with the RAGE V domain, suggesting that calgranulin C could act as an inhibitor of calgranulin B.
65. Kallberg E, Vogl T, Liberg D, et al.: S100A9 interaction with TLR4 promotes tumor growth. *PLoS One* 2012, 7:e3420

KEY POINTS

- Calgranulins have been identified as emerging biomarkers of inflammation.
- S100 proteins are differentially expressed in subtypes of CRS.
- S100 proteins demonstrate different expression patterns with respect to location of tissue sampling (*e.g.*, uncinate, inferior turbinate, nasal polyp, anterior ethmoid, blood, and nasal lavage fluid) and tissue subsite (*e.g.*, epithelium, basement membrane, seromucous glands, and subepithelial stroma).
- S100 proteins exert unique and concerted roles in mediating immunity in CRS.

The reported expression patterns of S100 proteins in chronic rhinosinusitis with respect to the results for each S100 gene or protein, analysis method, location of specimen sampling, and overall conclusions from each respective investigation.

Table 1:

Reference	Protein	Method	Specimen Analyzed	Comparison	Results	Proposed Biological Roles
Tieu et al., 2010[47]	S100A7	ELISA	Inferior turbinate, Uncinate	Control	S100A7 is significantly elevated in inferior turbinate tissue compared to uncinata tissue.	Reduced immune barrier function in patients with CRS might underlie their increased susceptibility to infections, bacterial and fungal colonization, or both in the upper airways and sinuses.
	S100A7	ELISA	Uncinate	Control vs. CRS Subtypes	S100A7 is significantly upregulated in uncinata tissue from CRSsNP compared to CRSwNP and Controls. The expression of S100A7 is significantly increased in CRSwNP compared to controls.	
	S100A8/A ₉	ELISA	Nasal polyp, uncinata, inferior turbinate	Control vs. CRS Subtypes	S100A8/A9 is significantly increased in polyp tissue compared to uncinata and inferior turbinate tissues from CRSwNP, CRSsNP, and controls.	
	S100A7	ELISA	NLF	Control vs. CRS Subtypes	S100A7 is significantly decreased in NLF from CRSwNP and CRSsNP compared to controls.	
	S100A8/A ₉	ELISA	NLF	Control vs. CRS Subtypes	S100A8/A9 is significantly decreased in NLF from CRSwNP compared to controls.	
	S100A7	ELISA	Serum	Control vs. CRS Subtypes	S100A7 is decreased (not significant) in serum from CRSsNP and CRSwNP compared to controls.	
	S100A8/A ₉	ELISA	Serum	Control vs. CRS Subtypes	S100A8/A9 is significantly decreased in serum from CRSwNP compared to CRSsNP and controls.	
	S100A7	IHC	Uncinate	Control vs. CRS Subtypes	S100A7 is present in the nasal respiratory epithelium, glands, and subepithelial stroma of uncinata tissues from CRSwNP, CRSsNP, and controls.	
	S100A8/A ₉	IHC	Uncinate	Control vs. CRS Subtypes	S100A8/A9 is present in the nasal respiratory epithelium, glands and subepithelial stroma of uncinata tissue of CRSwNP, CRSsNP, and controls.	
Van Crombruggen et al., 2016[52]	S100A8	ELISA	Nasal polyp, control inferior turbinate	Control vs. CRSwNP	S100A8 is significantly upregulated in polyp tissue compared to inferior turbinate tissue from controls.	The inflammatory and remodeling characteristics of CRSwNP specifically allow for the increased retention of S100A8, S100A9, and S100A8/A9 in the ECM of CRSwNP tissues. Upon release, homodimeric proteins

Reference	Protein	Method	Specimen Analyzed	Comparison	Results	Proposed Biological Roles
	S100A9	ELISA	Nasal polyp, control inferior turbinate	Control vs. CRSwNP	S100A9 is significantly increased in polyp tissue compared to inferior turbinate tissue from controls.	act as local danger signals, inducing inflammatory mediators, predominantly via TLR4 activation.
	S100A8/A9	ELISA	Nasal polyp, control inferior turbinate	Control vs. CRSwNP	S100A8/A9 was significantly increased in polyp tissue compared to inferior turbinate tissue from controls.	
	S100A8	IHC	Nasal polyp, control inferior turbinate, CRSwNP inferior turbinate	Control vs. CRSwNP	Inferior turbinate tissues from controls showed negligible S100A8 expression. Inferior turbinate and nasal polyp tissues from CRSwNP showed pronounced S100A8 expression around the basement membrane, blood vessels, and ECM structures and was also expressed by monocytes and neutrophils.	
	S100A9	IHC	Nasal polyp, control inferior turbinate, CRSwNP inferior turbinate	Control vs. CRSwNP	Inferior turbinate tissues from controls showed negligible S100A9 expression. Inferior turbinate and nasal polyp tissues from CRSwNP showed pronounced S100A9 expression around the basement membrane, blood vessels, and ECM structures and was also expressed by monocytes and neutrophils.	
	S100A8/A9	IHC	Nasal polyp, control inferior turbinate, CRSwNP inferior turbinate	Control vs. CRSwNP	Inferior turbinate tissues from controls showed negligible S100A8/A9 expression. Inferior turbinate and nasal polyp tissues from CRSwNP showed pronounced S100A8/A9 expression around the basement membrane, blood vessels, and ECM structures and was also expressed by monocytes and neutrophils.	
	S100A8	mRNA	Nasal polyp, control inferior turbinate	Control vs. CRSwNP	The mRNA expression level of S100A8 did not differ between nasal polyp tissues from CRSwNP and inferior turbinate tissue from controls.	
	S100A9	mRNA	Nasal polyp, control inferior turbinate	Control vs. CRSwNP	The mRNA expression level of S100A9 did not differ between nasal polyp tissues from CRSwNP and inferior turbinate tissue from controls.	
Richer et al., 2008[51]	S100A7	mRNA	Inferior turbinate, uncinata	Control vs. CRS Subtypes	The mRNA expression level of S100A7 is significantly decreased in inferior turbinate and uncinata tissues	This study showed marked reductions in the expression of several genes involved in epithelial barrier

Reference	Protein	Method	Specimen Analyzed	Comparison	Results	Proposed Biological Roles
					from CRSwNP and CRSsNP compared to controls.	maintenance and repair in the inflammatory state of CRS.
	S100A8	mRNA	Inferior turbinate, uncinata	Control vs. CRS Subtypes	The mRNA expression level of S100A8 is significantly decreased in inferior turbinate and uncinata tissues from CRSwNP and CRSsNP compared to controls.	
	S100A9	mRNA	Inferior turbinate, uncinata	Control vs. CRS Subtypes	The mRNA expression level of S100A9 is significantly decreased in inferior turbinate and uncinata tissues from CRSwNP compared to controls.	
Kvarnhammar et al., 2012[50]	S100A7	IHC	Inferior turbinate	Control	S100A7 is present in epithelium and seromucous glands of inferior turbinate tissue from controls.	S100A7 levels are diminished in rhinitis patients, suggesting that the antimicrobial defense system is compromised in patients with SAR.
Cho et al., 2015[48]	S100A8/A9	ELISA	Nasal polyp	Age of CRSwNP	The expression of S100A8/A9 decreases with ageing.	Epithelial barrier dysfunction may play a particularly important role in the pathogenesis of the CRSwNP in the elderly.
Bryborn et al., 2005[46]	S100A7	Two-dimensional gel electrophoresis with mass spectrometry	NLF	Control vs Allergic rhinosinusitis	The NLF expression of S100A7 is decreased in allergic rhinosinusitis and controls.	S100A7 is a potent chemotactic factor, and its downregulation during inflammation might be important in CRS.
Pulsipher et al., 2018[49]*	S100A12	ELISA	Anterior ethmoid	Control vs. CRS Subtypes	S100A12 is significantly increased in CRSsNP compared to CRSwNP and controls.	S100A12 is differentially expressed in subtypes of CRS and is significantly elevated in patients with CRSsNP and associated with CRS-specific disease severity. S100A12 expression is negatively correlated with HNE expression in CRSwNP.
	S100A12	IHC	Anterior ethmoid	Control vs. CRS Subtypes	S100A12 is increased in CRSsNP compared to CRSwNP and controls.	

CRS (chronic rhinosinusitis); CRSsNP (chronic rhinosinusitis without nasal polyps); CRSwNP (chronic rhinosinusitis with nasal polyps); ELISA (enzyme-linked immunosorbent assay); IHC (immunohistochemistry); mRNA (maternal ribonucleic acid); NLF (nasal lavage fluid); S100A7 (psoriasin); S100A8 (calgranulin A); S100A9 (calgranulin B); S100A8/A9 (calprotectin); S100A12 (calgranulin c)