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## Methods in Free Radical Biology and Medicine

## An overview of the chemistry and biology of reactive aldehydes



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## ABSTRACT

The nonenzymatic free radical generation of reactive aldehydes is known to contribute to diseases of sustained oxidative stress including rheumatoid arthritis, atherosclerosis, neurodegeneration, and a number of liver diseases. At the same time, the accumulation of lipid electrophiles has been demonstrated to play a role in cell signaling events through modification of proteins critical for cellular homeostasis. Given the broad scope of reactivity profiles and the ability to modify numerous proteomic and genomic processes, new emphasis is being placed on a systems-based analysis of the consequences of electrophilic adduction. This review focuses on the generation and chemical reactivity of lipid-derived aldehydes with a special focus on the homeostatic responses to electrophilic stress.

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## Introduction

Unsaturated fatty acids are abundant constituents sequestered in *sn*-1 and *sn*-2 esterified forms in glycerol phospholipids within cellular membranes. In addition, unsaturated free fatty acids are abundant within various cellular compartments. The fact that lipids constitute a major portion of the plasma and mitochondrial and endoplasmic membranes establishes the presence of massive concentrations of unsaturated fatty acids within membranous structures. Likewise, the concentration of unsaturated fatty acids, in the free form or bound to specific transport proteins, in cells is noteworthy. It is also well recognized that the polyunsaturated fatty acids are bioactive mediators of diverse pathways involved in cellular homeostasis or, in some cases, interact with cellular macromolecules resulting in cell death. These cellular responses may be a consequence of the vulnerability of unsaturated fatty acids to diverse oxidation reactions or radical reactions both of which result in formation of electrophilic lipid products. Certain of the oxidation reactions involving unsaturated fatty acids are enzymatically mediated by families of non-heme-containing metallo-enzymes including the lipoxygenases (LOX), cyclooxygenases (COX), and cytochrome P450. Because these reactions are integral for an organism's response to a range of stimuli, they are generally well controlled and generate a spectrum of bioactive products which are ligands for highly specific, receptor-mediated responses including vasodilation, vasoconstriction, and proinflammatory or anti-inflammatory cascades (for informative comprehensive reviews of the production and actions of enzymatically generated bioactive lipids see the entire volume of *Chem. Rev.* **111**:2011) [1].

The electrophilic lipid products generated by free radical-mediated lipid oxidation are different in many respects than

those generated by enzyme-mediated oxidation. First, free radical-mediated production of electrophilic products of polyunsaturated fatty acids proceeds by autocatalysis and is, as a result, not well regulated. Thus, free radical-mediated lipid peroxidation is more commonly associated with diseases of sustained oxidative stress including rheumatoid arthritis, atherosclerosis, neurodegeneration, and a number of liver diseases. Further, the overproduction of electrophilic lipids resulting from free radical-mediated peroxidation of polyunsaturated fatty acids has been demonstrated to initiate cell death through modification of proteins critical for cellular homeostasis. A series of recent reviews highlight the advances in identification of lipid substrates, mechanisms of lipid peroxidation, and protein targets of electrophilic lipid modification [2,3]. At the same time, there is emerging data demonstrating that, like the lipid products produced by the LOX, COX, and P450-mediated oxidation of unsaturated fatty acids, the electrophilic products of radical-generated lipid peroxidation also initiate responses that are cytoprotective and thus integral for cellular homeostasis and survival. This review will provide an overview of free radical-mediated lipid peroxidation involved in cellular injury followed by a more comprehensive examination of recent studies describing the potential of electrophilic products of lipid peroxidation to modulate signaling pathways involved in antioxidant responses. The final section of this perspective will assess the potential of antioxidant therapy to enhance or abrogate antioxidant responses initiated by electrophilic lipid signaling.

## Generation of reactive aldehydes

The generation of oxygen-derived free radicals and oxidants is a consequence of cellular metabolism and bioenergetic processes

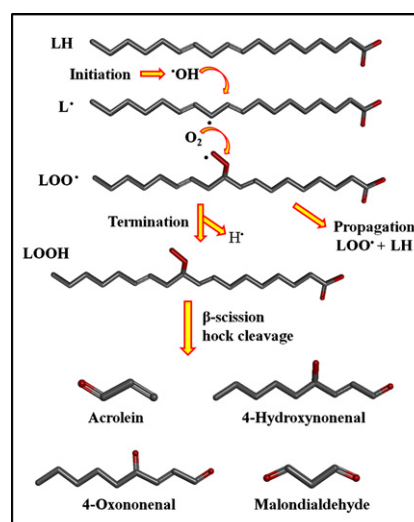
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throughout the animal kingdom [4]. It is evident that a host of biochemical mechanisms mediate persistent free radical generation and damage. An insightful review by Hermann Esterbauer details the integration of oxidative processes in human physiology [5]. In it, he states that the adult human with a daily energy requirement of 10,000 kJ consumes approximately 660 g of oxygen per day, with roughly 90–95% of that oxygen converted to harmless water through mitochondrial respiration. The remaining 5–10% of that oxygen undergoes univalent and divalent reduction, yielding reactive oxygen species, such as superoxide radicals. When taken in the context of the average human life span, the human body consumes a massive 17,000 kg of oxygen which results in the concomitant production of 800–1700 kg of oxygen radicals. These figures provide a fascinating perspective on the detoxifying mechanisms aerobic organisms have integrated into biology and the critical role they play in diffusing oxygen radicals and maintaining life.

The endogenous generation of reactive aldehydes has been studied for decades and is known to contribute to numerous disease pathologies by altering proteomic, genomic, cell signaling, and metabolic processes [6,7]. Reactive aldehydes compose a class of highly reactive organic chemical compounds obtained by oxidation of primary alcohols, characterized by the common group R-CHO consisting of a carbonyl center bonded to hydrogen and an R group [8]. These compounds arise predominantly as a consequence of oxidative stress within the cellular microenvironment, where prooxidant forces overcome natural antioxidant capacities. Numerous studies demonstrate that at low levels, these compounds contribute to regulating cell proliferation among many other processes; however, a delicate balance exists between basal levels of these aldehydes and cytotoxic concentrations [5,6,9,10].

The production of reactive aldehydes occurs through a number of pathways, including enzymatic and nonenzymatic processes. One of the most prominent sources for the generation of reactive aldehydes is through nonenzymatic free radical mechanisms [11]. Specifically, a profusion of reactive nitrogen species (RNS) and reactive oxygen species (ROS), including peroxyxynitrite, superoxide radicals, and hydroxyl radicals, provides an overabundance of initiating chemicals. A major source of these aldehydes is the autoxidation of polyunsaturated fatty acids (PUFA), including arachidonic and linoleic acid. Lipid peroxidation occurs in three major phases, comprised of an initiation event, chain propagation, and termination (Fig. 1) [3]. Typical initiating chemical species are hydroxyl and superoxide radicals. Both are present under normal physiologic conditions and are produced at much higher concentrations in situations of oxidative stress. Initiation occurs through the abstraction of a bis-allylic hydrogen from a lipid chain (LH) to yield a lipid radical ( $\dot{L}$ ). Propagation proceeds when oxygen is added to the carbon-centered radical, where  $\dot{L}$  is rapidly converted to an oxygen-centered peroxy radical ( $LOO\dot{O}$ ). The  $LOO\dot{O}$  reacts with another LH to generate  $\dot{L}$  and an unstable lipid hydroperoxide (LOOH), which in turn yields new peroxy and alkoxy radicals, degrading further to secondary products through  $\beta$ -scission and hock cleavage, among others. A single initiation reaction is postulated to result in 200 to 400 propagation cycles, rapidly amplifying free radical damage under highly oxidizing PUFA-rich environments [12]. Chain-breaking events are known to occur, as  $\alpha$ -tocopherol (vitamin E) radicals form while converting  $LOO\dot{O}$  to LOOH [13]. Termination of the propagation cycle occurs when two free radical species combine to yield nonradical species. The degradation of peroxy and alkoxy radicals into secondary products is thought to generate well over a hundred different reactive species, each with a wide range of reactivity, size, and specificity [5,6]. This overabundance of chemical species greatly increases the complexity of studies attempting to characterize lipid peroxidation-derived electrophilic damage. The



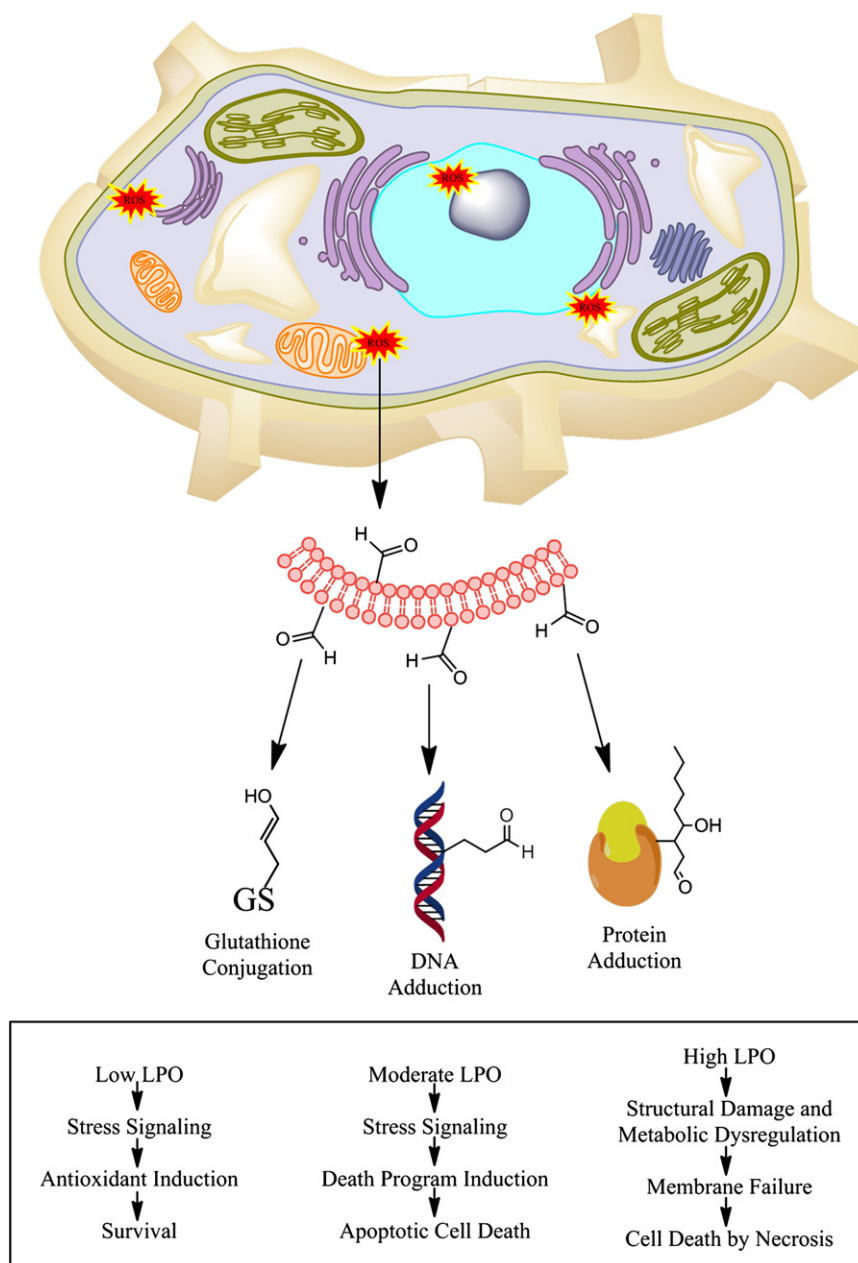
**Fig. 1.** The generation of reactive aldehydes through the initiation, propagation, and termination stages of lipid peroxidation. For simplicity, Fig. 1 shows the generation of reactive aldehydes from a C18 fatty acid; however, in cells, the fatty acids are largely esterified within phospholipids. Furthermore, reactive aldehydes can also occur in esterified form, which may also form adducts with proteins and other macromolecules, although this is a less well-studied area.

more prominent products 4-hydroxynonenal (4-HNE), malondialdehyde (MDA), and acrolein (ACR) have been examined under various disease models through *in vitro* assays, immunohistochemical staining, and analytical HPLC methods [7,14,15].

Cellular location is an important consideration in determining the extent of free radical-mediated damage resulting in the generation of reactive aldehyde species. As illustrated in Fig. 2, lipid-rich membranes provide an optimal environment for producing a high abundance of these cytotoxic compounds. Microsomal and mitochondrial intramembrane concentrations of these lipid peroxidation-derived electrophilic compounds have been estimated to accumulate to as high as 10 mM *in vivo* [9,16]. The regulatory function and cytotoxicity of these aldehydes hinges on abundance, reactivity, and longevity. Longevity varies greatly, with compounds such as 4-oxononenal (4-ONE) and 4-HNE displaying half-lives of roughly 1 s and 2 min, respectively [9,17]. Persisting on a scale of minutes, some aldehydes preferentially retain the ability to transiently modify distant proteins, membranes, and DNA. These factors may contribute to a localized bias for molecular adduction by reactive aldehydes, particularly those with short half-lives, as nucleophiles located within the vicinity of lipid peroxidation remain more susceptible to higher concentrations of electrophiles. However, long-lived species may have a more pronounced global impact by altering a broader array of proteomic and genomic targets due to their transient nature.

### Chemical mechanisms

The chemical specificity of reactive aldehydes is impacted through direct and indirect forces. Direct factors include characteristics of the electrophile and nucleophile species, such as  $pK_a$ , chemical potential, and electrophilic index. Indirect forces include solvent pH and adjacent peptide sequence, which can play an important role through steric hindrance and charge stabilization. Both factors play an integrated role in determining the specificity for any given set of electrophile–nucleophile pairs. To better understand the stability and reactivity of these adducts, a number



**Fig. 2.** Cellular effects of oxidative stress and lipid peroxidation. The nonenzymatic free radical generation of reactive aldehydes by lipid peroxidation occurs near lipid-rich cellular membranes and produces both membrane-bound and free electrophiles. These electrophilic species go on to modify proteomic and genomic processes. Antioxidant responses such as glutathione conjugation play a role in defending against the damaging effects of these reactive aldehydes. (LPO, lipid peroxidation.).

of researchers have employed computational models coupled to *in vitro* studies [18–22]. Importantly, these studies provide key mechanistic insights into the reactivity and specificity of reactive aldehydes toward cellular targets. A detailed analysis of the selectivity of electrophile–nucleophile interactions applying the hard, soft, acids, and bases (HSAB) theory is covered extensively in a recent perspective [23]. Basically, electrophiles and nucleophiles are classified as either soft (polarizable) or hard (nonpolarizable). Based on these parameters, among others, the specificity of an electrophile for any given nucleophile is determined with a certain degree of confidence. Therefore, these factors can be applied to chemical toxicology to aid in the identification of sites of modification and mechanisms of toxicant action. Further, through the application of quantum mechanical parameters to *in vitro* experiments, it was confirmed that soft electrophiles bind

more favorably with soft nucleophiles and hard electrophiles bind more favorably with hard nucleophiles [23,24]. An *in vitro* study using purified human glyceraldehyde-3-phosphate dehydrogenase (GAPDH) intricately demonstrates that a weak electrophile (acrylamide) was able to discriminate among highly reactive cysteines from less reactive residues [19]. Furthermore, a quantitative relationship was determined correlating adduct formation and protein dysfunction. Certainly, *in vivo* and *in vitro* model systems contain highly complex matrices of biomolecules which may confound the predictive nature of these experiments employing purified protein and theoretical computational models. However, these examples provide support for the combined application of computational analysis with physiologically applied models to better understand the chemical reactivity of these reactive aldehydes.

The covalent modification of biomolecules by reactive aldehydes is a well-documented phenomenon [25,26]; however, underlying chemical mechanisms of specificity and reactivity remain somewhat unclear. Basic mechanisms of aldehyde modification include Michael addition and Schiff base reactions with the thiol group of cysteine and secondary amines of lysine, histidine, and arginine [3,27]. DNA adducts arise through direct base adduction, for example, the Michael addition of deoxyguanosine by 4-HNE [21]. In general, these electrophilic adducts are thought to generate an array of stable, covalent modifications, yet numerous reports present a contrasting account of adduct stability [20,28–30]. Instability among these electrophilic adducts provides a likely explanation for the lack of reported *in vivo* adducts by mass spectrometry techniques. While continued research is providing an ever-expanding list of proteins modified *in vivo* by lipid electrophiles like 4-HNE [25,31], the modified amino acid is rarely identified. To circumvent potential instability of adducts, sodium borohydride treatment is used to stabilize these adducts through reduction, increasing the chances for identifying modified proteins [32]. Recently, a number of novel methods have been developed to capture these carbonylated proteins and provide a large toolset for identifying and characterizing these PTMs [33–36]. These strategies were recently reviewed and include a click chemistry approach as well as those based primarily on hydrazide reactivity with protein carbonyls, such as biotin hydrazide, aldehyde reactive probes, and Girard's P reagent [3].

The majority of research studying chemical mechanisms of aldehyde adduction has occurred under controlled experimental conditions. In these studies, protein targets previously identified through Western blotting or immunoprecipitation techniques are treated with known quantities of aldehyde in a test tube [18,37–39]. While these studies have revealed important information relating to amino acid targets and kinetic parameters for these reactive aldehydes, little insight is gained in establishing a substantial connection between the electrophilic modification of a specific protein and a related physiologic response. Importantly, a number of studies have demonstrated that these electrophilic adducts may be used as biomarkers for a range of disease pathologies; however, what role these adducts play in the initiation and/or progression of these diseases remains unclear [7]. Only recently has a global analysis of protein damage by a lipid electrophile been accomplished [25]. In this study, human colon carcinoma RKO cells were treated with increasing concentrations of 4-HNE followed by the isolation of modified proteins for MS/MS analysis. Proteomic surveys of protein carbonylation typically generate large lists of proteins (~500), where highly abundant proteins are found modified with greater frequency, which leads to the question of 4-HNE specificity versus concentration-type interactions. An important consideration when examining these adducts is whether they form through concentration-dependent mechanisms. It is anticipated that true protein targets of 4-HNE display an electrophile concentration-dependent relationship. As expected, this study confirmed previous conclusions regarding pseudo-first-order kinetics of 4-HNE protein adduction [40]. A key analysis is to examine if protein damage reflects specificity for cell components and processes. To address this particular issue, Codreanu et al. performed a networks and communities analysis of modified proteins [25]. By mapping the identified 4-HNE-adducted proteins to a human protein interaction network they were able to provide systems-level insight into the cellular impact of 4-HNE adduction. Importantly, the authors were able to utilize protein clustering coefficients to characterize protein communities targeted by 4-HNE and elucidate cellular processes that may be impacted.

Representing a paradigm shift in studying cellular responses to electrophiles, Jacobs and Marnett provide a novel approach to define the chemistry of protein modification using lipid electrophiles as a

model system [33]. Employing a 4-HNE-derived click chemistry treatment to RKO cells, the authors identified 4-HNE-activated transcriptional pathways through microarray, real-time PCR, Western blot, and luciferase reporter assays. Their global approach incorporated data analyses for electrophile–protein modification as well as alterations in gene expression. Informatics tools provided a theoretical connection by linking electrophile-induced proteomic and genomic effects through related transcription factors. Interestingly, pathways that were found to be activated by 4-HNE treatment include DNA damage, antioxidant, ER stress, and heat shock responses. This strategy correlates proteome-wide electrophile modification to cellular responses, bridging our understanding of the chemical reactivity of aldehydes to human health and disease. Indeed, electrophile-induced protein modification and downstream effects reveal an extremely complex cascade of cellular responses to oxidative stress and lipid peroxidation in particular.

### Homeostatic responses to electrophilic stress

The proposition that 4-HNE and other lipid electrophiles produced from the radical oxidation of lipids mediate cellular toxicity through adduction of proteins has been investigated extensively and is the subject of many recent reviews. However, investigators have observed that cellular responses are divergent depending on the dose or concentration of 4-HNE to which the cells are exposed. For instance, in the classic review presented by Esterbauer et al. [9] it is noted that cells exposed to subcytotoxic concentrations (0.5–2  $\mu\text{M}$ ) of 4-HNE displayed activation of phospholipase c activity or, in the case of smooth muscle vascular cells, proliferation. These observations were followed by a report [41] that subcytotoxic concentrations of 4-HNE increase expression of the glutathione S-transferases (GST) in a hepatoma cell line through activation of the antioxidant response element (ARE). Because GST-mediated conjugation of 4-HNE with glutathione (GSH) is a major pathway for detoxification of this lipid electrophile, the results of this study suggested that the exposure of cells to subcytotoxic concentrations of 4-HNE results in induction of pathways which facilitate its elimination. This notion was subsequently confirmed by a report that exposure of HBE1 cells to physiologically relevant concentrations of 4-HNE resulted in induction of GSH synthesis through induction of glutamate cysteine ligase (GCL) which is the rate-limiting step in glutathione synthesis [42]. The fact that GCL induction was attenuated by a JNK inhibitor demonstrated that 4-HNE signaled this response through the JNK pathway. Taken together, these early studies suggested that when present at low concentrations, 4-HNE has the ability to activate a spectrum of cytoprotective signaling cascades in order to maintain cellular homeostasis.

There is general agreement that the cellular accumulation of 4-HNE leads to apoptosis and the specific mechanisms by which 4-HNE modulates signaling stress-induced programmed cell death have received considerable attention and detailed in a recent review [43]. The involvement of 4-HNE in modulation of programmed cell death is multifactorial in that this lipid electrophile modulates signaling pathways involved in extrinsic and intrinsic pathways of apoptosis. In terms of the extrinsic pathway, reports have appeared describing the potential of 4-HNE to initiate binding of the death-associated protein Daxx with the intracellular surface of the Fas receptor [44–46]. Once Daxx binding to Fas occurs, apoptosis is modulated through the downstream signaling proteins, ASK1, JNK, and caspase 3. Interestingly, the silencing of Daxx results in increased sensitivity of Jurkat cells to 4-HNE-induced apoptosis, suggesting that this lipid electrophile, when present at low concentrations, can play a

self-regulatory or self-limiting role in apoptosis [46]. Activation of the intrinsic pathway of apoptosis involves the p53 tumor-suppressor protein [47]. The ability of 4-HNE generated by oxidative stress to initiate apoptosis has been attributed to the ability of this lipid electrophile to increase p53 expression followed by activation of p21, Bax, JNK, and caspase 3 [45,48]. The involvement of 4-HNE in apoptosis through extrinsic and intrinsic pathways is interesting in terms of self-regulating the extent of programmed cell death in response to oxidative stress through a common downstream signaling protein JNK. As this phenomenon continues to be investigated, it will be important to determine if programmed cell death by 4-HNE is regulated through the same mechanisms in all cell types.

The concept that cellular responses to 4-HNE and oxidant stress are supported by a decade old study documenting modulation of 35 genes in human retinal pigment epithelial cells exposed to 4-HNE or the oxidants H<sub>2</sub>O<sub>2</sub> or *tert*-butylhydroperoxide [49]. The mechanisms of gene modulation by 4-HNE were revealed in a more recent comprehensive report of gene profiling expression in RKO cells exposed to subcytotoxic concentrations of 4-HNE for 6 or 24 h [50]. Among the prominent responsive genes identified by these investigators were those involved in oxidative damage or ER stress. Of particular interest was the experimental approach which facilitated the observation that gene responsiveness was mechanistically linked to signaling responses associated with addiction of relevant proteins.

As noted earlier in this review, protein thiols are preferential targets for modification by 4-HNE. It is not surprising that the transcription factor Keap-1-Nrf2, a chief regulator of cellular antioxidant responses through redox-sensitive cysteine residues, is a sensitive target protein for 4-HNE adduction, resulting in activation of cellular antioxidant systems. The specific mechanisms of 4-HNE-Keap-1-Nrf2 interactions and cellular responses are detailed in a recent review [51]. Therefore, we will present only an overview of reports describing the interaction of 4-HNE with the thiol-sensitive systems involving Keap-1-Nrf-2 which, in turn, impact protective cellular responses to stress.

As detailed in two recent reviews [51,52] the involvement of the Keap-1-Nrf2 complex in antioxidant responses, through activation of antioxidant-response elements (AREs), has received considerable attention. The mechanistic aspects of this system are intriguing in that electrophiles such as 4-HNE target cysteine thiols in Keap-1, resulting in release of Nrf2 which translocates to the nucleus where it dimerizes with nuclear factors forming a complex activating expression of ARE genes for the phase II antioxidant proteins NADH-dependent quinone reductase, heme oxygenase-1, glutathione-S-transferases, and glutamate-cysteine ligase. The release of Nrf2 from Keap-1 is mediated by modification of specific thiol sensors in Keap-1 by electrophiles such as 4-HNE. Although Keap-1 contains multiple thiols, those at locations C151 and C288 appear to be specific for 4-HNE and other alkenals [53]. Interestingly, these specific cysteine residues are separate from those targeted by other stress-associated endogenous agents such as nitric oxide or Zn<sup>+</sup>, suggesting a high degree of specificity for the signaling capabilities of Keap-1 to endogenous signals.

The thiol sensitivity of the nuclear factor kappa B (NFκB) system was first proposed in a study describing what appeared to be inhibition of its activation in RKO cells and H1299 cells exposed to 4-HNE [54]. NFκB is a transcription factor commonly associated with proinflammatory signaling and is regulated by binding of the inhibitor kappa B (IκBα) protein. Thus, under homeostatic cellular conditions, IκBα binds to NFκB sequestering it in an inactive form. Under conditions of cell stress, the enzyme IκB kinase phosphorylates IκBα facilitating its dissociation from NFκB allowing translocation to the nucleus for transcription of

Bcl-2 family members which mediate antiapoptotic effects. Similarly, the expression of proinflammatory cytokines such as IL-6 is also mediated by NFκB. Interestingly, systematic evaluation of the NFκB inhibition revealed that 4-HNE inhibited IκB kinase (IKK), thus compromising IκBα degradation [54]. The sensitivity of IKK to 4-HNE inhibition is marked, as the studies with kupffer cells, which are the resident macrophage in liver, have demonstrated that inhibition of IL-6 release is severely impaired by submicromolar concentrations of 4-HNE [55]. Thus, 4-HNE produced during controlled lipid peroxidation, such as that occurring during homeostatic cell metabolism, may play an important role in regulating inflammatory responses. Taken together, the effects of 4-HNE on activation of phase II antioxidant enzymes through Keap-1 and repression of proinflammatory cytokine release by inhibition of IKK suggest that basal concentrations of this and other lipid electrophiles may play very important homeostatic roles in a variety of cell types.

### Considerations of pharmacologic strategies for mitigating lipoxidation-derived reactive aldehydes

There is substantial evidence that the overproduction of electrophilic lipid aldehydes such as 4-HNE results in protein carbonylation associated with a spectrum of disorders including diabetes, neurodegenerative diseases, and liver disease. Therefore, it is logical that a number of strategies have been evaluated to arrest oxidative stress through the use of antioxidants or attempts to chemically trap the reactive lipid electrophiles. To date, there is little evidence that prophylactic antioxidant therapy is effective in preventing or slowing progression of diseases involving oxidative stress. The approach of using specific agents to trap reactive aldehydes is based on the chemical properties of the thiol- and histidine-containing compounds that form Michael adducts with the lipid electrophiles. The agents evaluated to date are described in detail in a comprehensive review [56]. Briefly, one strategy includes utilizing endogenous pathways of detoxification such as spontaneous and GST-catalyzed glutathionylation, where irrigation with γ-glutamyl-cysteine may increase GSH levels to normalize pathophysiological 4-HNE concentrations to preinsult levels. Other interventions include ascorbate (vitamin C) and carnosine (β-alanyl-L-histidine), each used as a quencher of cytotoxic carbonyl species via Michael addition [7,56]. Whereas these compounds are effective in trapping reactive aldehydes in isolated cells subjected to significant oxidative stress, their efficacy in animal models or humans remains to be established. As with any potential therapeutic agent, a thorough assessment must be performed to identify off-target effects and resulting toxicities. This is complicated by the potential role of 4-HNE in regulating homeostatic responses to oxidative stress through ARE activation [53] or inhibition of the release of proinflammatory cytokines through modulation of NFκB [54,55]. Likewise, especially challenging is the selectivity of targeting specific protein thiols, such as those identified in keap-1, which selectively interact with 4-HNE rather than Zn<sup>+</sup> or nitric oxide [53]. Clearly, caution is needed when prophylactic treatment with antioxidant therapies such as vitamin E supplementation is employed. One must assess the overall impact to signaling cascades as well as oxidative stress using a system-wide approach.

### Conclusions

The generation of reactive aldehydes through nonenzymatic free radical mechanisms is a complex process resulting in the formation of over a hundred different species of electrophilic

compounds containing a broad spectrum of reactivity profiles. Given that oxidative stress is integrated throughout the cell, most notably through cellular respiration in the mitochondria, it is not surprising that multiple antioxidant responses exist. Indeed, a systems biology approach investigating both protective and deleterious effects of reactive aldehydes is essential to understanding the chemico-biological nature of aldehyde-regulated cellular homeostasis. While reactive aldehydes are known to alter proteomic, lipidomic, and genomic processes, very little is known about the role that these compounds play in the initiation and progression of disease pathologies. Further studies are required to elucidate the physiological impact these electrophiles have on biological processes.

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