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Minireview

CIN85/CMS family of adaptor molecules

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Abstract CIN85 and CMS belong to a family of ubiquitously expressed adaptor molecules containing three SH3 domains, a proline-rich region and a coiled-coil domain. By binding to numerous proteins they assemble multimeric complexes implicated in cell-specific signals controlling T-cell activation, kidney glomeruli function or apoptosis in neuronal cells. CIN85/CMS also associate with accessory endocytic proteins, components of the actin cytoskeleton as well as other adaptor proteins involved in receptor tyrosine kinase (RTK) signaling. These interactions enable CIN85/CMS to function within a network of signaling pathways that co-ordinate critical steps involved in downregulation and degradation of RTKs.

Key words: CIN85; CMS/CD2-associated protein; Cbl; Receptor tyrosine kinase; Endocytosis

1. Introduction

Adaptor molecules are non-catalytic polypeptides that contain one or more domains able to bind to other protein or non-protein ligands [1]. These molecules selectively control the spatial and temporal assembly of multiprotein complexes that transmit intracellular signals involved in regulation of cell growth, differentiation, migration and survival [1]. The importance of such signaling networks is well understood for signal transduction induced by receptor tyrosine kinases (RTKs) [2]. Binding of growth factors to RTKs promotes receptor activation leading to autophosphorylation and phosphorylation of numerous cellular proteins. This initiates a cascade of protein–protein interactions and formation of signaling networks that mediate proliferative and differentiating signals [1,2]. Activation of RTKs also leads to their rapid removal from the cell surface in a process dependent on receptor ubiquitination and dynamic interaction with accessory endocytic proteins [3]. For example, Cbl, a ubiquitin ligase, binds to and ubiquitinates activated RTKs and also recruits CIN85–endophilin complexes to mediate receptor internalization [4–6]. Once internalized, RTKs are delivered into the endosomal compartment where receptors get sorted for either recycling back to the cell surface or targeting to lysosomes for degradation [3]. This review describes a recently identified family of adaptor proteins known as the CIN85/CMS family that is involved in orchestrating multiple steps in RTK signaling and endocytosis.

2. Identification and structure of CIN85 and CD2-associated protein (CD2AP)

CIN85 was independently identified as Cbl-interacting protein of 85 kDa [7], Ruk (regulator of ubiquitous kinase) [8], SETA (SH3 domain-containing gene expressed in tumorigenic astrocytes) [9] and SH3KBP1 (SH3 domain kinase binding protein 1) [10]. These genes were isolated from either human (CIN85), rat (Ruk and SETA) or mouse (SH3KBP1) sources and share between 92% and 97% sequence identity, suggesting that they represent homologues of one gene. Since CIN85 was the first gene cloned we have used this name throughout the review. The CIN85 gene is localized on the distal arm of the X chromosome (Xp22.1–p21.3) in mouse [10]. A main 3.2 kb CIN85 mRNA is expressed in all adults and newborn tissues [7,8,11]. Due to alternative splicing and different promoter usage, multiple CIN85 mRNA messages have been detected, which showed a more restricted pattern of expression [8,11].

Another adaptor protein known as CD2AP [12], CMS (Cas ligand with multiple SH3 domains) [13] or METS-1 (mesenchyme-to-epithelium transition protein with SH3 domains) [14] displays high sequence and structural similarities to CIN85. CMS was cloned from human placenta by its ability to interact with p130Cas [13], while CD2AP/METS-1, a mouse homologue of CMS, was identified by binding to the adhesion receptor CD2 or as a gene strongly induced during kidney development, respectively [12,14]. The CMS gene is localized on chromosome 6 (6p12) in humans. A prominent 5.4 kb CMS mRNA is ubiquitously expressed in adult and fetal human tissues [13].

The overall domain organization of CIN85 and CMS/CD2AP is identical and they share 39% identity and 54% similarity in amino acid sequence (Fig. 1). At the amino-terminus CIN85 and CMS/CD2AP contain three SH3 domains known to mediate protein–protein interactions by binding to proline-rich motifs. These three SH3 domains share higher similarity among themselves than to any other SH3 domains, suggesting that they may have overlapping specificities in binding. A region rich in serine and threonine residues, which could be subject to phosphorylation, lies between the second and third SH3 domains. There are also three FxDxF sequences in the amino-terminus that may serve as binding sites for the clathrin adaptor protein AP2 [15]. A proline-rich region is found adjacent to the third SH3 domain of CIN85/CMS, providing potential recognition sites for other SH3 domain-

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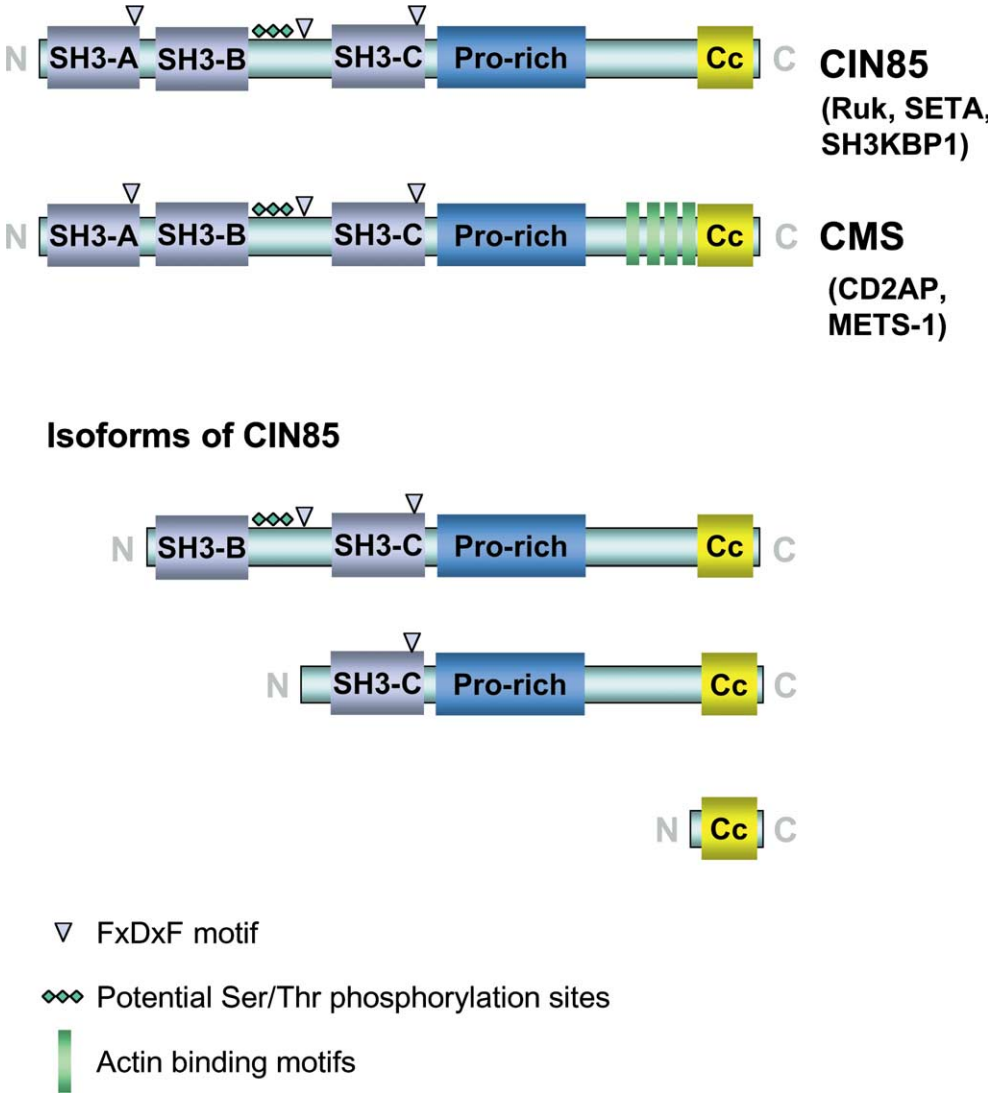

Fig. 1. Schematic representation of the domain structure of CIN85 and CMS. CIN85 and CMS contain three N-terminal SH3 domains, a centrally located proline-rich motif and a C-terminal coiled-coil domain. CMS additionally contains four actin binding motifs at the C-terminus. Alternative splice forms of CIN85 lack one or two SH3 domains or contain only the isolated coiled-coil domain. CIN85/CMS proteins were isolated in human, rat and mouse, while there are no orthologues found in *D. melanogaster*, *C. elegans* or yeast cells.

containing proteins. The carboxyl-terminal half of these proteins is predicted to be globular, and the last 50 amino acid residues show high propensity for coiled-coil structure. Coiled-coil structures are implicated in homotypic and heterotypic protein interactions [16]. In addition, CMS, but not CIN85, contains four putative actin binding sites in its carboxyl-terminus that are similar to the LKKTET motifs found in a number of actin binding proteins [13].

Due to their sequence and structural similarities CIN85 and CMS/CD2AP form a new family of adaptor proteins. Since CIN85 and CMS are the names of the human genes, this family was designated CIN85/CMS. Interestingly, the CIN85/CMS family is not phylogenetically conserved as there are no CIN85/CMS orthologues found in *Drosophila melanogaster*, *Caenorhabditis elegans* or yeast.

3. Proteins interacting with CIN85 and CMS

There is a rapidly growing list of proteins reported to interact with CIN85 and CMS (Table 1). Most of them were isolated using a yeast two-hybrid system and were subsequently confirmed for their interactions in vivo. Several groups have shown that the SH3 domains of CIN85 or CMS directly bind to members of the Cbl family of ubiquitin ligases [6,7,17], CD2 receptors [9,12], B-cell linker protein BLNK [18], apoptosis regulator AIP1 [11] and SETA binding protein SB1 [9]. The optimal binding site for SH3 domains of CIN85/CMS has not yet been determined. The first SH3 domain of CMS specifically binds to the type II PxxP motif present in the CD2 receptor [12]. On the other hand, multiple SH3 domains of CIN85/CMS are required for binding to the distal carboxyl-terminus of Cbl [6,7,17]. Interestingly, their binding was increased following tyrosine phosphorylation of Cbl in cells stimulated with growth factors [6,7,17], suggesting that phosphorylation of Cbl induces a conformational change of the protein, thereby unmasking putative SH3 binding motifs able to interact with CIN85/CMS. In accordance, the association of CMS with CD2 receptor was also induced upon T-cell activation [12]. Therefore, interactions between SH3 domains of the CIN85/CMS family and their targets

may be regulated in the cell by ligand stimulations. This is a novel characteristic of SH3 domain binding since it is generally believed that SH3 domains bind constitutively to polyproline motifs. There are currently no available data on whether interactions between CIN85 and BLNK, SB1 or AIP1 are also inducible by external signals [9,11,18,19]. More recently, a minimal binding domain in the distal part of Cbl and Cbl-b that is necessary for association with CIN85 was identified [20]. Interestingly, binding of CIN85 to this domain is not mediated via classical PxxP motifs [20]. Identification of the consensus binding motif for the CIN85/CMS SH3 domains and structural determination of their interactions would provide a molecular explanation for the mechanism by which CIN85 and CMS act as inducible scaffolding proteins.

Numerous SH3 domain-containing proteins were shown to be constitutively associated with proline-rich regions of CIN85/CMS (Table 1). Many of these proteins including Grb2, p130Cas, Crk and p85 phosphatidyl inositol-3 (PI-3) kinase are adaptor molecules linked to protein tyrosine kinase pathways [8,9,13,18]. CIN85 binding to the SH3 domain of the p85 PI-3 kinase inhibits its kinase activity and may thus block PI-3 kinase survival pathways in neuronal cells [8]. CMS was also shown to bind to Src family kinases and to co-localize with p130Cas in membrane ruffles and leading edges of cells; these interactions were implicated in dynamic regulation of the actin cytoskeleton [13]. More recently, two independent groups have shown that the endophilin family of accessory endocytic proteins constitutively binds to CIN85 and mediates internalization of RTKs. [5,6]. The interaction was mediated by the SH3 domain of endophilin bound to PKKPPPP and PKKPRPP motifs present in a proline-rich region of CIN85/CMS [5]. CIN85 and CMS were also shown to be homodimerized via their coiled-coil domains [9,13,18]. Finally, there is a list of kidney-specific proteins including nephrin, podocin and polycistin-2 that associate with the carboxyl-terminus of CMS [14,19,21,22].

4. Receptor clustering and formation of immunological synapse

The original discovery of CD2AP indicated its role in CD2 receptor clustering and cytoskeletal polarity in the contact area between T-cells and antigen-presenting cells [12]. This specialized junctional structure is conceptually analogous to a neuronal synapse and is called 'immunological synapse' [12]. Biochemical studies have indicated that the first SH3 domain of CD2AP binds to a single polyproline motif found in the carboxyl-terminal tail of CD2 and that their association is increased after T-cell engagement [12]. CD2AP was proposed to cluster CD2 receptors leading to organization of the immunological synapse [12]. However, it is not yet understood how CD2AP can mediate clustering of CD2 receptors if one CD2AP binds to only one CD2 receptor. One plausible explanation is that CD2AP is homo-oligomerized (due to coiled-coil interactions) and that CD2 receptor phosphorylation may lead to a conformational change in its carboxyl-terminal tail, thus favoring binding of CD2AP proteins that could in turn aggregate CD2 receptors. Another likely scenario is that all three SH3 domains of CD2AP may bind to the CD2 molecule enabling one CD2AP to cluster three CD2 receptors. The first SH3 domain of CD2AP binds with high affinity to a polyproline peptide in CD2 receptors, while it has not been tested whether the other two SH3 domains can also bind to the same peptide with similar or different affinities [12]. Moreover, CIN85 also binds to CD2 and is highly expressed in T-cells indicating that both proteins may have redundant roles in controlling CD2 function in the immunological synapse [9].

5. Building kidney architecture

A similar role for CD2AP was proposed in clustering a kidney-specific nephrin receptor [19,23]. Nephrin and CD2 are both immunoglobulin superfamily receptors involved in organization of specialized cell adhesive structures called slit diaphragm in podocytes and immunological synapse in T-cells, respectively [12,19]. An interacting network of clustered nephrin molecules is believed to be critical for glomerular filtration via the slit diaphragm. Nephrin binds to CD2AP and co-localizes with CD2AP in the slit diaphragm area [19,21,24]. A novel domain in the carboxyl-terminus of CD2AP was shown to associate with the cytoplasmic portion of nephrin [21]. Both proteins are also present in lipid rafts as well as linked to the actin cytoskeleton [23]. It was suggested that the main function of CD2AP is to anchor nephrin to the cytoskeleton, while other associated proteins may be involved in their clustering [19]. For example, a scaffolding protein

Table 1
CIN85/CMS binding partners

Protein	Interacts with	Function	Reference
c-Cbl	SH3-ABC of CIN85/CMS	Downregulation of RTKs	[6,7,17]
Cbl-b	SH3-ABC of CIN85	Downregulation of RTKs	[20]
BLNK: B-cell linker protein	SH3-ABC of CIN85	B-cell receptor signaling	[18]
SB1 (similar to NY-REN-45)	SH3-ABC of CIN85	Not yet defined	[9]
CD2	SH3-B of CD2AP and CIN85	T-cell receptor clustering. T-cell polarization	[9,12]
AIP1/Alix	SH3-B of CIN85	Apoptosis in glial cells	[11]
p85 subunit of PI-3 kinase	Proline-rich region of CIN85	Negative regulation of PI-3 kinase. Induction of apoptosis in neuronal cells	[8]
Grb2	Proline-rich region of CIN85	Regulation of RTK signaling	[9,13,18]
p130Cas	Proline-rich region of CMS/CIN85	Regulation of the actin cytoskeleton	[13,18]
Fyn, Src, Yes,	Proline-rich region of CMS	Regulation of Src family kinases	[13]
Endophilins A1, A2 and A3	Proline-rich region of CIN85	Regulation of RTK internalization	[5,6]
Nephrin	CD2AP C-terminus	Structural organization of kidney podocytes	[19,21]
Polycystin-2	CMS/CD2AP C-terminus	Maintenance of renal tubular structure	[14]
Podocin	CD2AP	Kidney glomerular architecture	[22]
CIN85/CMS	Coiled-coil region of CIN85/CMS	Homodimerization of CIN85/CMS	[9,13,18]
α-ear of AP2	FxDxF region of CIN85/CMS	Regulation of clathrin-mediated endocytosis	[15]

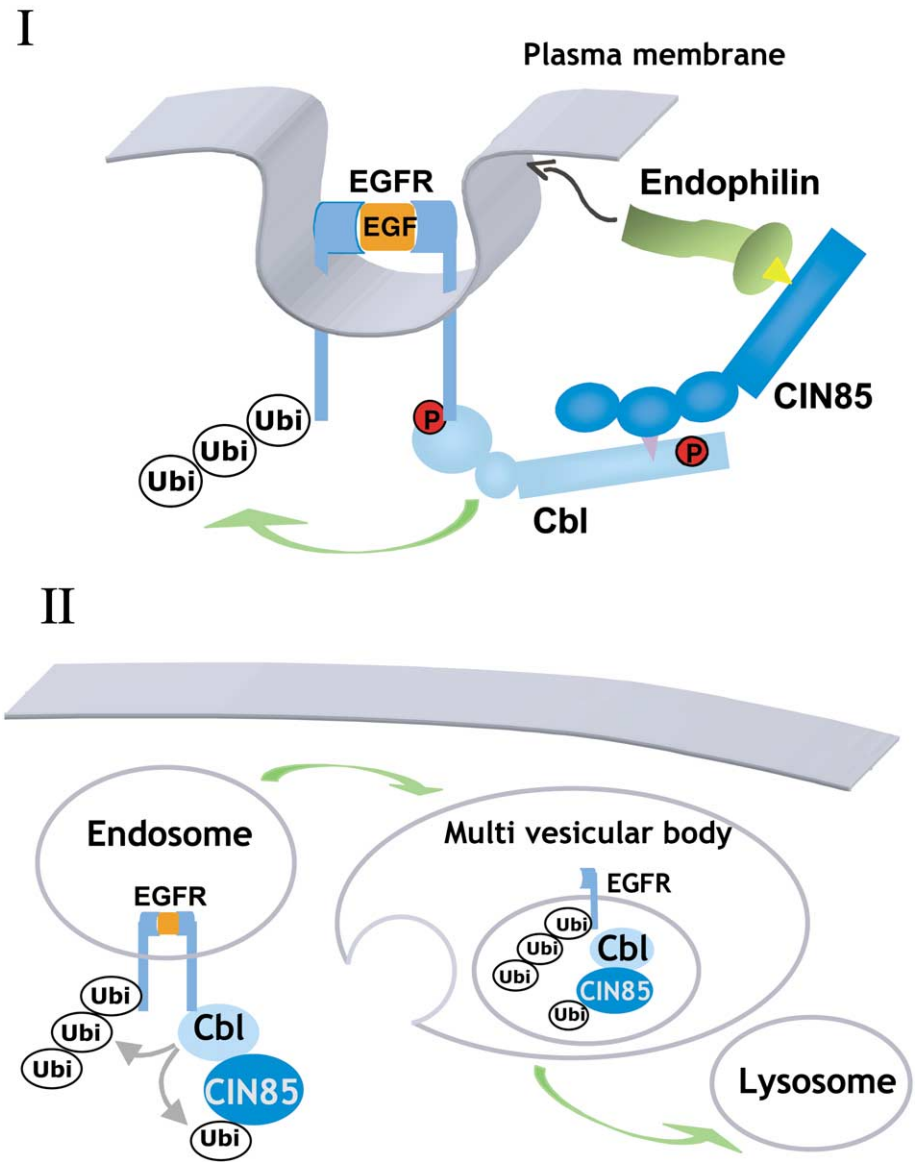

Fig. 2. Proposed role of CIN85/CMS in RTK endocytosis. I: Upon ligand-induced activation of EGF receptors, Cbl binds to phosphorylated receptors and promotes receptor ubiquitination (Ubi). Cbl is also tyrosine-phosphorylated in this complex leading to translocation of CIN85/endophilin in the vicinity of active EGF receptors, whereby endophilins in concert with dynamin and amphiphysin regulate clathrin-mediated internalization. II: CIN85 and CMS are also monoubiquitinated by Cbl in these complexes and are implicated in sorting of EGF receptors along the endocytic pathway for lysosomal degradation.

podocin is present in high-order oligomers in lipid rafts of the slit diaphragm and associates with both CD2AP and nephrin, thus promoting their aggregation [22]. Functional significance of these interactions is emphasized by phenotypes observed in CD2AP-deficient mice. They exhibited gross defects in glomeruli function leading to a premature death from renal failure [19]. T-cell proliferation and functions were also severely impaired [19]. In accordance, mutations in the nephrin gene cause a congenital nephrotic syndrome in humans [25]. CD2AP binds also to polycystin-2, a large transmembrane protein thought to be involved in cell–matrix adhesions as well as ion channel functions [14]. Taken together, it appears that interactions of CD2AP with nephrin, podocin and polycystin-2 may play important roles in building kidney architecture and maintaining its specialized functions.

6. Receptor endocytosis

Downregulation of RTKs is a critical step in modulating their activity [3]. This is a highly ordered and dynamic process controlled by the assembly of a large network of RTK-associated proteins as well as their post-translational modifications including phosphorylation and ubiquitination [3]. The Cbl family of ubiquitin ligases plays a major role in receptor ubiquitination, receptor sorting for degradation and ultimately in cessation of receptor-induced signal transduction [4].

Several lines of evidence support a role of CIN85/CMS in the regulation of Cbl-directed RTK downregulation (Fig. 2). CIN85 and CMS are rapidly recruited in Cbl–RTK complexes after growth factor stimulation [5,6,17,20]. In that way, CIN85 positions pre-associated endophilins in close proximity

of internalizing receptors [5,6]. Endophilins are regulatory components of clathrin-coated vesicle that could induce negative curvature and invagination of the plasma membrane during the early steps of RTK internalization [26–28]. This appears to be a general mechanism involved in internalization of multiple RTKs, including EGFR, PDGFR, c-Met and c-Kit [5,6,20]. CIN85/CMS may have additional roles in linking Cbl–RTK complexes with clathrin-coated endocytosis. One possible pathway, although not yet proven, includes binding of the AP2-α subunit appendage (α-ear) to FxDxF motifs found in the amino-termini of both CIN85 and CMS [15]. Different subunits of the AP2 adaptor complex bind to either internalization motifs found in RTKs, or to clathrin and accessory endocytic proteins [15]. It is possible that the dual binding of AP2 adaptor complex to RTKs and to CIN85/CMS may provide higher avidity interactions necessary to target and recruit preferentially activated RTKs into clathrin-coated vesicles.

The role of CIN85 in regulation of RTK downregulation is not only specific for clathrin-mediated internalization, but also participates in post-internalization processes such as endocytic trafficking and receptor degradation. CIN85 is co-localized with Cbl and activated RTKs in post-clathrin vesicles [5,6,20]. CIN85 is also targeted for common degradation with Cbl and EGFR in the lysosome [29]. Interestingly, CMS and CIN85 are monoubiquitinated by Cbl and Cbl-b in a trimeric complex with activated EGF receptors [29]. Monoubiquitination of proteins is a critical signal for endosomal sorting of proteins into multivesicular bodies and their targeting for degradation [30]. Several proteins involved in receptor endocytosis, including Eps15, Hrs and epsins, are monoubiquitinated following EGF stimulation [31]. Monoubiquitination of CIN85/CMS may therefore be a signal for their sorting to the lysosome, while non-ubiquitinated CIN85/CMS are recycled back to the cytoplasm [29]. More direct evidence for the role of CIN85 and CMS in regulating endosome functions is provided by data showing that overexpression of CIN85 and CMS leads to the formation of multiple vesicles in the cell [13,18]. These vesicles contain several markers of the endosomal compartment as well as Cbl and p130Cas proteins ([13] and Szymkiewicz and Dikic, unpublished data). Interestingly, deletion of the coiled-coil domain of CIN85 or CMS is sufficient to impair the formation of these vesicles ([13,18] and Szymkiewicz and Dikic, unpublished data). A coiled-coil fold exists in many endocytic proteins such as Hrs, epsins, Eps15, sorting nexins and others [16,32–34]. Similarly, deletion of coiled-coil domains in Hrs and sorting nexin 1 blocks the formation of endocytic vesicles upon overexpression of the respective proteins [32,33]. Although these results suggest that the coiled-coil domain of CIN85 is involved in sorting processes in the endosome, the molecular mechanism of its action is not yet understood. Cbl and CIN85/CMS are homo-oligomerized via their leucine zipper or coiled-coil domains, respectively [18,35]. Therefore, the increase in local concentrations of Cbl- and CIN85/CMS-anchored signaling complexes in the proximity of activated receptors may provide spatial and temporal co-ordination of critical steps in receptor endocytosis. The process of cargo delivery along the endosome may require polarization of vesicles as well as dynamic actin reorganization [36,37]. By its ability to bind both receptor cargo complex as well as components of the actin cytoskeleton CIN85/CMS may co-ordinate cross-talk between endocytic vesicles and the actin cytoskeleton. CIN85/CMS could also cluster and target Cbl–RTK complexes to one side of vesicles, thus providing sorting signals. This dual mode of action is similar to the role CMS plays in clustering of CD2 and nephrin receptors and anchoring them to specialized adhesion structures. This suggests a general mechanism by which CIN85 and CMS regulate functions of transmembrane receptors.

7. Conclusions and perspective

The CIN85/CMS family of adaptor proteins is an evolutionary divergent family of adaptor molecules with specific functions in higher eukaryotes. They are involved in formation of kidney glomeruli architecture, organization of specialized junctions in T-cells called immunological synapse and coordinating multiple steps in endocytosis of RTKs. A common mode of action has been revealed whereby CIN85/CMS proteins may regulate these processes by clustering transmembrane receptors and mediating dynamic interactions with the actin cytoskeleton. It will be interesting to test if CIN85/CMS play similar roles in organizing synapses or promoting synaptic vesicle endocytosis in neuronal cells. In the near future, we can expect a growing list of CIN85/CMS-interacting proteins that may provide a better understanding of their roles in regulation of endosome trafficking.

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References

[1] Pawson, T., Raina, M. and Nash, P. (2002) FEBS Lett. 513, 2–10.
[2] Schlessinger, J. (2000) Cell 103, 211–225.
[3] Waterman, H. and Yarden, Y. (2001) FEBS Lett. 490, 142–152.
[4] Thien, C.B. and Langdon, W.Y. (2001) Nat. Rev. Mol. Cell Biol. 2, 294–307.
[5] Petrelli, A., Gilestro, G.F., Lanzardo, S., Comoglio, P.M., Migone, N. and Giordano, S. (2002) Nature 416, 187–190.
[6] Soubeyran, P., Kowanetz, K., Szymkiewicz, I., Langdon, W.Y. and Dikic, I. (2002) Nature 416, 183–187.
[7] Take, H., Watanabe, S., Takeda, K., Yu, Z.X., Iwata, N. and Kajigaya, S. (2000) Biochem. Biophys. Res. Commun. 268, 321–328.
[8] Gout, I. et al. (2000) EMBO J. 19, 4015–4025.
[9] Borinstein, S.C., Hyatt, M.A., Sykes, V.W., Straub, R.E., Lipkowitz, S., Boulter, J. and Bogler, O. (2000) Cell Signal 12, 769–779.
[10] Narita, T., Amano, F., Yoshizaki, K., Nishimoto, N., Nishimura, T., Tajima, T., Namiki, H. and Taniyama, T. (2001) Cytogenet. Cell Genet. 93, 133–134.
[11] Chen, B., Borinstein, S.C., Gillis, J., Sykes, V.W. and Bogler, O. (2000) J. Biol. Chem. 275, 19275–19281.
[12] Dustin, M.L. et al. (1998) Cell 94, 667–677.
[13] Kirsch, K.H., Georgescu, M.M., Ishimaru, S. and Hanafusa, H. (1999) Proc. Natl. Acad. Sci. USA 96, 6211–6216.
[14] Lehtonen, S., Ora, A., Olkkonen, V.M., Geng, L., Zerial, M., Somlo, S. and Lehtonen, E. (2000) J. Biol. Chem. 275, 32888–32893.
[15] Brett, T.J., Traub, L.M. and Fremont, D.H. (2002) Structure (Camb.) 10, 797–809.
[16] Burkhard, P., Stetefeld, J. and Strelkov, S.V. (2001) Trends Cell Biol. 11, 82–88.
[17] Kirsch, K.H., Georgescu, M.M., Shishido, T., Langdon, W.Y., Birge, R.B. and Hanafusa, H. (2001) J. Biol. Chem. 276, 4957–4963.

[18] Watanabe, S., Take, H., Takeda, K., Yu, Z.X., Iwata, N. and Kajigaya, S. (2000) Biochem. Biophys. Res. Commun. 278, 167–174.
[19] Shih, N.Y., Li, J., Karpitskii, V., Nguyen, A., Dustin, M.L., Kanagawa, O., Miner, J.H. and Shaw, A.S. (1999) Science 286, 312–315.
[20] Szymkiewicz, I., Kowanetz, K., Soubeyran, P., Dinarina, A., Lipkowitz, S. and Dikic, I. (2002) J. Biol. Chem., in press.
[21] Shih, N.Y., Li, J., Cotran, R., Mundel, P., Miner, J.H. and Shaw, A.S. (2001) Am. J. Pathol. 159, 2303–2308.
[22] Schwarz, K. et al. (2001) J. Clin. Invest. 108, 1621–1629.
[23] Yuan, H., Takeuchi, E. and Salant, D.J. (2002) Am. J. Physiol. Renal Physiol. 282, F585–F591.
[24] Palmen, T., Lehtonen, S., Ora, A., Kerjaschki, D., Antignac, C., Lehtonen, E. and Holthofer, H. (2002) J. Am. Soc. Nephrol. 13, 1766–1772.
[25] Kestila, M. et al. (1998) Mol. Cell 1, 575–582.
[26] Farsad, K., Ringstad, N., Takei, K., Floyd, S.R., Rose, K. and De Camilli, P. (2001) J. Cell Biol. 155, 193–200.
[27] Ringstad, N., Gad, H., Low, P., Di Paolo, G., Brodin, L., Shupliakov, O. and De Camilli, P. (1999) Neuron 24, 143–154.
[28] Schmidt, A. et al. (1999) Nature 401, 133–141.
[29] Haglund, K., Shimokawa, N., Szymkiewicz, I. and Dikic, I. (2002) Proc. Natl. Acad. Sci. USA, in press.
[30] Hicke, L. (2001) Cell 106, 527–530.
[31] Polo, S. et al. (2002) Nature 416, 451–455.
[32] Raiborg, C., Bremnes, B., Mehlum, A., Gillooly, D.J., D'Arrigo, A., Stang, E. and Stenmark, H. (2001) J. Cell Sci. 114, 2255–2263.
[33] Zhong, Q. et al. (2002) Proc. Natl. Acad. Sci. USA 99, 6767–6772.
[34] Di Fiore, P.P. and De Camilli, P. (2001) Cell 106, 1–4.
[35] Bartkiewicz, M., Houghton, A. and Baron, R. (1999) J. Biol. Chem. 274, 30887–30895.
[36] Takenawa, T. and Itoh, T. (2001) Biochim. Biophys. Acta 1533, 190–206.
[37] Lanzetti, L., Di Fiore, P.P. and Scita, G. (2001) Exp. Cell. Res. 271, 45–56.