Nitric Oxide xxx (2013) xxx–xxx

Contents lists available at SciVerse ScienceDirect

Nitric Oxide

journal homepage: www.elsevier.com/locate/yniox

Review

Hydrogen sulfide chemical biology: Pathophysiological roles and detection ☆,☆☆

Gopi K. Kolluru, Xinggui Shen, Shyamal C. Bir, Christopher G. Kevil *

Department of Pathology, LSU Health-Shreveport, United States

ARTICLE INFO

Article history:
Available online xxxx

Keywords:
Sulfide
Cysteine
Nitric oxide
Cardiovascular
Oxidative stress

ABSTRACT

Hydrogen sulfide (H_2S) is the most recent endogenous gasotransmitter that has been reported to serve many physiological and pathological functions in different tissues. Studies over the past decade have revealed that H_2S can be synthesized through numerous pathways and its bioavailability regulated through its conversion into different biochemical forms. H_2S exerts its biological effects in various manners including redox regulation of protein and small molecular weight thiols, polysulfides, thiosulfate/sulfite, iron-sulfur cluster proteins, and anti-oxidant properties that affect multiple cellular and molecular responses. However, precise measurement of H_2S bioavailability and its associated biochemical and pathophysiological roles remains less well understood. In this review, we discuss recent understanding of H_2S chemical biology, its relationship to tissue pathophysiological responses and possible therapeutic uses.

Contents

☆☆ Funded by NIH HL11331 to C.G.K and LSU Health Sciences Center Feist Cardiovascular Fellowships to G.K.K. and S.C.B.

* Corresponding author. Address: Department of Pathology, LSU Health Sciences Center-Shreveport, 1501 Kings Hwy, Shreveport, LA 71130, United States.
E-mail address: ckevil@lsuhsc.edu (C.G. Kevil).

1089-8603/$ - see front matter © 2013 The Authors. Published by Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.niox.2013.07.002

Please cite this article in press as: G.K. Kolluru et al., Hydrogen sulfide chemical biology: Pathophysiological roles and detection, Nitric Oxide (2013), http://dx.doi.org/10.1016/j.niox.2013.07.002

Introduction

Hydrogen sulfide (H_2S) has emerged as an important gaseous signaling molecule playing numerous roles in health and disease, along with CO and NO [1,2]. It acts as a relaxant of smooth muscle and thus a vasodilator, a regulator of cardiac function and *N*-methyl *D*-aspartate (NMDA) receptor in the brain, and cytoprotectant and mediator for cardiovascular therapeutic approaches [3–8]. Understanding precise pathophysiological signaling mechanisms and the metabolism of H_2S is a topic of active research. And unraveling its interactions within different tissues, with other biochemical molecules and various signaling mediators is becoming ever more complex. Thus, in this rapidly growing field it is important to appreciate key findings and their potential implications regarding H_2S chemical biology that are now known, as well as to identify critical areas for further understanding and clarification. In this review, we address numerous salient issues regarding biochemical synthesis and metabolism of H_2S, measurement techniques used to detect levels of H_2S both *in vitro* and *in vivo*, biological functions regulated by H_2S and the potential of therapeutic approaches employing H_2S based therapies for various clinical applications.

H_2S biosynthesis: enzymatic and non-enzymatic

The production of H_2S can occur via two pathways – enzymatic and non-enzymatic. Enzymatic synthesis of H_2S occurs through three enzymes that are cystathionine gamma-lyase (CGL also abbreviated as CSE), cystathionine beta-synthase (CBS) and 3-mercaptopyruvate sulfurtransferase (3-MST). These enzymes have been reported to be organ-specific depending on the type of enzyme. CBS is predominantly found in the brain, nervous system, and liver [4], while CSE is mostly found in the vasculature and liver, and 3-MST can be found in the brain and vasculature [9]. However, all three enzymes are distributed across many tissues and are often jointly present such as CBS and CSE being most prominently found in the liver and kidney [9]. Importantly, while CSE and CBS are hemeproteins primarily located in the cytosol, 3-MST is a zinc-dependent protein found in both the mitochondria and cytosol.

Cystathionine is a critical intermediate metabolite involved in many sulfur-containing amino acids, formed by CBS through condensation of homocysteine along with serine. CSE is also involved in a reaction that converts L-cystathionine and cysteine to form L-cysteine and α-ketoglutarate (α-KG). CSE and CBS ultimately produce H_2S through a reaction involving the generation of L-cysteine, pyruvate, and ammonia. Likewise, 3-MST produces H_2S through a reaction involving the generation of pyruvate from 3-mercaptopyruvate (3-MP). It has recently been shown that 3-MST might also cleave mercaptopyruvate to form pyruvate and H_2S, or catalyze the transsulfuration of a thiol to a persulfide, which can subsequently join a second thiol to form a disulfide and release H_2S [10]. 3-MP substrate is provided through the metabolism of cysteine and α-KG by cysteine aminotransferase (CAT). Fig. 1 illustrates how these enzymes coordinately regulate transsulfuration activity controlling physiological H_2S levels in a complex and overlapping manner.

Apart from enzymatic synthesis pathways, endogenous production of H_2S can also occur through other non-enzymatic processes that are less well understood. Non-enzymatic production of H_2S occurs through glucose, glutathione, inorganic and organic polysulfides (present in garlic) and elemental sulfur. H_2S can be generated from glucose either via glycolysis or from phosphogluconate via NADPH oxidase. Glucose reacts with methionine, homocysteine or cysteine to produce gaseous sulfur compounds – methanethiol and hydrogen sulfide. H_2S is also produced through direct reduction of glutathione and elemental sulfur. Reduction of elemental sulfur to H_2S is mediated through reducing equivalents of the glucose oxidation pathway such as NADH, or NADPH [11]. H_2S formation from thiosulfate results from a reductive reaction involving pyruvate, which acts as a hydrogen donor. Thiosulfate is an intermediate of sulfur metabolism from cysteine and a metabolite of H_2S that can also lead to the production of H_2S [12–14]. Though the involvement of mitochondria in oxidizing sulfide has been well known [15], the specific affinity of the enzyme 3-MST to thiosulfate and in its production has been recently reported [16,17]. Further, rhodanese is involved in metabolizing thiosulfate into H_2S and sulfite [13]. Fig. 1 summarizes various enzymatic and non-enzymatic H_2S synthesis pathways that have been described.

Different biochemical forms of sulfide

Apart from its free state, H_2S can react with different biochemical molecules establishing different bioavailable pools including stable, acid-labile and bound sulfide forms (Fig. 2). In the stable pool, sulfur atoms are not readily reactive or liberated upon chemical treatment with acid or dithiothreitol [14]. These compounds exist in a reduced divalent form such as H_2S or oxidized hexavalent form such as sulfate anion. Sulfide can also be categorized based on the form in which they are stored in a biological system such as free sulfides or in bound forms such as acid-labile forms and bound sulfane sulfur [18]. Free or unbound sulfide exists as S^{2-}, HS^- or H_2S, where its acidic dissociation constants (pKa) range between >12 and 7.0, respectively (Fig. 2). Acid-labile sulfide is mainly in the form of iron–sulfur (Fe–S) complexes and persulfides, which play a critical role in redox reactions in cytoplasm and mitochondria. The critical pH below which H_2S is released from acid-labile sulfur like Fe–S is 5.4 [19]. Conversely, bound sulfane sulfur exists as a compound containing sulfur-bonded sulfur [20]. This includes compounds like polysulfides, thiosulfate, polythionates, thiosulfonates bisorganyl-polysulfanes or monoarylthiosulfonates and elemental sulfur. Bound-sulfane sulfur compounds such as polysulfides release H_2S under reducing conditions suggesting that cellular redox state is important for regulating its bioavailability [21]. Moreover, free H_2S can be incorporated into proteins as bound sulfane sulfur, where its divalent sulfur form binds only to the elemental sulfur, persulfides and polysulfides [22]. These various biochemical forms represent complex and diverse ways in which H_2S bioavailability can be maintained. However, the movement

Please cite this article in press as: G.K. Kolluru et al., Hydrogen sulfide chemical biology: Pathophysiological roles and detection, Nitric Oxide (2013), http://dx.doi.org/10.1016/j.niox.2013.07.002