Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Inorganic Biochemistry 99 (2005) 1–22
JOURNAL OF Inorganic Biochemistry
www.elsevier.com/locate/jinorgbio

Heme-based sensors: defining characteristics, recent developments, and regulatory hypotheses

Marie-Alda Gilles-Gonzalez *, Gonzalo Gonzalez

Department of Biochemistry, University of Texas Southwestern Medical Center, 5323 Harry Hines Boulevard, Dallas, TX 75390-9038, USA

Received 27 March 2004; received in revised form 5 August 2004; accepted 24 October 2004

Abstract

In a great variety of organisms throughout all kingdoms of life, the heme-based-sensor proteins are the key regulators of adaptive responses to fluctuating oxygen, carbon monoxide, and nitric oxide levels. These signal transducers achieve their responses by coupling a regulatory heme-binding domain to a neighboring transmitter. The past decade has witnessed an explosion in the numbers of these modular sensory proteins known, from just two recognized members, FixL and soluble guanylyl cyclase (sGC), to four broad families comprising more than 50 sensors. Heme-based sensors so far feature four different types of heme-binding modules: the heme-binding PAS domain, globin-coupled sensor (GCS), CooA, and heme–NO-binding (HNOB). The transmitters for coupling to such heme-binding domains include histidine protein kinases, cyclic nucleotide phosphodiesterases, chemotaxis methyl-carrier protein receptors, and transcription factors of the basic helix-loop-helix and helix-turn-helix classes. Some well-studied sensors are the FixL, *Ec*Dos, *Ax*PDEA1, NPAS2, HemAT-*Bs*, HemAT-*Hs*, CooA, and sGC proteins. This review elaborates the defining characteristics of heme-based sensors, examines recent developments on those proteins, and discusses the regulatory hypotheses proposed for those sensors. A general, "helix-swap", model is also proposed here for signal transduction by PAS domains.

Keywords: Hemoglobin; Myoglobin; Oxygen sensor; Guanylyl cyclase; Response regulator; Sensor kinase

1. Introduction

The past decade has introduced a greater variety of ligand-binding heme proteins than all previous years put together. This has expanded our knowledge of the strategies with which these proteins achieve their architectures and control their properties. This has also made us realize that much of what we thought we could conclude about ligand-binding heme proteins may have been the characteristics of individual classes of proteins. For example, many heme–protein enzymes are actually signal transducers where the heme center is directly concerned with regulation rather than catalysis [1–12]. The traditional relations of the affinities of hemoglobins for ligands, i.e. $O_2 << CO <<< NO$, do not extend to other heme proteins designed for ligand binding, or even O_2 binding [8,13,14]. Many proteins with hexacoordinate heme iron function in reversible binding of ligands rather than transfer of electrons [8,15–24]. For at least one class of O_2-binding heme proteins, the iron–histidine bond stretches determined by resonance Raman spectroscopy do not correlate with O_2 affinity [25]. The protein scaffold for a heme may consist entirely of β strands, a mixture of α helices and β strands, or α helices differing in number and arrangement from those in myoglobins [26–30]. Study of a greater variety of these ligand-binding heme proteins should reveal some unifying principles about their behaviors. Although currently it is again difficult

* Corresponding author. Tel.: +1 214 648 9438; fax: +1 214 648 8856.
E-mail address: magg@biochem.swmed.edu (M.-A. Gilles-Gonzalez).

0162-0134/$ - see front matter © 2004 Elsevier Inc. All rights reserved.
doi:10.1016/j.jinorgbio.2004.11.006

to rationalize those behaviors, it is also quite exciting to consider them from first principles. Many of the changes in thinking about heme-based sensors began with their recognition as a distinct functional class and the expectation that their diverse requirements for sensing would lead to a broad range of characteristics [1]. Discovery of heme-based sensors has rapidly accelerated (Fig. 1) [1–3,8–12,15,16,25,31–33].

2. What constitutes a biological heme-based sensor?

In a biological heme-based sensor, a regulatory heme-binding domain or subunit controls a neighboring transmitter region of the same protein [1]. Such signal-transducing heme proteins govern adaptative responses to fluctuations in O_2, CO, or NO: all three being diatomic gases that are now appreciated as physiological messengers. As a class of heme proteins, the sensors are distinct from the carriers of gases and the catalysts of oxygen-atom and electron transfer reactions [1]. The transmitter regions of heme-based sensors typically feature modules that also transduce signals in many non-heme proteins; such modules include histidine protein kinase, cyclic-dinucleotide phosphodiesterase, nucleotide cyclase, chemotaxis receptor, and DNA-binding transcription-factor activities [1–3,8–12,15,16,31–33]. The regulatory domains, on the other hand, have several architectural and sequence motifs that are entirely novel for heme binding, such as Per-Arnt-Sim (PAS), cAMP-receptor-like, and modified-globin domains [26,28,30]. Heme-based sensors provide excellent models for study of signal transduction, with the iron center supplying a built-in probe of the sensor's status and the transmitter reporting the switching potential of any state of the heme.

An effective sensor must:

- Bind its signal ligand over the concentration range appropriate for "switching" an activity.
- Switch an activity on binding of its signal ligand, i.e., trigger a change from an active to an inactive state, or vice versa (Fig. 2).
- Discriminate against false signals if this is physiologically necessary (Figs. 2(b) and (c)).
- Switch its conformation on binding of its signal ligand.

2.1. Signal binding in an appropriate range and discrimination at the binding step

Heme-based sensors are biochemical tools for physiological adaptation. As such, they recognize their signal based principally on the organism and environment in which they have evolved to function. For example, the *Rm*FixL O_2 sensor ($K_d \sim 50$ μM) triggers the expression of *Sinorhizobium meliloti nif* and *fix* genes as the *Rhizo-*

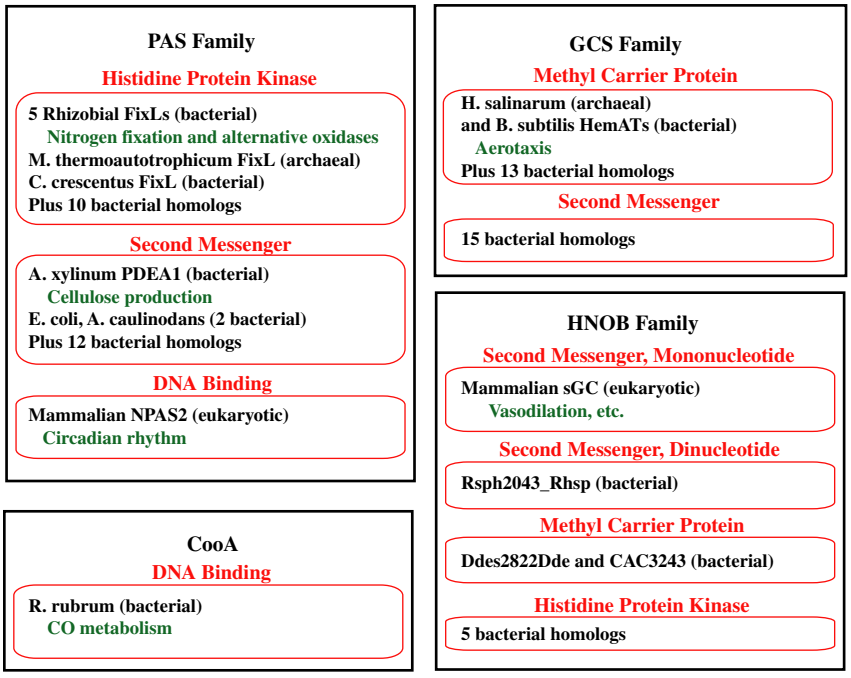

Fig. 1. Families of heme-based sensors. A distinctive heme-binding domain defines each family of sensors. Subgroups (red boxes) within the families couple their heme-binding domain to different transmitters for signal transduction. Those proteins specifically named are ones that have been purified and established as heme proteins. The physiological functions, if known, are highlighted in green. The last line in each category notes the numbers and kingdom of additional members expected from sequence homology.