HNE (4-hydroxynonenal) Metabolism & Its Age Dependency

HNE (4-hidroksinonenal) Metabolizması ve Yaşlanmayla İlişkisi

Werner SIEMS,[a]
P. VOSS,[b]
Nikolaus BRESGEN,[c]
Peter ECKL,[c]
Tilman GRUNE[b]

[a]KortexMed Inst.
of Medical Education,Research Institute
of Physiotherapy and Gerontology,
Hindenburgring 12A,
D-38667 Bad Harzburg, GERMANY,
[b]Institute of Biological Chemistry and
Nutrition,
University Hohenheim, Garbenst
28, D-70593 Stuttgart, GERMANY,
[c]Department of Cell Biology,
University Salzburg, Hellbrunnerstr.
34, A-5020 Salzburg, AUSTRIA

Yazışma Adresi/*Correspondence:*
Werner SIEMS
KortexMed Inst.
of Medical Education,Research Institute
of Physiotherapy and Gerontology,
Hindenburgring 12A,
D-38667 Bad Harzburg, GERMANY,
siems@kortexmed.de

ABSTRACT Mammalian cells possess highly active pathways of aldehyde including 4-hydroxynonenal (HNE) metabolism. As primary HNE products in many cell types the HNE-glutathione conjugate (HNE-GSH), the hydroxynonenoic acid (HNA) and the corresponding alcohol – 1,4-dihydroxynonene (DHN) were identified. The very rapid HNE metabolism underlines the role of HNE degrading pathways as important part of the secondary antioxidative defense mechanisms in order to protect proteins from modification by oxidative stress products.

From blood plasma and various tissues of human beings and further mammalian species it is known, that HNE levels increase with increasing age. It is not clarified if that is due to whether accelerated formation of HNE or to diminished metabolism of this compound. In the model of human skin fibroblasts from donors of different age the age dependency of HNE degradation rate was analyzed. It was found, that the overall HNE degradation rate strongly decreases with age. Both the formation of HNE-GSH (due to reducing intracellular GSH) and of HNA are reduced with increasing age. It is concluded, that the drastically diminishing HNE metabolism contributes strongly to increasing HNE levels with age.

Key Words: HNE Metabolism, 4-hydroxynonenal, Fibroblasts, Aging, Lipid peroxidation

ÖZET Memeli hücreleri, 4-hidroksinonenal (HNE) metabolizması dahil olmak üzere yüksek derecede aktif aldehid yollarına sahiptirler. Çoğu hücre türündeki birincil HNE ürünü olarak, HNE-glutation konjugat (HNE-GSH), hidroksinonenoik asit (HNA) ve ilgili alkol– 1,4-dihidroksinonen (DHN) belirlenmiştir. Çok hızlı HNE metabolizması, proteinleri oksidatif stres ürünleri ile modifikasyondan korumak için ikincil antioksidatif savunma mekanizmalarının önemli bir parçası olarak HNE degrade edici yolların rolünü ortaya koyar.

Plazmadan, çeşitli insan dokularından ve diğer memeli türlerinden, HNE seviyelerinin artan yaş ile birlikte yükseldiği bilinmektedir. Bunun sebebinin HNE oluşumunun hızlanması mı yoksa bu bileşenin azalan metabolizması mı olduğu bilinmiyor. Farklı yaştaki donörlerden alınan insan cildi fibroblastlarından, HNE degradasyon oranının yaşa bağımlılığı analiz edildi. Genel HNE degradasyon oranının yaş ile birlikte yüksek oranda düştüğü bulundu. Hem HNE-GSH (azalan hücreler arası GSH nedeniyle) ve HNA oluşumu artan yaş ile birlikte düşüşe geçmektedir. Büyük ölçüde zayıflayan HNE metabolizması yaş ile birlikte artan HNE seviyelerine büyük katkı sağlar.

Anahtar Kelimeler: HNE Metabolism, 4-hdroksinonenal, Fibroblast, Yaşlanma, Lipid peroksidasyonu

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HNE (4-HYDROXYNONENAL) LEVEL AS BIOMARKER OF OXIDATIVE STRESS AND AGING

The levels of lipid peroxidation (LPO) products such as MDA (malondialdehyde) and HNE (4-hydroxynonenal) increase in human serum with increasing age. That age-dependency is also true for MDA, HNE, and also for protein carbonyl concentrations in different cells and tissues. The MDA and HNE accumulation in serum correlates with a decrease of GSH and an increase of disulfide in red blood cells.[1-4] The accumulation of LPO products such as HNE may be due to an increased formation rate and/or to a decreased rate of the removal of LPO products.

THE PATHWAYS OF HNE METABOLISM

HNE is rapidly metabolized in eucariotic cells.[5,6] The velocity and the pattern of HNE metabolism were studied in various cell types, furthermore in subcellular organelles, and even in whole organisms. HNE metabolism was studied in hepatocytes, hepatoma cells, ascites tumor cells, mucosal cells, synovial fibroblasts, thymocytes, in vascular smooth muscle cells and also in organs such as heart and kidney.[7-25] The rapid HNE degradation and intracellular metabolism implicates that HNE enters rapidly the cells. The velocity of HNE entry into the cells was tracerkinetically observed in various cell types.[17]

In all studies the overall metabolic rate of exogenously added HNE was so high, that already within a few minutes or even faster equilibrium concentrations in the nanomolar range were obtained. From these experiments one can also conclude, that HNE even at very high lipid peroxidation rates never can accumulate to higher and higher levels. The highest level which we ever measured was 6.5 μM.[26] That was analyzed in experiments with rat intestine during postischemic reperfusion period. In all experimental models which were investigated a multitude of HNE metabolites was identified and quantified. However, one may separate these metabolites into primary and secondary intermediates. The primary metabolites – HNE-GSH, HNA and DHN which are formed by the main enzymes involved in HNE metabolism: glutathione transferases, aldehyde dehydro-genases, and alcohol dehydrogenases - are undergoing further metabolic conversion leading to secondary intermediates.5,6 Some of the secondary intermediates are stable end products. Possibly one may use beside the HNE itself stable HNE products – such as mercapturic acids - as biomarkers for oxidative stress and aging, too.

HNE METABOLISM AS AN IMPORTANT COMPONENT OF ANTIOXIDATIVE DEFENSE

The catabolism of HNE is a very important part of the antioxidative defense system of cells and organisms.[5,6] This is due to the fact, that HNE as other aldehydic products of LPO is able to exert cytotoxic, mutagenic, signal, and cancerogenic effects. If HNE can be degraded to less toxic intermediates, that diminishes the reactions between HNE and biomolecules and, therefore, effectively contributes to the antioxidative protection of cells and organisms. This protective effect is more effective as more rapid the metabolism of HNE leads to stable HNE products, which can be easily excreted.[5,6,22-25]

As more slow the metabolic pathways of HNE are as more HNE reacts directly with biomolecules such as proteins and peptides leading to functional deterioration up to functional loss of these biomolecules. The primary importance of HNE-degrading pathways as one important part of the antioxidative defense system, which functions also at physiological and pathophysiological HNE levels, seems to be the protection of proteins from modification by aldehydic LPO products. That would be valid especially in regions with high HNE formation rate such as in postischemic or reperfused tissues or in synovial tissue of joints during rheumatoid arthritis and under other inflammatory conditions.[5,6,27-37]

HNE METABOLISM IN FIBROBLAST

The velocity of HNE metabolism in primary synovial fibroblasts was high, in one range with that in hepatocytes, thymocytes etc.[10,17,18] The oxidative pathways of HNE metabolism were markedly higher than the reductive pathways. HNA and secondary products of HNA such as members of citric acid cycle and water – formed mainly in the mitochondria - and GSH-HNE conjugate were the quantitatively leading products in HNE degradation of fibroblasts. Similar data on the pattern of HNE metabolites were obtained with skin fibroblasts.

AGE-DEPENDENT DECREASE OF HNE DEGRADATION RATE IN HUMAN SKIN FIBROBLASTS

The HNE degradation rate was measured in human skin fibroblasts of donors of different age (between 20 and 90 years). Additionally, the rate of oxidative pathways was quantified by HNA measurement. Furthermore, glutathione was analyzed to establish GSH consumption for the formation of HNE-GSH.

The most important results which were obtained were: In skin fibroblasts the HNE degradation rate strongly decreases with increasing age of the cell donors. In fibroblasts the reduction of the HNE metabolism is due at least to two factors: one factor is the lowering glutathione concentration with age leading to a decreased capacity for the formation of HNE-GSH conjugate, a major primary HNE intermediate. A second factor is the decreased formation of HNA as another major primary HNE metabolite.

The decrease of overall HNE degradation rate in fibroblasts from donors with increasing age is an indicator for a reduction of the metabolism of aldehydic LPO products such as HNE during aging.

The findings confirm the assumption that increases in LPO product levels during aging are due rather to lowering rates of metabolism of LPO products than to increased oxidative stress including lipid oxidation itself. Hormetic pro-aging protection, therefore, could be directed to improved secondary antioxidative enzymatic mechanisms such as HNE metabolism rather than to direct antioxidative protection against LPO.

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