Protein palmitoylation: Palmitoyltransferases and their specificity

Sabina Tabaczar, Aleksander Czogalla, Joanna Podkalicka, Agnieszka Biernatowska and Aleksander F Sikorski

Department of Cytobiochemistry, Faculty of Biotechnology, University of Wrocław, 50-383 Wrocław, Poland
Corresponding author: Sabina Tabaczar. Email: sabina.tabaczar@uwr.edu.pl

Impact statement

Protein palmitoylation is one of most important reversible post-translational modifications of protein function in cell-signaling systems. This review gathers the latest information on the molecular mechanism of protein palmitoyl transferase action. It also discusses the issue of substrate specificity of palmitoyl transferases. Another important question is the role of depalmitoylation enzymes. This review should help to formulate questions concerning the regulation of activity of particular PATs as well as of depalmitoylating enzymes (APT).

Abstract

A plethora of novel information has emerged over the past decade regarding protein lipidation. The reversible attachment of palmitic acid to cysteine residues, termed *S*-palmitoylation, has focused a special attention. This is mainly due to the unique role of this modification in the regulation of protein trafficking and function. A large family of protein acyltransferases (PATs) containing a conserved aspartate–histidine–histidine–cysteine motif use ping-pong kinetic mechanism to catalyze *S*-palmitoylation of a substrate protein. Here, we discuss the topology of PAT proteins and their cellular localization. We will also give an overview of the mechanism of protein palmitoylation and how it is regulated. New information concerning the recent discovery of depalmitoylating enzymes belonging to the family of α/β-hydrolase domain-containing protein 17 (ABHD17A) is included. Considering the recent advances that have occurred in understanding the mechanisms underlying the interplay between palmitoylation and depalmitoylation, it is clear that we are beginning to understand the fundamental nature of how cellular signal-transduction mediates membrane-level organization in health and disease.

Keywords: Lipidation, *S*-palmitoylation, protein acyl transferases, aspartate–histidine–histidine–cysteine protein, depalmitoylation, palmitoyltransferases

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Introduction

The attachment of deprotonated fatty acid to the cysteine residues of proteins via a thioester linkage is called *S*-acylation. It is often referred to as *S*-palmitoylation (the terminology used throughout this paper), with palmitate C16:0 being the most common fatty acid present in *S*-acylated proteins, among other long chain fatty-acyl-coenzyme As (CoAs), such as palmitoleate C16:1, stearate C18:0, and oleate C18:1.[1–3]

This reversible post-translational modification influences protein stability, function[4] and is often indispensable in the trafficking of a protein to the membrane[5] or to specific membrane domains.[6] Moreover the defects in *S*-palmitoylation of some proteins have been linked to the specific disorders.[7,8]

Although *S*-palmitoylation can occur spontaneously *in vitro*, *in vivo* it is believed to be catalyzed by the class of polytopic transmembrane proteins called protein acyl transferases (PATs) with zinc-finger and aspartate–histidine–histidine–cysteine (zDHHC) domains.[9] The *ZDHHC* family of genes in mammals consists of 23 members, which were reviewed previously.[10]

The conserved DHHC motif or DHYC in yeast Akr1p, Akr2p and Pfa5 proteins, and DQHC in the protein encoded by the mammalian *ZDHHC*13 gene;[11] was long regarded as critical for *S*-acylating activity of all PATs. Recent findings, however, show that, despite the mutations in the DHHC motif of yeast *S*-acyltransferases Swf1 and Pfa4, their activity is partially preserved.[12] The labile character of the palmitic acid attachment suggests that acylation/deacylation cycles must be tightly regulated. Although it is not known how the process is regulated, it is possible that it might occur on different levels, e.g. the availability of palmitoyl-CoA, the activity of palmitoylating and/or depalmitoylating enzymes, and protein-substrate localization and/or recognition.

The aim of this mini-review is to discuss the potential factors that might influence the palmitoylation process, with a high emphasis on PAT activity. Moreover, recent findings are presented that point toward the role of

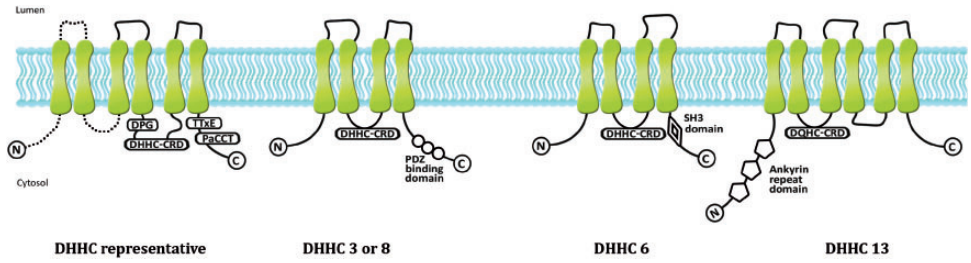

Figure 1 Predicted structure of aspartate–histidine–histidine–cysteine (DHHC) proteins. DHHC acyltransferases usually have four (rarely, six—shown as dotted lines) transmembrane domains. The conserved, catalytic DHHC motif, the site where protein becomes autopalmitoylated, is located in a cytosolic loop. DHHC signature is indicative of a cysteine rich domain (CRD-domain). DHHC proteins contain a conserved DPG motif facing the cytosol. Moreover, most DHHC enzymes have a C-terminal threonine-threonine-x-glutamate (TTxE; x stands for any amino acid residue) and palmitoyltransferase conserved C-terminus (PaCCT) motifs that are predicted to localize to the cytosol. DHHC13 is the only DHHC protein with DQHC motif. Protein–protein interacting domains such as a PDZ-binding motif (PSD95-Dlg-1-ZO), an SH3 domain and ankyrin repeats were identified in some of the DHHC proteins. (A color version of this figure is available in the online journal.)

depalmitoylating enzymes in the dynamic maintenance of the palmitoylation machinery.

DHHC proteins: Structure

Although the crystal structure of a DHHC protein has not been resolved so far, it is predicted that these proteins share common structural characteristics, e.g. they contain transmembrane domains with N- and C- termini in the cytosol (Figure 1).[13,14] Moreover, they contain a DHHC domain oriented in the cytosol, as was shown in the study involving yeast DHHC protein Akr1-invertase chimeras. The coding sequences of yeast invertase were inserted in different positions within *Akr1* gene. Since the invertase is extensively glycosylated in the lumen of the ER/Golgi complex, only those Akr1-invertase chimeras which had a luminally oriented invertase segment underwent glycosylation.[13] From this study, the typical topology of DHHC proteins is predicted as shown in Figure 1.

Usually, the DHHC signature is indicative of a cysteine-rich domain (CRD-domain).[13,15] The cysteine of the DHHC motif was shown to be a site where protein becomes acylated via a process known as autoplamitoylation. This residue is crucial for the activity of several PATs, as its mutation results in non-functional enzyme both in *in vitro* and *in vivo* studies.[3,11,16] This is not the case, however, for two yeast PATs, Pfa4, and Swf1, because palmitoylation still occurred when their DHHC cysteine residue was mutated into arginine or alanine.[12] In view of the recent work of Hemsley and Grierson,[17] it might be explained that it is possible for PATs to be acylated in *trans* when the cysteine of the DHHC motif is mutated. Moreover, at least in the case of human DHHCs 5, 6, and 8, autoplamitoylation was detected in the C-terminal region defined as CCX7-13C(S/T) motif, where three cysteine residues are present.[18] Neither the remaining human PATs nor yeast PATs, however, have this motif.

DHHC proteins also share other sequence similarities such as threonine-threonine-x-glutamate (TTxE; x stands for any amino acid residue) and aspartate–proline–glycine (DPG) motifs, which are located outside the cysteine-rich domain but on the same surface of the membrane as the DHHC domain.[19] Most DHHC enzymes also contain a C-terminal palmitoyltransferase conserved C-terminus (PaCCT) motif (Figure 1).[20] The mutation of tyrosine 323 that is located within PaCCT results in the lack of Swf1 palmitoylation activity *in vivo*. Additionally, a diminished function of yeast Pfa3 was observed when PaCCT phenyloalanine 250 was mutated to alanine.[20] Conserved structural motifs are shown in Figure 2.

Two of the mammalian PAT proteins (DHHC13 and 17) differ from the remaining family members by the presence of an ankyrin repeat (ANK) domain located at the N-terminus, which faces the cytosol (Figure 1).[19] ANK domains have been shown to be involved in protein–protein interactions[21] and, in the case of DHHC enzymes, play a role in substrate recognition and interaction.[22] Additionally, other protein binding domains, such as SH3 (*S*rc *H*omology 3) and PDZ (*P*SD95-*D*lg-1-*Z*O) domains, have been identified in some of the DHHC proteins (Figure 1).[23,24]

The DHHC-CRD of PATs displays some homology with the C_2H_2 zinc-finger and non-Cys_2His_2 motif and has been termed NEW 1.[25,26] Recent work indicates that the integrity of the CRD domain depends on two CCHC zinc-fingers, which are responsible for the binding of zinc at a 2:1 zinc/protein molar ratio by this domain.[15,27] Several residues have been predicted to play a role in the co-ordination of zinc and their mutations results in the formation of unstable protein, probably due to misfolding of the zinc-finger domain. On the other hand, it has been suggested that various structure-stabilizing elements, such as hydrogen bonds, might contribute to the proper folding of PAT making zinc-coordination not required.[27]

Despite many years of research on DHHC proteins, their crystal structure still awaits to be resolved. Such a discovery would certainly contribute to the better understanding of their function and substrate-specificity.

DHHC proteins: Localization

The first report on DHHC localization arose from the work of Ohno *et al.*,[14] where the Golgi and the ER were indicated as the two organelles in which most human DHHC proteins are localized. They also showed that human DHHC5 and 20 could be found at the plasma membrane when overexpressed in HEK 293T cells and yeast DHHCs; Pfa3 resides in the vacuole and Pfa5 in the plasma membrane.

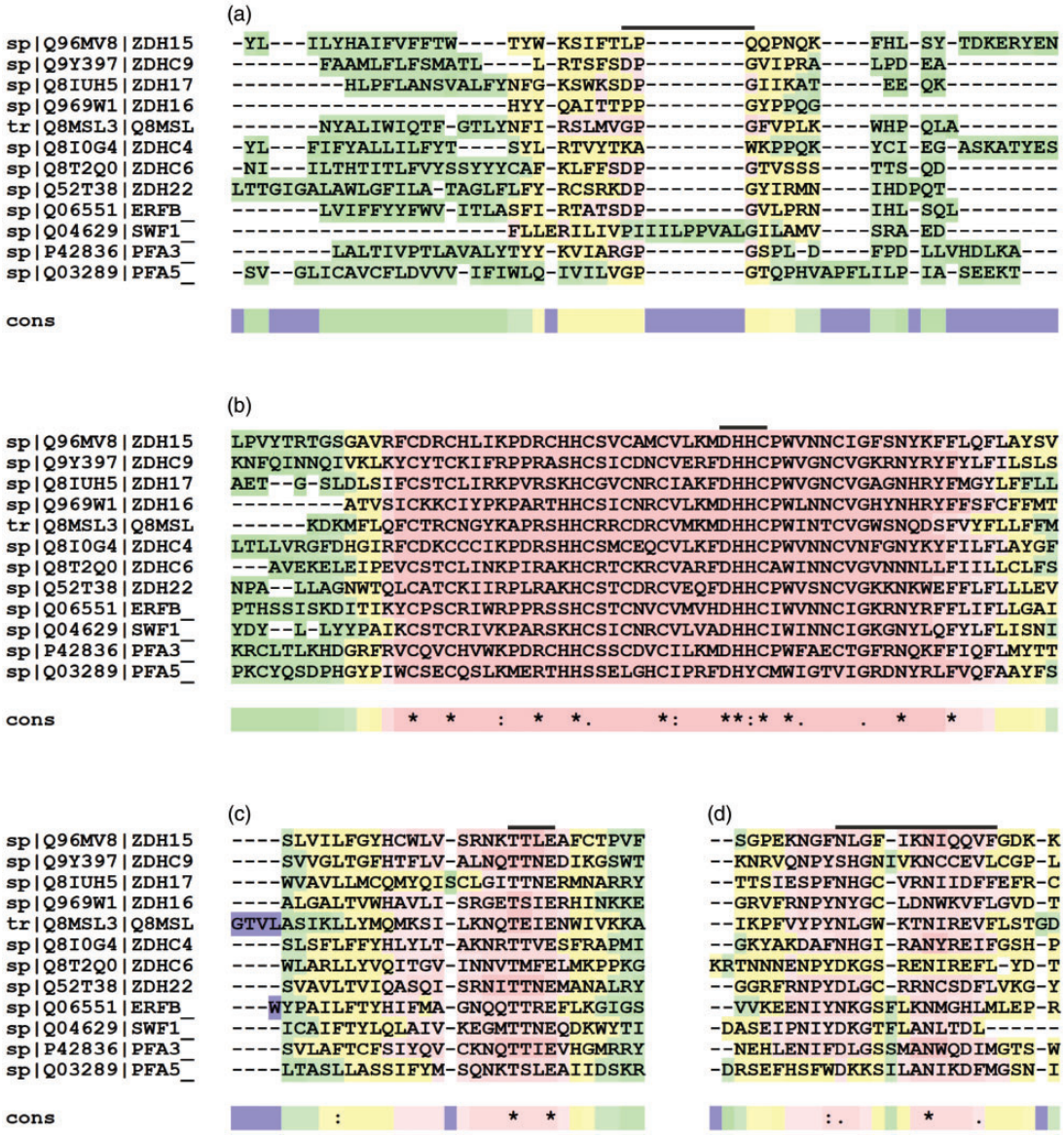

Figure 2 Homology of palmitoyl transferase proteins. Multiple sequence alignment of the conserved regions (highlighted in black) comprising: (a) the DPG motif, (b) cysteine-rich domain (CRD-domain) containing aspartate–histidine–histidine–cysteine (DHHC) motif, (c) the threonine–threonine–x-glutamate (TTxE), and (d) the palmitoyltransferase conserved C-terminus (PaCCT) for selected PATs from model organisms. Amino acid residue sequences of PATs were extracted from UniProt database and alignments were generated using T-Coffe Algorithm.[69] The sequence identification numbers (UniProt) for the model organisms are as follows: Q96MV8—palmitoyltransferase ZDHHC15, *Homo sapiens*; Q9Y397—palmitoyltransferase ZDHHC9, *H. sapiens*; Q8IUH5—palmitoyltransferase ZDHHC15, *H. sapiens*; Q969W1—palmitoyltransferase ZDHHC16, *H. sapiens*; Q8MSL3—palmitoyltransferase, *Drosophila melanogaster*; Q8I0G4—zinc-finger DHHC domain-containing protein 4, *Caenorhabditis elegans*; Q8T2Q0—putative ZDHHC-type palmitoyltransferase 6, *Dictyostelium discoideum*; Q52T38—protein *S*-acyltransferase 24, *Arabidopsis thaliana*; Q06551—palmitoyltransferase ERF2, *Saccharomyces cerevisiae*; Q04629—palmitoyltransferase SWF1, *S. cerevisiae*; P42836—palmitoyltransferase PFA3, *S. cerevisiae*; Q03289—palmitoyltransferase PFA5, *S. cerevisiae*. (A color version of this figure is available in the online journal.)

Recent findings point toward Golgi membranes as the major site of residency of active human DHHC protein.[22,24,28,29]

DHHC proteins can cycle between different compartments, as was shown for DHHC2 which moves between the plasma membrane and endosomes. Special attention has to be paid to what co-localization markers are used for determining the protein disposition within the cell, particularly when considering closely positioned cellular compartments such as Golgi, trans-Golgi network (TGN), and recycling endosomes.[30]

It is not clear what governs DHHC proteins to their specific localization. The recent studies of Chamberlain's group suggest that the C-terminal pentapeptide might play a role as a targeting signal. It was shown, for example, that DHHC4 and 6 have a dilysine consensus sequence at their C-termini (KKEK in the case of DHHC4 and KKXX in the case of DHHC6) that targets them to the ER. The removal of these residues, in turn, results in the redistribution of these PATs to different cellular compartments.[31] Similarly, the intracellular distribution of DHHC2 and DHHC15 was proven to be dependent on the C-terminal sequence (68 amino-acid residue long in the case of DHHC2, and a shorter but corresponding sequence of DHHC15).[30] Another study reports that, when a highly conserved C-terminal

phenylalanine (F233) of DHHC21 is deleted, the protein changes its location from the Golgi to ER membranes.[32]

It would be interesting to determine whether the C-terminus sequence of all DHHC proteins contains the coding information that determines their cellular localization. Thus, more work is needed in this field.

ZDHHC proteins: Substrate-specificity

The substrate-specificity of PATs remains the most controversial, yet unresolved issue concerning their biology. This has mainly arisen from conflicting studies which show either that an individual DHHC is indispensable for a certain substrate, i.e. the depletion of a specific PAT results in a diminished palmitoylation of the substrate, or that different DHHCs have overlapping activities.[33] Examples of the first case are DHHC21, for the endothelial nitric oxide synthase (eNOS) substrate,[28] or DHHC17 for huntingtin.[34]

If the hypothesis concerning PATs specificity is correct, then their differential substrate preference must be due to some differences that lie within both the enzyme and the substrate structure. Indeed, taking a domain from the active DHHC3 and putting it into an enzyme which is not active (DHHC15) toward a specific substrate (SNAP23), did not confer specific activity of the latter.[35] Moreover, it is not only the CRD-domain that is important for a substrate recognition and interaction, but also distal sequences could also be involved, as shown before.[34]

Additionally, even a subtle change within the palmitoylated motif of a substrate, such as that used in the study of Greaves, where the cysteine residue of SNAP23 (which is not a substrate for DHHC3) was substituted by an phenylalanine residue, as in SNAP25b (which is a substrate for DHHC3), resulted in the palmitoylation of a chimeric protein, indicates that specificity may depend upon substrate structure as well.[35]

The protein–protein interacting domains of either DHHC proteins or their substrates might be an additional factor involved in substrate recognition. A recent report highlights the important role of DHHC-ANK domains in the recognition of the substrate sequence.[36] The new substrate consensus sequence recognized by the DHHC17 and 13 ANK domains was detected in several proteins including, among others, SNAP25, SNAP23, and huntingtin.[36] Similarly, it was shown that the PDZ domain of DHHC5 binds glutamate receptor interacting protein (GRIP1b), facilitating the palmitoylation of GRIP1 band its subsequent trafficking to its dendritic localization.[37] PDZ-mediated interactions between DHHC5 and neuronal PSD-95 protein was previously shown[38] and, in a separate study, a mutant of DHHC5 with a deletion of the PDZ domain (DHHC5 ΔPDZb) could not bind to PSD-95.[39] Moreover, SH3 domain and SH3-binding domain interactions between selenoprotein K (Selk) and DHHC6 were reported to be required for palmitoylation of the Ca^{2+} channel protein, inositol 1,4,5-triphosphate (IP3) receptor: IP3R.[40]

On the other hand, the protein interaction/recognition domains of DHHC proteins might play roles separate to substrate recognition as, for example, in the case of the C-terminal residues of DHHC5, which were shown to be involved in the interaction with the SH3 domain of Fyn, a member of the Src-family of kinases. Mutation of the most critical prolines that lie within this sequence resulted in a protein that was unable to bind to the SH3 domain of Fyn which, in turn, blocked the Fyn-mediated phosphorylation of DHHC5, thus enhancing the dissociation of DHHC5 from the membrane.[39]

Mention should also be made of the study that argues against any specific DHHC-substrate interactions, stating that, in order for a protein to be palmitoylated, it only needs to have an accessible cysteine residue that can transiently locate at Golgi membranes; the basic site in the cell where PATs reside.[24] Thus, further studies are needed to finally resolve the issue of PAT substrate-specificity.

Mechanism of palmitoylation

DHHC proteins use a two-step transfer mechanism to palmitoylate their substrates. During this process an enzyme first becomes autoacylated and then transfers its attached palmitoyl-residue to the target protein, as shown in a real-time autopalmitoylation fluorescence assay and in a single turnover assay with a radiolabelled acyl group (Figure 3(a)).[41,42] A two-stage mechanism has also been suggested for yeast Pfa3-mediated *S*-palmitoylation.[33] Most probably, the cysteine of the DHHC motif is the primary one which is autoacylated, as shown in many studies, although secondary sites of autoacylation might also exist.[12,18,42] From *in vitro* kinetics studies, the stoichiometry is one acyl chain per DHHC motif.[42]

In order for autoacylation to occur, the target cysteine residue has to be in a deprotonated state as a thiolate anion that acts as a nucleophile for palmitoyl-CoA. The reactivity of the thiol group of cysteine varies with pH and it also differs between proteins as well as upon its localization within protein. An approximation for the acid dissociation constant (pK_a) value for an average cysteine residue in proteins is ~8.5. Considering that the pH of the cytosol is around 7.2–7.4, it is clear that some other factor (or factors) must modulate the local conditions in order to favor the formation of a thiolate. As shown before, depressing the pK_a of cysteine can be achieved via hydrogen bond donation and dipolar interactions.[43] Additional factors that can influence the pKa of cysteine albeit, previously reported, as being less important, could be neighboring amino-acid residues, e.g. lysine, arginine, or protonated histidine side chains.[44] Indeed, the studies of Mitchell *et al*.[41] and Gonzalez Montoro *et al*.[12] suggest that the first histidine within the DHHC motif plays a significant role in the second step of the palmitoylation mechanism, involving the transfer of palmitoyl residue from PATs to the target protein. This might be due to histidine-aided deprotonation of the cysteine residues of the acceptor protein. The mutation of the first histidine in the DHHC motif into alanine resulted in the Erf2 DAHC being unable to acylate its substrate, Ras2, despite it being autoacylated.[41] In addition, in both *in vitro* and *in vivo* experiments, the yeast Akr1p mutant, AAYC, was neither autopalmitoylated or was able to demonstrate any *S*-palmitoylation activity.[11] On the contrary, a DQHC mutant of Swf1 undergoes

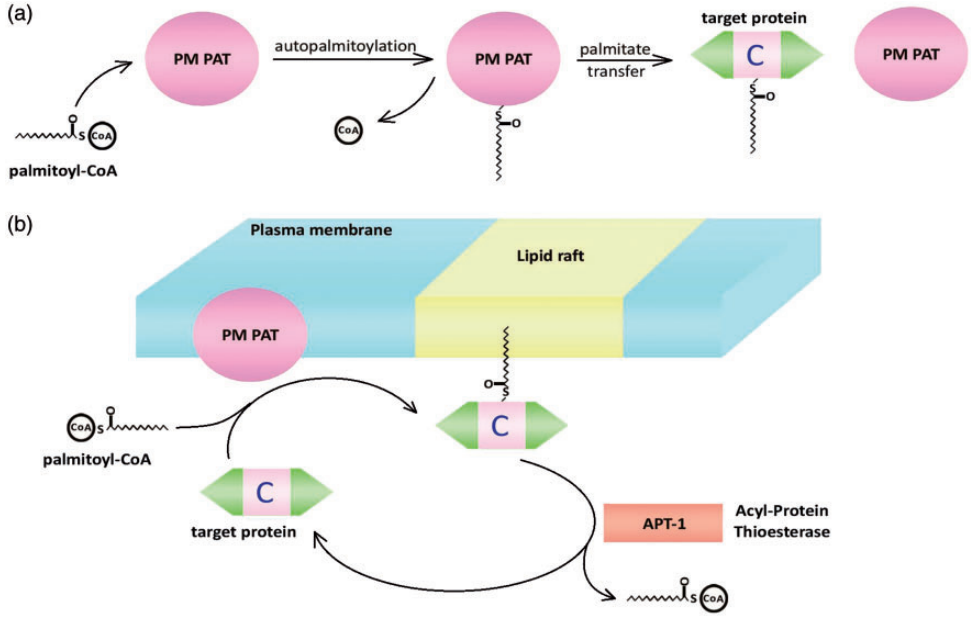

Figure 3 (a) The ping-pong mechanism of PAT-mediated palmitoylation. The reaction is seen to occur in two steps. In the first, palmitoyl-CoA reacts with the enzyme, leading to the formation of a PAT intermediate, with a concomitant release of CoA. In the second step, palmitate is transferred to the target protein. The binding of the two substrates (palmitoyl-CoA and a target protein) to PAT causes the enzyme to switch back and forth between two stages (palmitoylated versus non-palmitoylated) (ping-pong).[42] (b) palmitoylation-depalmitoylation cycle. PAT-mediated attachment of palmitic acid to the target protein might allow protein to be localized to different cell compartments, e.g. membrane microdomains, i.e. lipid rafts. In turn, enzymatic removal of palmitoyl group by acyl-protein thioesterase causes the target protein to translocate back to the cytosol. (A color version of this figure is available in the online journal.)

autoacylation and has partial PAT activity.[12] Moreover, the human DHHC13 protein contains a natural DQHC motif and actively palmitoylates huntingtin protein and ClipR-59.[45,46]

It is not only intramolecular factors that can modulate the mechanism of palmitoylation, but intermolecular factors can also be involved. The study of Duncan and Gilman[47] showed an augmented *in vitro* palmitoylation of Giα1 when a 'partner' protein $\beta\gamma$ is present. Similarly, the protein–protein interaction between SNAP-25 and the SNARE protein, syntaxin 1, affects palmitoylation efficiency *in vitro*.[48] In addition, some DHHC enzymes might also require the presence of additional protein cofactors. For example, in order for DHHC9 to become autopalmitoylated and to then be able to transfer palmitoyl-CoA to the substrate, it needs to be in a complex with a Golgi protein, GCP16.[49]

The intracellular pool of palmitoyl-coenzyme A and its availability are other factors influencing the efficiency of the palmitoylation process. The ubiquitously expressed acyl-CoA-binding protein, also known as diazepam-binding inhibitor (DBI)[50] plays a key role in the maintenance of acyl-coenzyme A homeostasis.[51] DBI belongs to acyl-CoA binding domain-containing (ACBD) family of proteins and binds medium- and long-chain acyl-CoA esters (C14–C22).[52,53] The equilibrium dissociation constant (K_D) values obtained by microcalorimetry titration method for octanoyl-CoA, dodecanoyl-CoA and hexadecanoyl-CoA binding to recombinant bovine acyl-CoA-binding protein were $0.24 \pm 0.02 \times 10^{-6}$ mol/L, $0.65 \pm 0.02 \times 10^{-8}$ mol/L, and $0.45 \pm 0.02 \times 10^{-13}$ mol/L, respectively.[54]

Acyl-CoA sequestration by ACBD-containing protein has a negative impact on the spontaneous palmitoylation of short polypeptides with sequences corresponding to the natural *S*-palmitoylation sites of protein.[55] Conversely, other reports state that, when a physiological ratio of ACBD-containing protein and acyl-CoA (1.5:1) was used, enzymatic, PAT-mediated, palmitoylation seemed not to be affected in the presence of ACBD.[9] On the other hand, the suppressing effect of ACBD-containing protein was also observed in the case of non-enzymatic palmitoylation of proteins such as G-protein α-subunits[9] and MPP1/p55 erythroid membrane protein.[56] Palmitoyl-CoA complexed with rhACBD cannot serve as a substrate for enzymatic protein palmitoylation in the case of MPP1/p55.[56]

Depalmitoylation

The reversible nature of protein palmitoylation suggests that the mechanism that controls protein depalmitoylation also exists and is under tight control. Therefore depalmitoylation likewise palmitoylation participates in controlling cellular trafficking and membrane localization of many proteins (Figure 3(b)). The cytoplasmic enzymes, acyl-protein thioesterases 1 and 2 (APT1 also known as Lypla1 and APT2), belonging to the metabolic serine hydrolase (mSH) superfamily[57] have been shown to have a depalmitoylating activity against various proteins.[58–60] When APT1 enzyme activity was blocked by hexadecylfluorophosphonate (HDFP), proteins such as Ras GTPases, G proteins, and MAGUK proteins (MPP1 and MPP6) were not depalmitoylated.[61] Additionally, a newly developed compound

containing a β-lactone core, palmostatin B, effectively blocked the APT1/APT2-mediated hydrolysis of palmitate from Ras proteins.[62,63]

APT1 and APT 2 differ in their substrate-specificity, e.g. APT-2 but not APT-1 is implicated in the deacylation of the growth-associated protein-43 (GAP-43)[60] while, in another study, APT-1 but not APT-2 was able to depalmitoylate the cysteine residues present on the S0-S1 loops of calcium/voltage-gated, large conductance potassium (BK) channels.[64] So far, the substrate consensus sequence for each of these APTs has not been resolved.

Unexpectedly, APT1 and APT2 are also the substrates for PAT with their cysteine-2 residue being palmitoylated.[4,18] This modification allows their translocation from the cytosol and the membrane attachment of these acyl-protein thioesterases. Increased dosing of the potent palmitoylation inhibitor, 2-bromopalmitate, resulted in the gradual decrease of the membrane APT fractions, which correlated with localization change of APTs substrates, i.e. H-ras and GAP-43. Moreover, APT1 was shown to undergo auto-depalmitoylation and to catalyze the hydrolysis of palmitoyl-CoA from APT2.[4]

Besides, APT1 and APT2 APT-like thioesterase (APTL1), known also as LYPLAL1, has been shown to participate in the depalmitoylation of the BK channel. APTL1 was shown to be a distant homolog of APT1.[64] However, considering the structure of LYPLAL1 that shows the presence of a shallow binding pocket, it has been suggested that its depalmitoylating activity is rather limited, with a preference for shorter-chain lipid substrates.[65]

Recently, the discovery of new enzymes that can, either alone, or in combination with APT1/2, play a role in the depalmitoylation of various proteins has been reported. Namely the existence of other, yet to be defined, depalmitoylating enzymes has been predicted in a study reporting APT1/2-independent depalmitoylation of the SNARE-like protein44, R7 RGS-binding protein (R7BP).[66] Indeed, the mammalian αβ hydrolase-domain (ABHD) containing proteins have emerged as potential novel depalmitoylating enzymes. Three isoforms of membrane-anchored ABHD, namely, ABHD17; ABHD17A, 17B, and 17C were shown to depalmitoylate PSD-95.[67] Similarly, enhanced palmitate removal by ABHD17 was observed in the case of overexpressed N-Ras in HEK293T cells, but not when endogenous N-Ras in the neuronal cells was studied.[67,68]

The amino acid residues which are indispensable for the catalytic activity of ABHD17 included amino acid S170, together with residues D235 and H264, as their mutation resulted in inactive protein.[67] Similarly to APT1 and APT2, the palmitoylation of ABHD17 proteins is required for their targeting to the plasma membrane[61] and for conferring their depalmitoylating activity toward PSD-95.[67]

The discovery of new depalmitoylating enzymes suggest the existence of a new layer of complexity in the palmitoylation/depalmitoylation network. Considering that the enzymes that mediate palmitate removal are also the substrates for PATs, it is obvious that specific interplay exists between these two processes. Moreover, the repertoire of depalmitoylation enzymes might be larger than was originally thought to be.

Conclusion

Protein palmitoylation is one of most important reversible post-translational modifications of proteins functioning in cell-signaling systems. Palmitoylation affects a variety of proteins and is a way of controlling their cellular trafficking and membrane localization. Here, we have discussed the role of palmitoyltransferases and their substrate specificity, along with the factors that might influence the palmitoylation process. Despite the recent advances in the field, especially in terms of the methods that allow better detection of palmitoylation, there are still many questions that need to be answered, particularly those concerning substrate specificity. Although substantial progress has been made in research of molecular mechanism of palmitoylation reaction, the precise mechanism of how palmitoylation and depalmitoylation cycles are regulated awaits further studies.

Authors' contributions: All authors participated in initial discussion concerning the concept of the review. ST and AFS wrote the main text, AC prepared the homology search and diagram. All authors discussed the final form of the review.

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DECLARATION OF CONFLICT OF INTEREST

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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