Free Radical Biology and Medicine 111 (2017) 196–208

Contents lists available at ScienceDirect

Free Radical Biology and Medicine

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

4-Hydroxynonenal metabolites and adducts in pre-carcinogenic conditions and cancer

Françoise Guéraud

Toxalim (Research Centre in Food Toxicology), Université de Toulouse, INRA, ENVT, INP-Purpan, UPS, Toulouse, France

ARTICLE INFO

Keywords:
4-hydroxynonenal
Lipid peroxidation
Oxidative stress
Cancer
Genotoxicity
Etheno-adducts
Propano-adducts
Electrophilic
Nrf2
Keap1
GST
Mercapturate
Heme iron
Colorectal cancer

ABSTRACT

4-hydroxy-2-nonenal (HNE) is an amazing reactive compound, originating from lipid peroxidation within cells but also in food and considered as a "second messenger" of oxidative stress. Due to its chemical features, HNE is able to make covalent links with DNA, proteins and lipids. The aim of this review is to give a comprehensive summary of the chemical properties of HNE and of the consequences of its reactivity in relation to cancer development. The formation of exocyclic etheno-and propano-adducts and genotoxic effects are addressed. The adduction to cellular proteins and the repercussions on the regulation of cell signaling pathways involved in cancer development are reviewed, notably on the Nrf2/Keap1/ARE pathway. The metabolic pathways leading to the inactivation/elimination or, on the contrary, to the bioactivation of HNE are considered. A special focus is given on the link between HNE and colorectal cancer development, due to its occurrence in foodstuffs and in the digestive lumen, during digestion.

1. Introduction

Cancer, which can be defined as an uncontrolled proliferation of cells, is a multi-step and multi-factorial disease. After an initial genotoxic event, leading to a mutation of a critical gene in a cell that will proliferate and give a clone of mutated cells, the development of cancer requires the accumulation over periods of many years of abnormalities and alterations related to the regulation of cell proliferation, survival and differentiation.

Reactive oxygen species (ROS) are involved in many of those cancer-related cellular processes, such as proliferation and apoptosis, differentiation, cell migration and DNA damage. However, as ROS have a very short half-life, their effects remain localized to the very near vicinity of their production. However, the attack of cellular polyunsaturated acids by ROS gives rise to a series of secondary lipid oxidation products, among which reactive aldehydes, such as 2-alkenals seem to be the most abundant and the most reactive. Those cytotoxic and genotoxic compounds have a much longer half-life than ROS and can diffuse to other cell compartments, to other cells and probably to other tissues. For this reason, lipid peroxidation (LPO), as a consequence of oxidative stress, plays an important role in health and disease. The reactive secondary lipids oxidation products seem to be at least partly responsible for the biological/toxic effects of lipid peroxidation and has been often qualified as "second toxic messengers" of oxidative stress [1].

4-hydroxy-2-nonenal (HNE), one of the most abundant and reactive aldehydic compounds formed upon lipid peroxidation of rat liver microsomes [2] has been discovered in the early 1960s by Esterbauer and his colleagues in Graz. Since then, it has been widely studied, not only as a toxic compound but also as an actor of cellular regulations,

Abbreviations: ABC transporters, ATP-binding cassette transporters; AhR, aryl hydrocarbon receptor; AKR, aldo-keto reductase; Akt, protein kinase B; ALDH, aldehyde dehydrogenase; AOR, alkenal/one oxidoreductase; AP-1, activator Protein-1; AR, aldose reductase; ARE/EpRE, antioxidant response element/electrophilic response element; BER, base excision repair; CRC, colorectal cancer; COX-2, cyclooxygenase 2; Daxx, death domain-associated protein; DHN, 1,4-di-hydroxy-nonene; DHN-MA, DHN-mercapturic acid; EGCG, epigallocatechin-3-gallate; FMO, flavin monooxygenase; EGFR, epidermal growth factor receptor; GSH, glutathione (reduced); GS-HNE, glutathionyl-HNE; GST, glutathione *S*-transferase; HNA, 4-hydroxy-2-nonenoic acid; HNE, 4-hydroxy-2-nonenal; HNE-sat, 4-hydroxynonanal; HO-1, heme oxygenase-1; iNOS, inductible NO synthase; JNK, c-Jun N-terminal kinases; Keap1, Kelch-like ECH-associated protein 1; LEC rats, Long Evans Cinnamon rats; LPO, lipid peroxidation; MAP kinase, mitogen-activated protein kinase; MDA, malondialdehyde; MRP, multidrug resistance associated proteins; NER, nucleotide excision repair; NQO1, NAD(P)H:quinone oxidoreductase 1; Nrf2, Nuclear factor E2-related factor 2; PKC, protein kinase C; PDGFRβ, β-type platelet-derived growth factor receptor; Pin-1, peptidyl-prolyl cis/trans-isomerase A1; SCE, sister chromatid exchange; TBARS, thiobarbituric acid reactive substances; TBHQ, *tert*-butylhydroquinone; UDP, uridine diphosphate; UGT, UDP-glucuronosyl transferase

E-mail address: Francoise.Gueraud@inra.fr.

http://dx.doi.org/10.1016/j.freeradbiomed.2016.12.025
Received 2 November 2016; Received in revised form 14 December 2016; Accepted 17 December 2016
Available online 06 January 2017

even in a physiological state. This compound and its metabolites, including its adducts with cellular biomolecules, has also served as biomarkers of lipid peroxidation, under various conditions of oxidative stress/lipid peroxidation/inflammation related diseases.

The aim of this review is to give a general overview of the relation existing between HNE and its metabolites and the development of cancer, as a participant and/or a biomarker of the process. Other LPO derived 2-alkenals, and particularly other hydroxyl- or oxo-alkenals, will share the same properties due to similar chemical functions. A special focus is done on cancers of the digestive tract, especially on colorectal cancer, because HNE comes also from the peroxidation of dietary polyunsaturated fatty acids, in food and probably during digestion. Digestive tract cells are then the first targets of this dietary HNE.

2. Structure/activity of alkenals

HNE is an amphiphilic 4-hydroxy-2-alkenal formed upon peroxidation of polyunsaturated fatty acids of the omega-6 family, especially linoleic and arachidonic acids [1]. It is found in diverse tissues, organs and fluids at various concentrations, depending on the tissue and on the pathophysiological state. In human plasma, the mean value was 0.074 μM [3], while 0.33 nmol/g were found in rat liver [1]. However, as HNE is formed within membranes upon peroxidation of phospholipid bound fatty acids, much higher concentrations were reported locally in those membranes, reaching 5 mM under conditions of oxidative stress [4].

2-Alkenals (Fig. 1) represent a class of compounds with special reactivity due to the carbon to carbon double bond that is conjugated to an electron withdrawing group, namely the carbonyl group on carbon 1. This structure renders those molecules electrophilic, with the carbon 3 as the electrophilic center of the molecule. As a consequence, those compounds, considered as “soft electrophiles”, are prone to nucleophilic attack by cellular nucleophilic groups present in protein, DNA and lipids [1,4,5], but also with the tri-peptide glutathione, a major cellular anti-oxidant compound (see part D), *via* its cysteine moiety. It is noteworthy that the electron withdrawing hydroxyl group on carbon 4 in 4-hydroxy-2-alkenals such as HNE further exacerbates the electrophilicity of the carbon 3 in these compounds [4]. This Michael addition on carbon 3 seems to be a stable link, although an excess of thiol (as glutathione or cysteine) can reverse it [1]. The amino acids involved in this addition are, by decreasing order of reactivity, cysteine in its thiolate form, the imidazole group of histidine and the ε-amino group of lysine [4,6]. In the case of 4-hydroxy-2-alkenals, a cyclisation as a five-membered hemiacetal occurs (Fig. 2).

The aldehyde function is also reactive and can be either oxidized or reduced by biotransformation enzymes, leading to an alcohol or a carboxylic acid, which electrophilicity is reduced compared to the parent aldehyde. A further Michael addition to glutathione or to cysteine/histidine/lysine residue in protein is then unlikely to occur. The aldehyde function can bind covalently to primary amines of amino acid residues in proteins, forming a Schiff base (Fig. 2). This covalent link is easily reversed under mild acidic conditions [1]. However, the Schiff base resulting from the adduction of HNE to lysine gives a very stable pyrrole compound following cyclisation [4]. As lysine can react with HNE on both carbon 3 *via* Michael addition and carbon 1 *via* Schiff base formation, protein cross-linking might occur. Subsequent chemical rearrangements can give HNE derived lipofuscin-like fluorophores.

Due to their high reactivity, 4-hydroxy-alkenals can make exocyclic adducts with DNA (Fig. 3). Two kinds of those adducts were reported: propano-adducts that bear a 6-membered additional saturated ring and etheno-adducts with a 5-membered additional one. Those adducts were formed *in vitro* when nucleosides or DNA were exposed to HNE [7–9].

Propano-type adducts come from the direct Michael addition of HNE to deoxyguanosine, followed by Schiff base formation and ring closure, giving HNE-dGuo (substituted 1, N^2-propanodeoxyguanosine). HNE may also interact directly with other DNA bases. The reversibility of the ring closure may give rise to inter- and intra-DNA and to DNA-proteins cross-links [10–12]. Propano-adducts of HNE have been found in human and rodent tissues [13,14], and their levels dramatically increased upon glutathione depletion in the liver of rats [15,16]. Propano-adducts of HNE were shown to be increased in various tissues

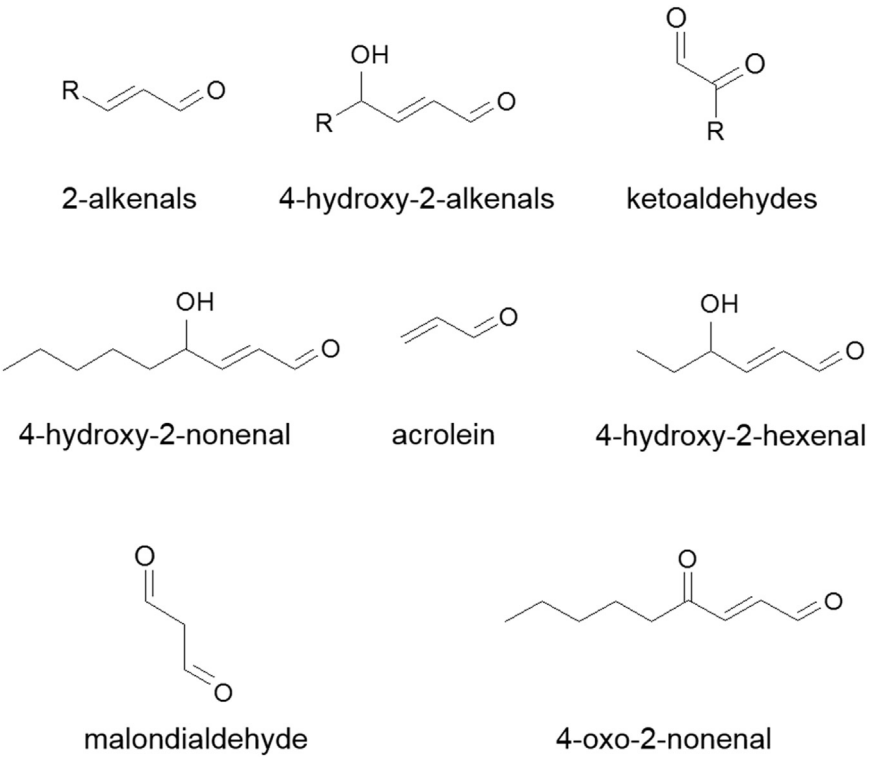

Fig. 1. Chemical structure of lipid oxidation derived reactive alkenals.