Biochimica et Biophysica Acta 1864 (2016) 1707–1713

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

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

Review

Function of membranous lysyl-tRNA synthetase and its implication for tumorigenesis

Young Ho Jeon [a,b], Jung Weon Lee [b,c,*], Sunghoon Kim [b,c]

[a] College of Pharmacy, Korea University, Sejong 30019, Republic of Korea
[b] Medicinal Bioconvergence Research Center, Seoul National University, Seoul 08826, Republic of Korea
[c] Research Institute of Pharmaceutical Sciences, College of Pharmacy, Seoul National University, Seoul 08826, Republic of Korea

ARTICLE INFO

Article history:
Received 13 July 2016
Received in revised form 28 August 2016
Accepted 19 September 2016
Available online 20 September 2016

Keywords:
Lysyl-tRNA synthetase
Laminin receptor
Cancer metastasis
Protein-protein interaction
Anti-cancer drug
Membrane localization

ABSTRACT

Aminoacyl-tRNA synthetases (ARSs) are essential enzymes that conjugate specific amino acids to their cognate tRNAs for protein synthesis. Besides their catalytic activity, recent studies have uncovered many additional functions of these enzymes through their interactions with diverse cellular factors. Among human ARSs, cytosolic lysyl-tRNA synthetase (KRS) is often highly expressed in cancer cells and tissues, and facilitates cancer cell migration and invasion through the interaction with the 67 kDa laminin receptor on the plasma membrane. Specific modulation of this interaction by small molecule inhibitors has revealed a new way to control metastasis. Here, we summarize the pro-metastatic functions of KRS and their patho-physiological implications.

1. Introduction

Aminoacyl-tRNA synthetases (ARSs) are ancient enzymes that conjugate specific amino acids to their cognate tRNAs for protein synthesis. However, these enzymes have adopted additional non-catalytic domains that have contributed to the expansion of their function beyond protein synthesis [1,2]. For last decade, the number of reports showing new functions of ARSs in the control of transcription, translation, RNA splicing in the context of immune responses, angiogenesis and cell fate determination has rapidly increased [3]. The numerous catalytic and non-catalytic activities of ARSs are also pathologically associated with various human diseases [4,5]. Considering the presence of cytosolic and mitochondrial sets of ARSs and their structural diversitifcation via alternative splicing and post-translational modification, many more functions and roles are anticipated to be unveiled for this group of enzymes. This review will focus on the new functions of lysyl-tRNA synthetase (KRS) in the plasma membrane and its patho-physiological implications.

2. Catalytic activity of KRS

Protein biosynthesis is essential for all the living organisms, requiring diverse cellular components and steps. ARSs are necessary for the process of translating mRNAs to proteins [1], by attaching amino acids to their cognate tRNAs. The resulting aminoacyl-tRNAs are then transferred to ribosome, which incorporates amino acids to the growing end of the polypeptide chain.

Like other ARSs, KRS accomplishes catalysis in two steps [6]. The first reaction (1) involves the activation of lysine, where KRS selectively binds lysine by using one molecule of ATP.

$$\text{Lysine} + \text{ATP} + \text{KRS} \rightarrow (\text{Lysine} - \text{AMP})\text{KRS} + \text{PPi} \quad (1)$$

The second reaction (2) involves the transfer of the activated lysine (lysine-AMP) to the acceptor end of $tRNA^{Lys}$ [4].

$$(\text{lysine} - \text{AMP})\text{KRS} + tRNA^{Lys} \rightarrow \text{lysyl} - tRNA^{Lys} + \text{KRS} + \text{AMP} \quad (2)$$

Combining the reactions of [(1) + (2)],

$$\text{ATP} + \text{lysine} + tRNA^{Lys} = \text{AMP} + \text{PPi} + \text{lysyl} - tRNA^{Lys} \quad (3)$$

KRS is normally located in the cytosol as a component of the multi-tRNA synthetase complex (MSC) that consists of nine different ARSs and three auxiliary factors named AIMP1, 2 and 3 [7,8]. A homodimer of KRS is linked to the N-terminal peptide of AIMP2, and the MSC contains two units of the KRS homodimer-AIMP2 monomer complex. This suggests that KRS would exist in the MSC as a tetramer that is present at a relatively higher stoichiometry, compared to other components that are bound to the MSC as monomers or dimers [9] (Fig. 1). This

* Corresponding author at: College of Pharmacy, Medicinal Bioconvergence Research Center, Seoul National University, Seoul 08826, Republic of Korea.
E-mail address: jwl@snu.ac.kr (J.W. Lee).

http://dx.doi.org/10.1016/j.bbapap.2016.09.009
1570-9639/

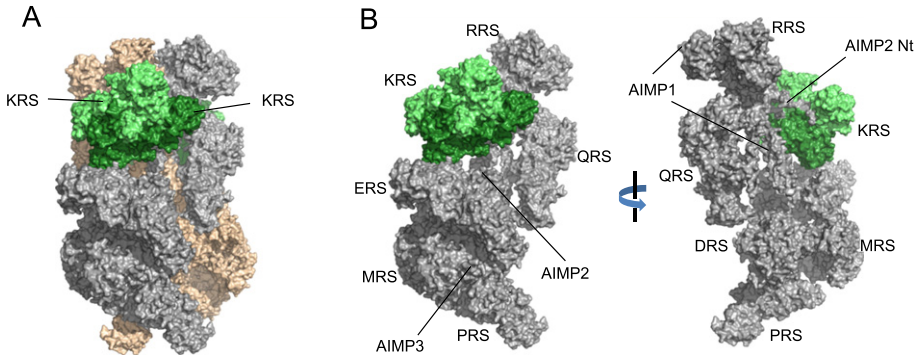

Fig. 1. Structure of KRS bound to the MSC. The heterotetrameric complex of the MRS-AIMP3-EPRS-AIMP2GST domains [67] and the ternary complex of QRS-AIMP1-RRS [68] are used as a platform for building the MSC model. The KRS dimer is linked to the N-terminus of AIMP2. AIMP2 is linked to the EPRS and AIMP1, which are linked to AIMP3-MRS and QRS-RRS. DRS is linked to the GST domain of AIMP2 at the opposite side of KRS binding. Assuming each AIMP2 binds to the KRS dimer, two KRS dimers (denoted in green and palegreen) are contained in the MSC complex. The components of the MSC are symmetrically duplicated as (A) (grey and orange), and one pair of sub-complexes are shown in (B). Due to lack of the structure information, LRS and IRS are not included in this model.

would make it possible for KRS to dissociate from the MSC to perform additional activities in other cellular locations without losing its catalytic capability of protein synthesis.

3. Non-catalytic roles of KRS

Although the canonical role of KRS in protein translation is important, many studies on KRS in different systems show diverse non-canonical functions of the enzyme including production of the signaling molecule diadenosine tetraphosphate [10], death signaling of stressed cells [11], cytokine-like immune signaling [12], transcriptional roles for HIV replication [13], interaction with mutant superoxide dismutase 1 in amyotrophic lateral sclerosis (ALS) [14], and regulation of cell migration [15]. Since other roles of KRS at the transcriptional levels in HIV replication [16,17] and at the gene expression level in immune response [10] are well documented elsewhere, this review will focus on KRS-dependent regulation of cell migration in cancer metastasis.

3.1. Secretion and nuclear translocation of KRS and its functional implications

Several different ARSs are secreted for distinct extracellular activities [12,18–21]. Although KRS is normally bound to the MSC in the cytosol as described above, its secretion is induced by TNF-α [12]. Secreted KRS is bound to macrophages and peripheral blood mononuclear cells to promote the TNF-α production and cell migration. Although its pathological implication is not currently understood, the secreted KRS may play a crucial role in mediating cancer cell survival and metastasis.

KRS is translocated into the nucleus when mast cells are activated by the exposure to antigens. For its nuclear localization, KRS requires phosphorylation at S207 by the IFN-g-induced MAPK pathway. Since S207 is located at the junction between the C-terminal catalytic and N-terminal anticodon-binding domains, its phosphorylation provides the repulsive force necessary to push the two domains apart [22]. In this conformation, KRS cannot work to deliver lysine to the acceptor end of its substrate tRNA. Instead, the S207 phosphorylated KRS generates diadenosine tetraphosphate (Ap4A), which is considered as a second messenger [22]. The nuclear translocated KRS binds to a transcription factor, MITF, that is maintained in an inactive form by complexing with HINT. In the nucleus, KRS binds to MITF, and the KRS-generated Ap4A binds to HINT to reduce its affinity to MITF [23]. Thus, KRS releases MITF from HINT via production of Ap4A to trigger the expression of its target genes in the immune response [10].

3.2. Membrane localization of KRS

Besides extracellular secretion and nuclear translocation, KRS has also been reported to locate to the plasma membrane upon stimulation by laminin but not by collagen or fibronectin [24]. The laminin-induced dissociation of KRS from the multi-tRNA synthetase complex (MRC) and its membrane translocation depends on phosphorylation of the Thr52 residue of KRS via the PI3K-p38MAPK pathway. Once in the plasma membrane, KRS can associate with other proteins or receptors to transduce intracellular signaling processes. Cellular proteins that bind to human KRS have been identified by yeast 2-hybrid assay using HeLa cell cDNA libraries [25]. These proteins include AIMP2/p38 (gene identification: 7965) in the MSC [26] and FANCC-interacting protein (FAZF; gene identification: 27033) (binding to the full-length KRS), and hypoxanthine phosphoribosyltransferase 1 (HPRT1; gene identification: 3251), laminin receptor (LR/RPSA) (the monomeric form of laminin receptor is also known as ribosomal subunit p40 or 37LRP, gene identification: 3921), and cyclophilin B (cypB, gene identification: 5479) (binding to the N-terminal peptide of KRS). Among the identified proteins, p40 was of particular interest and was, therefore, studied further to understand the functional implications of the p40-KRS association. p40 is converted to the 67 kDa laminin receptor (p67LR) by dimerization and, thus, it is likely that membrane localization of KRS upon laminin stimulation would be meaningful for sensing extracellular microenvironmental cues and transducing intracellular signaling pathways [27].

3.2.1. Protein interaction of KRS in plasma membrane

The membranous KRS has been shown to bind p67LR, which can induce laminin-dependent cell migration [24]. The p67LR also associates with integrin α6β1 [28] or α6β4 [29]. The integrins are important for cell-extracellular matrix (ECM) adhesion, and are involved in cell migration and invasion [30], as well as activation of signal transductions activation leading to tyrosine phosphorylation of focal adhesion kinase (FAK), paxillin, and c-Src during cell-ECM adhesion [31]. During cell adhesion, cells activate intracellular signaling pathways involving RhoA GTPases, FAK, c-Src, ERKs and paxillin [32]. These activations can lead to the reorganization of F-actin cytoskeletal networks to form new integrin-ECM interactions and subsequent tractive force generation [33].

KRS is often highly expressed in cancer cells [34–36]. In cancer cell lines in which KRS is highly expressed, therefore, laminin-induced membrane translocation of KRS positively leads to the stabilization of p67LR on the plasma membrane. Thereafter, the complex formation consisting of KRS, p67LR, and other integrins transduces intracellular