Biochimica et Biophysica Acta xxx (2014) xxx–xxx

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

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

Review

Mammalian lipoxygenases and their biological relevance☆

Hartmut Kuhn [a,*], Swathi Banthiya [a], Klaus van Leyen [b]

[a] Institute of Biochemistry, University Medicine Berlin — Charite, Charitеplatz 1, CCO-Building, Virchowweg 6, D-10117 Berlin, Germany
[b] Neuroprotection Research Laboratory, Department of Radiology, Massachusetts Genrel Hospital and Harvard Medical School, Charlestown, MA, USA

ARTICLE INFO

Article history:
Received 12 June 2014
Received in revised form 30 September 2014
Accepted 3 October 2014
Available online xxxx

Keywords:
Eicosanoid
Inflammation
Atherosclerosis
Cancer
Stroke
Infection

ABSTRACT

Lipoxygenases (LOXs) form a heterogeneous class of lipid peroxidizing enzymes, which have been implicated not only in cell proliferation and differentiation but also in the pathogenesis of various diseases with major public health relevance. As other fatty acid dioxygenases LOXs oxidize polyunsaturated fatty acids to their corresponding hydroperoxy derivatives, which are further transformed to bioactive lipid mediators (eicosanoids and related substances). On the other hand, lipoxygenases are key players in the regulation of the cellular redox homeostasis, which is an important element in gene expression regulation. Although the first mammalian lipoxygenases were discovered 40 years ago and although the enzymes have been well characterized with respect to their structural and functional properties the biological roles of the different lipoxygenase isoforms are not completely understood. This review is aimed at summarizing the current knowledge on the physiological roles of different mammalian LOX-isoforms and their patho-physiological function in inflammatory, metabolic, hyperproliferative, neurodegenerative and infectious disorders. This article is part of a Special Issue entitled "Oxygenated metabolism of PUFA: Analysis and biological relevance."

1. Introduction

Lipoxygenases (LOXs) are non-heme iron-containing dioxygenases [1,2] that catalyze dioxygenation of polyunsaturated fatty acids containing at least two isolated cis-double bonds (Fig. 1). In mammalian cells linoleic acid (C18:Δ2, n−6) and arachidonic acid (C20:Δ4, n−6) are the most abundant polyenoic fatty acids that serve as substrates for the different mammalian LOX-isoforms. In general, mammalian LOXs prefer free fatty acids as substrate over polyenoic fatty acid containing ester lipids but the cellular concentration of free fatty acids is rather low. Thus, an active LOX pathway requires liberation of substrate fatty acids from the ester lipids localized in the cellular membranes. After hydrolytic cleavage of the membrane ester lipids catalyzed by cytosolic phospholipase A2 [3] the liberated fatty acids [mainly arachidonic acid (AA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA)] are alternatively oxygenated by cyclooxygenases (COXs) to G-prostaglandins (PGG2 in the case of AA, PGG3 in the case of EPA) or by LOX isoforms to various hydroperoxy derivatives of the substrate fatty acids [2]. The primary products of the LOX pathway are subsequently converted to a large array of bioactive lipid mediators, which include leukotrienes [4], lipoxins [5], hepoxilins [6], eoxins [7], resolvins [8], protectins [9] and others originating from AA, EPA and DHA. However, the classical concept of the arachidonic acid cascade may not be the only way, by which LOXs exhibit their bioactivity. There are at least two alternative scenarios (Fig. 2): i) Some LOX isoforms are capable of oxygenating polyenoic fatty acids if they are constituents of phospholipids [10] or cholesterol esters [11]. The introduction of a hydrophilic peroxide group into the hydrophobic tail of a fatty acid changes the physicochemical properties of the ester lipids. Clustering of oxidized lipids within the lipid bilayer of a biomembrane leads to the formation of "hydrophilic pores". By this mechanism the barrier function of the membrane is impaired which may lead to cellular dysfunction. ii) The cellular redox state is of major cell physiological relevance. It impacts the gene expression pattern of a given cell population [12] on transcriptional and post-transcriptional levels and thus determines the cellular phenotype. In each cell the redox homeostasis is maintained by the balanced equilibrium of pro- and anti-oxidative processes and LOXs constitute some of the key pro-oxidative players in the redox homeostasis. LOX-catalyzed formation of hydroperoxy lipids impacts the activity of redox-dependent transcription and/or translation factors [13], which in turn leads to up- and/or down-regulation of the expression of redox sensitive genes.

The molecular details of how the different LOX-isoforms exhibit their bioactivity have been explored for many years and a large number of reports employing various loss-of-function (siRNA-mediated expression knockdown, knockout mice) as well as gain-of-function (cellular transfection studies, transgenic animals) strategies have provided a deeper insight into the biological importance of LOXs in health and

Abbreviations: LOX, lipoxygenase; COX, cyclooxygenase; LT, leukotriene; HETE, hydroxyeicosatetraenoic acid; PPAR, peroxisome proliferator activated receptor; MICAL, molecule interacting with CasL; ARCI, autosomal recessive congenital ichthyosis; GPCR, G-protein coupled receptors; NOD, nonobese diabetes; RSV, respiratory syncytial virus; NDGA, nordihydroguaiaretic acid; ALX, lipoxin; OXE, oxo-eicosanoid

☆ This article is part of a Special Issue entitled "Oxygenated metabolism of PUFA: Analysis and biological relevance."

* Corresponding author. Tel.: +49 30 450528040; fax: +49 30 450528905.
E-mail address: hartmut.kuehn@charite.de (H. Kuhn).

http://dx.doi.org/10.1016/j.bbalip.2014.10.002
1388-1981/

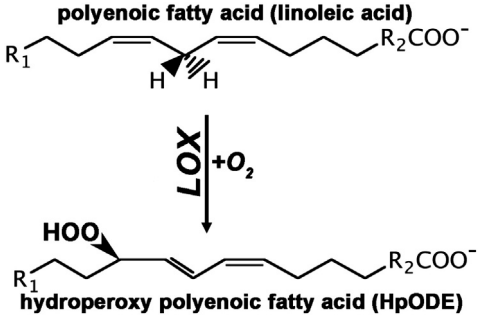

Fig. 1. Simplified scheme of the lipoxygenase reaction. LOXs convert polyenoic fatty acids containing at least one 1,4-pentadiene system to their corresponding hydroperoxy derivatives. Atmospheric oxygen serves as second substrate.

disease. Nonetheless, our knowledge of the biological role of various LOX-isoforms, in particular for *ALOX15*, *ALOX15B*, and *ALOX12* is still somewhat limited. This review is aimed at summarizing and critically evaluating the experimental data characterizing the physiological and patho-physiological roles of various LOX-isoforms in mammals. Of course, LOXs have been the topic of previous reviews and a PubMed search with the keywords "lipoxygenase and review" yielded some 1700 hits. However, most of these reviews cover selected areas of LOX research such as LOX enzymology [1], *ALOX5* pathway and leukotriene signaling [2] or LOX in bone disease [14]. To the best of our knowledge there is no recent review paper summarizing the current knowledge of the biological role of mammalian LOX isoforms in health and disease.

During the past decades LOX research has developed rapidly and a PubMed search with the keyword "lipoxygenase" gave some 15,600 hits. Since 2003 about 500 articles have been published annually and because of space limitations it was not possible to reference here even 10% of these reports. Thus, although we tried to make a balanced selection we might have overlooked important articles and we apologize to those distinguished colleagues whose work we have not had sufficient space to reference.

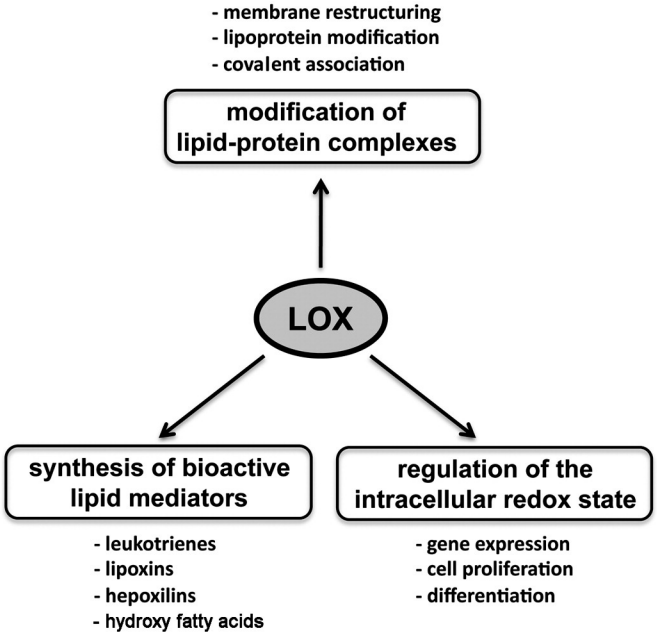

Fig. 2. Biological function of lipoxygenases. Lipoxygenases may exhibit their biological functionality via three different mechanistic scenarios. i) Formation of bioactive lipid mediators, ii) structural modification of complex lipid–protein assemblies and iii) modification of the cellular redox homeostasis, which alters the gene expression pattern.

2. Lipoxygenase distribution, classification and properties

LOXs occur in two (bacteria, eukarya) of the three domains of terrestrial life [1,15] but their occurrence in archaea remains unclear (Fig. 3). The genomic sequences of selected archeae (*Methanococcus voltae*, *Halorubrum kocurii*) also contain LOX-like sequences but in the absence of any functional data it remains unclear if these sequences encode for a functional LOX-isoform. When we performed multiple amino acid alignments of these putative LOX sequences with the primary structure of well-characterized pro- and eukaryotic LOXs we observed only low (<25%) degrees of amino acid conservation. Moreover, we did not find conservation of the iron liganding residues suggesting that the sequences of interest may not encode for functional LOXs. The occurrence of LOX in single cell organisms, in plants and lower metazoa [15–17], has been reviewed before but the distribution of LOX isoforms in multicellular vertebrates has not been summarized systematically.

2.1. Classification of mammalian lipoxygenases and LOX genes

The human genome involves six functional LOX genes (ALOX15, ALOX15B, ALOX12, ALOX12B, ALOXE3, ALOX5), which encode for six different LOX-isoforms [18]. Except for the ALOX5 gene, which was mapped to chromosome 10, all other LOX genes are localized in a joint gene cluster on chromosome 17. The corresponding mouse genes [18] were detected in syntenic regions on chromosomes 6 (Alox5) and 11 (other LOX-isoforms). Originally, the human LOX isoforms were classified with respect to their specificity of arachidonic acid oxygenation but this nomenclature turned out to be misleading and caused confusion among scientists not working in the LOX field [1]. These days the gene nomenclature is frequently employed to define the LOX isoenzyme. Table 1 summarizes human and murine LOX-isoforms and assigns names of the genes to the different isoenzymes. For this review we will use the names of the genes also when we talk about the corresponding enzymes. To differentiate between genes and proteins we employ italic capitalized letters when referring to enzymes but use normal letters when referring to the genes (small letters for mouse and capital letters for human genes).

The ALOX15 gene encodes for the 12/15-LOX, which is expressed at high levels in eosinophils [19], broncho-alveolar epithelial cells [20] and interleukin 4 treated monocytes [21]. The ALOX15B gene encodes for 15-LOX2, which is highly expressed in epithelial cells [22,23]. The ALOX12 gene encodes for the platelet-type 12-LOX (pl12-LOX), which is not only expressed at high levels in blood platelets [24] but also occurs in the skin [25]. The ALOX12B gene [26,27], which encodes for a 12R-lipoxygenating enzyme, and the ALOXE3 gene [28,29] encode for two distinct epidermis-type LOX isoforms, which are co-expressed in the skin. These enzymes have been implicated in epidermal differentiation [30] and appear to be important for the development of the epidermal

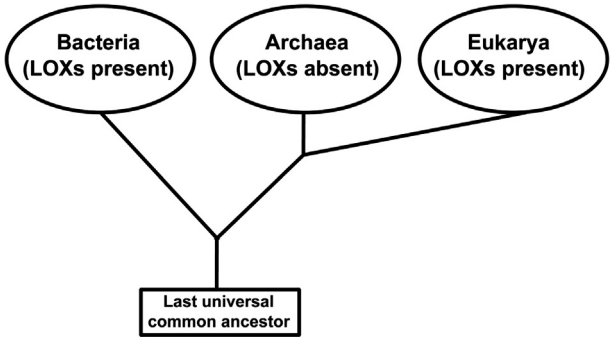

Fig. 3. Distribution of lipoxygenases in the kingdoms of terrestrial life. Lipoxygenase genes have been detected in two (bacteria, eukarya) of the three kingdoms of terrestrial life. Although LOX-like sequences have also been described in archaea, no functional LOX enzyme has been reported to occur.

Please cite this article as: H. Kuhn, et al., Mammalian lipoxygenases and their biological relevance, Biochim. Biophys. Acta (2014), http://dx.doi.org/10.1016/j.bbalip.2014.10.002