DOI 10.1515/pterid-2013-0004 — Pteridines 2013; 24(1): 105–109

Review

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Fatty aldehyde dehydrogenase, the enzyme downstream of tetrahydrobiopterin-dependent alkylglycerol monooxygenase

Abstract: The tetrahydrobiopterin-dependent degradation of ether lipids by alkylglycerol monooxygenase (AGMO) produces fatty aldehydes, which are toxic to cells. Therefore, it is of great physiological importance that these harmful compounds are converted into their corresponding, less toxic fatty acids by fatty aldehyde dehydrogenase (FALDH). Dysfunction of this enzyme causes Sjögren-Larsson syndrome. This severe inherited disorder is accompanied by symptoms such as ichthyosis, mental retardation and spasticity. Surprisingly, fatty alcohols and not fatty aldehydes were found to accumulate in fibroblasts of Sjögren-Larsson syndrome patients, suggesting that there can be wide-ranging alterations in the lipid composition of patient cells. In particular, this has to be considered when searching for possible treatment options for patients suffering from Sjögren-Larsson syndrome. For example, inhibition of fatty aldehyde producing ether lipid degradation would have multiple implications on ether lipid- and fatty alcohol-mediated signalling pathways.

Keywords: alkylglycerol monooxygenase; fatty aldehyde dehydrogenase; Sjögren-Larsson syndrome; sphingosine-1-phosphate; tetrahydrobiopterin.

Enzymes: alkylglycerol monooxygenase (E.C. 1.14.16.5); fatty aldehyde dehydrogenase (long-chain aldehyde dehydrogenase, E.C. 1.2.1.48); lysoplasmalogenase (alkenylglycerophosphocholine hydrolase, E.C. 3.3.2.2); lysoplasmalogenase (alkenylglycerophosphoethanolamine hydrolase, E.C. 3.3.2.5); sphingosine-1-phosphate lyase (sphinganine-1-phosphate aldolase, E.C. 4.1.2.27).

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Tetrahydrobiopterin-dependent ether lipid degradation

The class of ether-linked glycerol lipids comprises a number of highly bioactive lipid compounds [1–4], which feature a characteristic alkyl residue attached to the sn-1 hydroxyl group of their glycerol backbone. Some well-known ether lipids are plasmalogens, for example, platelet-activating factor (PAF) and seminolipids [5]. Ether lipids can be found ubiquitously in nature [6, 7]. The ether bond increases the resistance towards chemical and enzymatic hydrolysis, thereby making these compounds excellent hydrophobic, membrane-permeable signalling molecules [8]. In mice with deficient ether lipid de novo synthesis, it has been shown that these lipids are involved in maintaining proper brain function, spermatogenesis and lens organisation [1, 4, 9].

The metabolic network of ether lipids consists of a number of enzymatic reactions that are responsible for the synthesis and interconversion of these compounds [10]. Furthermore, effective degradation pathways of ether lipids are important to regulate the levels of these bioactive molecules in cells. The two major ether lipid-degrading pathways are illustrated in Figure 1. The tetrahydrobiopterin-dependent cleavage of the ether bonds of alkylglycerols by alkylglycerol monooxygenase (E.C. 1.14.16.5) is one major route for this degradation. In this reaction, a glycerol derivative and a toxic fatty aldehyde are produced [11]. An alternative degradation pathway is mediated by lysoplasmalogenases (E.C. 3.3.2.2 and E.C. 3.3.2.5), which catalyse the degradation of the vinyl ether bonds of lysoplasmalogens and again produce fatty aldehydes [12]. As fatty aldehydes tend to form Schiff base adducts with free amino groups of lipids and proteins, they have to be readily metabolised into the less toxic fatty acids by fatty aldehyde dehydrogenase (E.C. 1.2.1.48) [11].

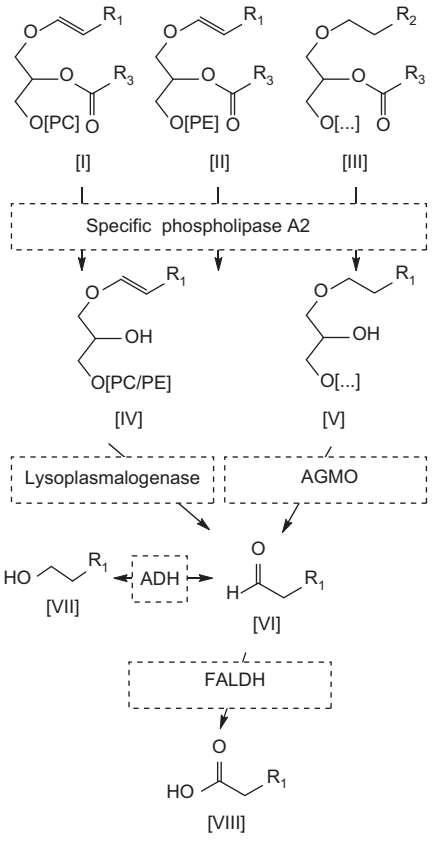

Figure 1 Mammalian ether lipid degradation pathways. Plasmalogens (I) and (II), and alkylacylglycerols (III) are hydrolysed at the sn-2 position by specific phospholipases A2. The resulting lysoplasmalogens (IV) and alkylglycerols (V) are then further degraded by lysoplasmalogenases (E.C. 3.3.2.2 and E.C. 3.3.2.5) and alkylglycerol monooxygenase (AGMO, E.C. 1.14.16.5), respectively. In each of these reactions a fatty aldehyde (VI) and a glycerol derivative are formed. The toxic aldehyde is further metabolised into the corresponding fatty acid (VIII) by fatty aldehyde dehydrogenase (FALDH, E.C. 1.2.1.48) or can be converted into a fatty alcohol (VII) by an alcohol dehydrogenase (ADH).

Alkylglycerol monooxygenase

In 1964, Tietz and coworkers demonstrated that the degradation of ether lipids in tissue homogenates is dependent on the presence of the tetrahydrobiopterin cofactor [13]. They monitored the formation of fatty acids in the presence of tetrahydrobiopterin and NAD, whereas with tetrahydrobiopterin alone, fatty aldehydes were formed. This suggested the presence of a second, NAD-dependent enzyme responsible for the conversion of fatty aldehydes into fatty acids.

It is known that alkylglycerol monooxygenase hydroxylates the C_a atom of the ether bond in alkylglycerols with the help of molecular oxygen and tetrahydrobiopterin, thereby inducing the cleavage into a fatty aldehyde and a glycerol derivative. Alkylglycerol monooxygenase accepts a broad range of alkylglycerol substrates (alkyl residue chain length 11–20 carbon atoms) [14]. Alkylglycerol monooxygenase has nine transmembrane domains [15] and is localised in the endoplasmic reticulum [16]. It is widely expressed in mouse and rat tissues with highest enzyme activities in liver, intestine and cerebellum, and low levels in skeletal muscle and heart [11, 16].

Fatty aldehyde dehydrogenase

As described above, fatty aldehydes are formed by ether lipid-degrading activities of alkylglycerol monooxygenase and lysoplasmalogenases (Figure 1). Additionally, fatty aldehydes are formed during enzymatic degradation of sphingosines via sphingosine-1-phosphate lyase (E.C. 4.1.2.27) [17], but they can also be chemically produced by lipid-derived oxidative stress or originate from the diet.

There are 19 human aldehyde dehydrogenases known that comprise the superfamily of aldehyde dehydrogenases [18]. These enzymes convert a large diversity of different aldehydes into their corresponding acids, thereby protecting cells from the potentially toxic effects of these compounds [18]. The NAD-dependent fatty aldehyde dehydrogenase (FALDH; gene symbol: ALDH3A2, E.C. 1.2.1.48) is the only membrane-bound aldehyde dehydrogenase isozyme and has a specificity for the conversion of long-chain aliphatic aldehyde substrates into their corresponding fatty acids [19]. Fatty aldehyde dehydrogenase prefers fatty aldehydes with a chain length between 8 and 20 carbon atoms, including monounsaturated and branched aldehydes, as substrates [20–22]. It has been shown that fatty aldehyde dehydrogenase is active as a homodimer [23] and is anchored to membranes by one C-terminal transmembrane domain per monomeric subunit. The ALDH3A2 gene consists of 11 exons, which can be spliced into two variants [24]. Of the total fatty aldehyde dehydrogenase, 90% is expressed as the major variant and is localised into the endoplasmic reticulum. The second splice variant has an altered carboxy-terminus which targets it to peroxisomes [24].

Substrate specificity of fatty aldehyde dehydrogenase and other ALDHs

All known aldehyde dehydrogenases are dependent on NAD(P) as cofactor [25] and together they cover a very

diverse spectrum of aldehyde substrates. Despite the large functional diversity of aldehyde dehydrogenase isozymes, they often have somewhat overlapping substrate specificities [26].

ALDH3A1 is the closest relative to fatty aldehyde dehydrogenase. ALDH3A1 was formed from ALDH3A2 during a duplication event [27]. Although both enzymes share 66% sequence identity they have very different physiological functions. In contrast to fatty aldehyde dehydrogenase, ALDH3A1 is a cytosolic enzyme. However, in a recent study we could show that ALDH3A1 can partially take over fatty aldehyde dehydrogenase activity [28]. In the same experiment no fatty aldehyde degrading activity was measured for ALDH1A1, another aldehyde dehydrogenase isozyme with 28% sequence similarity. Retinal and acetaldehyde have been reported to be the optimal substrates for ALDH1A1, whereas fatty aldehyde dehydrogenase shows almost no conversion and high K_m values in the millimolar range for these compounds [21].

To pin down the physiological function of the individual aldehyde dehydrogenase isozymes, a simple comparison of K_m values is not sufficient but also the subcellular localisation, tissue-specific expression and bioavailability of substrates have to be conscientiously considered.

Sjögren-Larsson syndrome

The overlapping substrate specificities of aldehyde dehydrogenases can sometimes compensate the dysfunction of another isozyme, as, for example, a common polymorphism of ALDH2 in parts of the Asian population only results in a decreased tolerance towards ethanol, whereas more severe symptoms are absent [29].

Fatty aldehyde dehydrogenase is the only aldehyde dehydrogenase isozyme with an associated genetic disorder, the Sjögren-Larsson syndrome. This recessive autosomal disease was first described by Sjögren and Larsson in 1957 [30]. So far, more than 70 mutations in the ALDH3A2 gene causing Sjögren-Larsson syndrome are known [31]. The major symptoms of Sjögren-Larsson syndrome are ichthyosis, mental retardation and spastic di- and tetraplegia [32].

As described above, ALDH3A1 can partially take over the fatty aldehyde-degrading function. However, differences in the expression patterns of fatty aldehyde dehydrogenase and ALDH3A1 can be the cause for the tissue-specific occurrence of symptoms such as ichthyosis in the skin and neurological symptoms such as spasticity and mental retardation in the brain. Although the highest fatty aldehyde dehydrogenase expression levels are found in the liver [23], no severe dysfunctions of this tissue have been reported in Sjögren-Larsson syndrome patients [33]. This shows that when interpreting the physiological consequences of Sjögren-Larsson syndrome, in addition to the function of fatty aldehyde dehydrogenase, biochemical parameters, subcellular localisations and tissue expression profiles of other aldehyde dehydrogenase isozymes are also of great importance.

Measuring metabolite profiles in Sjögren-Larsson syndrome patient cells as a key to identify the cause of Sjögren-Larsson syndrome symptoms

Often the dysfunction of one enzymatic step within a metabolic pathway causes changes in the neighbouring metabolome. To investigate this in fatty aldehyde dehydrogenase deficiency, we have recently compared the changes in fatty aldehyde, fatty acid, alkylglycerol and fatty alcohol handling of cultured Sjögren-Larsson syndrome patient and control cells [28]. In contrary to previous reports, which demonstrated fatty aldehyde accumulation in Sjögren-Larsson syndrome patient cells [34], we found that fatty aldehydes can be degraded at the same rate in patient fibroblasts and controls [28]. We observed that instead of the corresponding fatty acid, the respective fatty alcohols accumulate from fatty aldehydes in patient cells. This is in agreement with the occurrence of elevated fatty alcohol concentrations in patient cells published by Rizzo and coworkers [35]. Interestingly, we also found that the fatty alcohol hexadecanol is approximately threefold more toxic to patient and control cells than the corresponding fatty aldehyde [28].

There is additional evidence that the metabolic changes in cells with impaired fatty aldehyde dehydrogenase function also have far-ranging implications on other lipid pathways, as both abnormal sphingosine-1-phosphate [36] and ether lipid levels have been reported [35].

Possible treatment options for Sjögren-Larsson syndrome

So far, there is only symptomatic treatment of this disease possible [37, 38]. As discussed above, the impaired function of fatty aldehyde dehydrogenase is the initial cause of this disease, but the related symptoms are a result of a more extensive alteration of connected metabolic processes and signalling pathways. As a consequence, these

findings have to be incorporated into the search for potential treatment options for Sjögren-Larsson syndrome.

As a critical portion of fatty aldehydes is formed by degradation of important lipid signalling molecules, dietary restriction as well as enzymatic inhibition of fatty aldehyde precursors does not appear to be promising.

A more promising approach would be to search for potential pharmacological chaperones that are able to stabilise the protein folding of mutated fatty aldehyde dehydrogenase and thereby increase its residual enzymatic activity in cells. As a large portion of Sjögren-Larsson syndrome causing mutations does not affect the catalytic core itself but rather affects proper folding of fatty aldehyde dehydrogenase protein, such an approach could be a promising step towards a possible treatment option for many Sjögren-Larsson syndrome patients.

Conclusion

Dysfunction of FALDH causes deregulation of the related fatty alcohol, ether lipid and sphingolipid metabolism. Although it can be partially compensated by the action of other aldehyde dehydrogenases, several severe, tissue-specific symptoms remain. Treatment of Sjögren-Larsson syndrome patients should aim to restore the natural metabolite composition by increasing the ability of the cells to metabolise fatty aldehydes and fatty alcohols. The search for pharmacological chaperones that stabilise the protein folding of mutated fatty aldehyde dehydrogenase could thereby be a promising step towards new treatment options for Sjögren-Larsson syndrome patients.

Acknowledgments: The authors thank Petra Loitzl, Rita Holzknecht and Nina Madl for expert technical assistance. This work was supported by the Austrian Research Funds "zur Förderung der wissenschaftlichen Forschung" (P22406) and the "Tiroler Wissenschaftsfonds" (UNI-0404/589). M.A.K is a recipient of a DOC fellowship from the Austrian Academy of Science.

Received March 11, 2013; accepted March 27, 2013; previously published online May 3, 2013

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