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Teneurins and Latrophilins: Two Giants Meet at the Synapse

Demet Araç^{1,2,*}, Jingxian Li^{1,2}

¹Department of Biochemistry and Molecular Biology, The University of Chicago, Chicago, IL 60637, USA

²Grossman Institute for Neuroscience, Quantitative Biology and Human Behavior, The University of Chicago, IL 60637, USA

Abstract

Teneurins and latrophilins are both conserved families of cell adhesion proteins that mediate cellular communication and play critical roles in embryonic and neural development. However, their mechanisms of action remain poorly understood. In the past several years, three-dimensional structures of teneurins and latrophilins have been reported at atomic resolutions and revealed distinct protein folds and unique structural features. In this review, we discuss these structures which, together with structure-guided biochemical and functional analyses, provide hints for the mechanisms of *trans*-cellular communication at the synapse and other cell-cell contact sites.

Keywords

Teneurin; ODZ; adhesion GPCR; Latrophilin/ADGRL; synapse; structure; alternative splicing; cellular communication; signaling

1. Introduction

Cells in multicellular organisms have the ability of adhering to each other and exchanging information. Teneurins/ODZs (TENs) and latrophilins/ADGRLs (LPHNs) are cell-surface molecules that mediate intercellular adhesion and communication at the synapse and at other cellular junctions (Figure 1) [1–13]. With four TEN (TEN1–4) and three LPHN (LPHN1–3) homologs in humans, they make up some of the most conserved and essential families of proteins. Genetic studies revealed critical roles for both TENs and LPHNs in embryonic development (especially of the brain) [3,14], and in neurobiology (especially in synaptogenesis, axon pathfinding, axon-dendrite partner matching and neural circuit-wiring) [15–18]; and linked them to numerous diseases including neurological disorders, developmental impairments, multiple types of cancers (such as glioblastoma and ovarian cancer), attention deficit/hyperactivity disorder and congenital general anosmia (the inability

*Corresponding author: Demet Araç: arac@uchicago.edu, Phone: (773) 834-2691, Fax: (773) 702-0439.

During the review of our manuscript, another paper reported that teneurin -SS isoform, when coexpressed with FLRT, induces excitatory synapse formation. Thus, postsynaptic latrophilins promote excitatory synapse formation by simultaneous binding of two unrelated presynaptic ligands [75].

Conflict of interest statement
Nothing declared.

to smell) [19–25]. Considering that many drugs target membrane receptors to regulate receptor function, and have excellent therapeutic benefits, TENs and LPHNs may be promising targets for drugs to treat numerous diseases once mechanistic details about the components that regulate their functions are understood.

TENs and LPHNs have large extracellular regions that form a tight *trans*-cellular complex [26,27]. The TEN/LPHN interaction likely perform a subset of the abovementioned physiological functions, whereas TENs and LPHNs individually have other functions independent of each other. In spite of the recent exciting advances, the mechanisms of TEN and LPHN action, and how they can mediate such broad range of functions remained majorly unknown partly due to lack of structural information on them. Recently, the structures of the large extracellular region of TENs and several structures of LPHNs in isolation and in complex with ligands revealed highly unusual structures and provided surprising insights into how TENs and LPHNs may perform their biological functions.

2. Teneurins

2.1. Teneurins have multiple essential functions in animals.

TENs are found in all multicellular organisms, including choanoflagellates, the eukaryotic unicellular organism that is often referred to as the origin of multicellularity [28]. Consistent with an ancient origin, TENs have multiple essential functions in animals that can be broadly categorized into functions in embryonic development and functions in the nervous system. During embryonic development, TENs are strongly expressed in various tissues including the heart and the brain [3,4,6–8,29]. In TEN mutant embryos, both of these tissues are severely malformed[3]. During brain development, TENs guide axons to the correct targets [15,16,30–33] and regulate synapse formation[34–36]. TEN perturbations cause synapse loss and impair synaptic organization *trans*-synaptically suggesting TENs act as bi-directional *trans*-cellular signaling molecules [5,15,16,26,27].

TENs have a large extracellular region that is involved in several homophilic and heterophilic interactions and mediate specific roles in different functions of TENs (Figure 2a) [34–36]. The strong heterophilic interaction of TENs with *trans*-cellular LPHNs regulates synapse formation and organization [26,27]. In mammalian cell culture, TEN2 interacts with LPHNs and mediates *trans*-cellular signaling to modulate cAMP levels in neighboring cells; supporting an important role for the TEN/LPHN interaction in cellular communication [37]. On the other hand, a *trans*-homodimerization function was suggested for TENs. Studies in *Drosophila* showed that the critical role of TEN in selecting the right targets between individual presynaptic and postsynaptic neurons is mediated by homophilic cell-adhesion of TENs [15,16]. Furthermore, TEN ECRs were reported to undergo multiple proteolytic events that may release soluble fragments, including a short C-terminal fragment called teneurin C-terminal associated peptide (TCAP) that stimulates neurons by an unknown mechanism [38,39]. These proteolytic events may convert TEN cell-adhesion molecules into diffusible signals, which could act during axonal pathfinding.

2.2. Teneurins have unusual domain architecture.

TENs are large type-II transmembrane proteins that are composed of an N-terminal cytoplasmic sequence, a single transmembrane region (TMR), and a large extracellular region (ECR) (Figure 2a) [3,7,40,41]. The ECR of TENs comprises eight epidermal growth factor (EGF) motifs. TENs form constitutive *cis*-dimers via highly conserved disulfide bonds formed between their EGF repeats in close proximity to the transmembrane pass [38,41,42]. The EGF motifs are followed by a large unknown region (>2000 amino-acids) that lacks the requisite domains generally observed in cell-adhesion molecules, such as classical cadherin-domain, LNS-domain, Ig-domain or integrin domain (Figure 2a). Even though TENs are highly conserved [28], sequence analyses have not revealed any identifiable domains in this region. The lack of information on the structure of TEN ECR has been one of the limiting factors in delineating their mechanisms of action.

A recent study by Li *et al.* showed negative stain electron microscopy images of the full-length extracellular region of human TEN2 ECR forming *cis*-dimerization of two large globular domains extended by thin flexible linkers that are tied together through a stalk-like region at their end and confirmed previous studies (Figure 2b) [37,41,42]. Deletion of the stalk-like region corresponding to the EGF repeats (grey) from this construct yield a soluble monomeric protein with a molecular mass of 215 kDa (ECR2-5). The structure of the monomeric TEN2 ECR2-5 was determined by single-particle cryo-EM at 3.1 Å resolution (Figure 2c–e) [37]. The structure revealed that TEN2 has an unusual domain architecture with no similarity to any other eukaryotic protein.

The TEN2 ECR assemble as a large cylindrical barrel composed of a three-layer spiral of β -hairpins and sealed at both ends (Figure 2c–e). The bottom of the barrel is capped with an Ig-like domain that extends to the inner barrel cavity, while a flexible six-bladed β -propeller domain is attached perpendicularly to the barrel bottom. The C-terminal part of the protein is partially encapsulated within the barrel and emerges to the solvent through an opening in the mid-barrel region. The external part of the C-terminal region adopts a stable fold and is anchored to the barrel outer surface. A similar fold arrangement was also observed for the chicken TEN2 and mouse TEN3 suggesting that this is a domain organization conserved between species and isoforms [43].

2.3. Teneurins have striking homology to bacterial Tc-toxins.

The fold of TEN2 has not been encountered in any eukaryotic protein. However, despite a limited protein sequence identity (12%), the TEN2 β -barrel has a striking structural similarity with the BC-components of bacterial Tc-toxins (Figure 2f–g) [44,45]. Tc-toxins are tripartite complexes composed of A, B and C protein components that are individually secreted from bacteria and assemble into a megadalton sized complex outside the producing cell [44–46]. The B and C components of Tc-toxins compose a large macromolecular barrel that encapsulates the covalently attached C-terminal toxin domain (Figure 2f–g). The C-component of Tc-toxins acts as an aspartyl protease and is capable of autoproteolytic processing of the toxin domain. The cleaved toxin domain travels through the barrel tunnel and is released into the host cell by an injection mechanism. In the host cell, it mediates its toxic effects via multiple mechanisms including DNA hydrolysis and transcriptional

regulation. The unusual similarity of TENs to bacterial toxins raises questions about the function of TENs.

Sequence alignment of TENs with the catalytic pocket region of bacterial toxins revealed a high level of conservation (Figure 2h–i), with all catalytic residues present in all TENs apart from the studied TEN2 variant, which carries a single aspartic acid to glycine alteration (D2345G) within catalytic residues [37]. Structural comparison between the pocket in TEN2 and the active catalytic pocket in bacterial toxins indicates that two conserved residues that were shown to mediate the catalytic activity in toxins (R2302 and D2315) are similarly oriented in both structures. However, the glycine residue in TEN2 is pulled away from the catalytic pocket, mainly due to several contacts of the encapsulated domain with the barrel inner cavity as well as with the Ig-like domain extension penetrating into the barrel. These observations suggest that TENs may undergo autoproteolysis to release the toxin-like domain and raised numerous questions and possibilities about how TENs function.

Interestingly, domain deletion experiments showed that toxin-like region of Ten mediates interaction of TEN with LPHN (Figure 3, left). LPHN, on the other hand, uses its Lectin domain to interact with TEN (Figure 3). However, the molecular details of the TEN/LPHN complex are not known. Furthermore, the TEN structure revealed the structural organization of the TCAP peptide within the TEN structure and positioned it on the toxin-like domain outside of the barrel. However, it is still unclear whether TEN functions similar to toxins or to peptide hormones.

2.4. Alternative splicing is a switch for ligand interactions and functions of TENs.

TENs are alternatively spliced at two canonical sites that are localized to the ECR and include nine-amino-acid and seven-amino-acid insertions at the EGF repeats and the β -propeller regions, respectively [29]. Alternative splicing within the β -propeller domain acts as a switch to regulate selective binding of TEN2 to adhesion partners, thus controlling the cellular functions TENs perform (Figure 3) [37]. The TEN variant that includes the seven-amino acid insertion (NKEFKHS) is unable to mediate *trans*-cellular adhesion with full-length LPHN presented on the neighboring cell (Figure 3, right side), whereas the variant without the insertion interacted with LPHN with high affinity (Figure 3, left side) [37]. The alternatively spliced site, however, localizes at least 8 nm away from the toxin-like region where LPHN binds, and, is thus unlikely to participate in any interactions with the LPHN lectin domain [37]. However, puzzlingly, both TEN isoforms can bind to the soluble lectin domain of LPHN. These observations altogether suggest that alternative splicing may facilitate a conformational change of the entire TEN ECR that makes the LPHN binding site inaccessible to the full-length LPHN on the neighboring cell (Figure 3). Future studies will reveal the structural basis for this intriguing phenomenon.

Further studies showed that the same seven-amino acid alternatively spliced site within the propeller domain also regulates TEN homophilic cell-adhesion and that TEN isoform with the splice insert (but not the one without) homodimerizes in *trans* in addition to in *cis* [17] (Figure 3, right side). However, the understanding of the molecular details of a *trans*-homodimeric interaction is in its infancy. The domains that mediate *trans*-homodimerization and how alternative splicing may regulate it are unstudied.

Unexpectedly, studies on the function of different splice variants of TENs showed that the TEN2 splice variant that is incapable of interacting with LPHN is a potent inducer of inhibitory synapses, indicating that it has functional impact on specific synapse subtypes through interactions with alternative binding partners that remain to be discovered (Figure 3, right side). Specifically, the TEN2 variant including the splice insert induced inhibitory postsynaptic differentiation, suggesting a LPHN-independent role for TEN in synapse formation. No excitatory presynaptic specialization was induced by splice isoforms of TEN, possibly because unknown co-factors may be required for excitatory but not inhibitory synapse formation. Overall, these results reveal that alternative splicing of TENs may serve as a general principle to regulate their diverse interactions with multiple adhesion partners, giving rise to the multifaceted roles they play. Future studies investigating developmental and spatial patterns of alternative splicing will generate important insights into the regulation of synapse formation and specification as well as embryogenesis and tissue development by TENs. The recent structure of TEN2 will form the structural framework to start delineating the diverse mechanisms of TEN function.

3. Latrophilins

3.1. Biological functions of latrophilins

LPHNs have roles in embryogenesis, tissue polarity and synapse development, interestingly almost identical to the functions of TENs [9–13,26,27,47,48]. LPHN1 was identified as the calcium independent receptor for α -latrotoxin, a black widow spider toxin that triggers massive neurotransmitter release from nerve terminals [9–13]. In *Caenorhabditis elegans* (*C. elegans*), LPHN is required for the alignment of cell division planes during development [49]. In vertebrates, the extracellular regions of LPHNs mediate excitatory synapse formation [50]. At present, among the many cell adhesion proteins involved in synapse formation, latrophilin-2 is the one whose deletion causes the strongest synapse-formation phenotype known [47]. LPHN mutations are linked to attention deficit hyperactivity disorder (ADHD) as well as numerous cancers in humans [51–53]. Given the co-expression patterns of LPHNs and TENs during development, and in the brain, it is possible that the heterophilic interaction of LPHNs with TENs mediates their respective functions during embryonic development and neural development.

3.2. Latrophilin is a model adhesion GPCR with a conserved GAIN domain

Latrophilins belong to the adhesion G-Protein Coupled Receptor (GPCR) family that contain the typical 7-transmembrane (TM) domain similar to canonical GPCRs (Figure 4a). Biochemical and structural work on LPHNs led to the better understanding of how adhesion GPCRs function. Unlike other GPCRs, adhesion GPCRs also have large extracellular regions (ECRs) that are autoproteolytically cleaved from their 7TM regions. Autoproteolysis occurs during biosynthesis, resulting in a fractured adhesion GPCR that nevertheless remains tightly associated at the cell surface [9,10]. A recent report provided pivotal details by discovery and analysis of a novel and evolutionarily conserved domain that mediates the autoproteolysis function of adhesion GPCRs (Figure 4a) [54]. This novel domain, termed the GPCR Autoproteolysis INDucing (GAIN) domain fine-tunes the chemical environment to catalyze peptide bond hydrolysis at a GPCR Proteolysis Site (GPS) (Figure 4b) [9,54–57].

Strikingly, the GAIN domain is the only extracellular domain shared by all human adhesion GPCRs, and thus, together with the TM domain constitutes the core of all adhesion GPCRs. Furthermore, the GAIN domain is evolutionarily conserved from tetrahymena to mammals and is the locus of multiple human disease mutations [54]. The cleaved and uncleaved GAIN domain structures revealed that autoproteolysis site is between the last two β -stands of the GAIN domain [54]. Autoproteolysis does not lead to dissociation of the cleaved peptide because the cleaved peptide is hidden within the core of the GAIN domain. The discovery of the GAIN domain was a breakthrough as it redefined the poorly studied aGPCR family, showing that members of this family share a large, unique and widespread autoproteolytic domain that may be involved in downstream signaling [54]. This prediction was proven correct by recent reports.

A current model suggests that ligand binding to the ECR and exerting force can cause dissociation at the autoproteolysis site, removing most of the ECR and revealing a tethered peptide agonist (the last β -strand of the GAIN domain) which can then bind to and activate the receptor [58–61]. A second model for aGPCR activation suggests that transient interactions between the ECR and 7TM directly regulate receptor signaling [62–65]. Synthetic proteins that bind to the adhesion GPCR ECRs including the GAIN domain have been shown to modulate receptor signaling via this mechanism [65]. These two models are not mutually exclusive and current data suggests that adhesion GPCRs can be activated by both mechanisms and maybe by other mechanisms as well. Both mechanisms can be biologically important in different contexts; for instance, the former is an irreversible mechanism that might induce a large cellular response similar to protease activated receptors (PARs), whereas the latter is a reversible mechanism that modulates receptor signaling in a subtle manner similar to most GPCRs.

In addition to the GAIN domain, LPHNs have an N-terminal lectin domain, an olfactomedin domain, a serine/threonine-rich region and a hormone-binding domain (HormR) in their ECR. (Figure 4a) [10,54,66,67]. The structures of all individual domains are available (Figure 4b–c) [54,68,69]. The multi-domain ECR is a protein-protein interaction surface for numerous other proteins. In addition to TENs, the ECRs of LPHNs interact with Fibronectin leucine-rich repeat transmembrane (FLRTs) proteins [50], with neurexins [70] and with α -Latrotoxin [10] (Figure 1). However, the roles of these interactions in LPHN function are yet to be studied.

3.3. Heterotetrameric and trimeric structures of latrophilins with FLRTs and UNC5s.

The interaction of LPHNs with FLRTs mediate excitatory synapse formation [50]. This interaction occurs between the olfactomedin domain of LPHNs and the LRR repeats of FLRTs. FLRTs are essential cell-adhesion molecules in cortical development and synapse formation. FLRTs additionally interact with UNC5/Netrin receptors to control the migration of neurons in the developing cortex (Figure 1) [50,71]. Moreover, FLRT proteins interact with each other and form cis-dimers on the cell surface although previous studies had reported a homotypic cell adhesion function. The involvement of the LRR repeats of FLRTs in numerous heterophilic interactions suggest a central role for FLRTs in neural development and raises the necessity to identify the specific binding sites for each of LRR

domain interactions (such as LPHN, UNC5 and homophilic FLRT binding sites on FLRT) in order to unambiguously dissect the role of each interaction for FLRT function.

Several recent structures together with biochemical analysis revealed the structural organization and competing and non-competing interactions of LPHNs and FLRTs. The structure of isolated FLRTs showed that the FLRT LRR repeats dimerize sideways in a parallel fashion whereby the N-termini of each protomer face one direction and the C-termini face the other (Figure 4d) [72]. The same dimer interface is observed in mouse FLRT3, human FLRT3 structures as well as two FLRT/LPHN complex structures, all of which were crystallized in different space groups [68,72,73], whereas a previous study reported a different FLRT dimerization interface [73].

The structure of the FLRT3/LPHN3 complex revealed that the olfactomedin domain binds to the concave surface of the FLRT3 LRR horseshoe (Figure 4e) [68,72]. This surface is spatially separated from the FLRT dimerization site, and thus two LPHN3 molecules bind to the FLRT dimer forming a heterotetramer (Figure 4e) [72]. The LPHN/FLRT interaction surface spreads over the extensive complementary surfaces of both proteins. The major interaction hotspot is at the N-terminal side of the LRR horseshoe and at the long loops emerging from the second and third blades of the LPHN3 olfactomedin beta-propeller (Figure 4e). Mapping of the mutations of LPHN3 genes that are linked to attention deficit/hyperactivity disorder and cancers on the FLRT3/LPHN3 complex structure revealed that most mutations are at the FLRT3/LPHN3 binding interface where the two proteins come closest to each other suggesting protein-protein interaction defects.

Crystal structures and biophysical experiments showed that the LPHN binding surface on FLRT is also spatially separated from the UNC5 binding site on FLRT3 (Figure 4f) [72,73]. Thus, LPHN3 and UNC5 can simultaneously bind to FLRT3, forming a trimeric complex (Figure 4f) [72]. FLRT3 may form transsynaptic complexes with both LPHN3 and UNC5 [72]. Moreover, UNC5 binding site overlaps with the FLRT dimerization interface, thus FLRT3 homodimerization and FLRT/UNC5 interaction are incompatible (Figure 4f) [72]. As FLRT dimerization has weaker affinity than FLRT/UNC5 interaction, the presence of UNC5 may break the FLRT dimer and the FLRT/LPHN heterotetramer in solution [72] and in the crystal [74]. How these interactions may be regulated in physiological conditions on the cell membrane is unknown and further studies will reveal their physiological roles. On the other hand, the FLRT3 binding site on LPHN3 is distant from the teneurin binding site on LPHN3 as teneurin binding is largely mediated by the LPHN lectin domain, which is N-terminal to the olfactomedin domain (Figure 4e) [72]. It is thus likely that LPHN can interact with TEN and FLRT simultaneously, introducing further complexity to the *trans*-cellular interactions at the synaptic junction (Figure 4 and Figure 5).

4. A combinatorial code generated by a mesh of interactions at the synapse.

Neural development, similar to other cellular communication induced processes, is a complex phenomenon that is mediated by the coordinated interactions of numerous cell-surface proteins on neurons and glial cells. Structural and functional studies summarized in

this review suggest that the interaction network of teneurins, latrophilins and their other ligands generate a combinatorial code of protein-protein interactions that dictates downstream functional events such as synapse formation. The presence or absence of certain proteins and their certain splice isoforms likely mediates various specific outcomes. As the complexity of complexes increase, understanding how the mesh of protein interactions are regulated at the structural level will help reveal how distinct *trans*-cellular signals are transduced at the synapse.

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Figure 1: Schematic diagram of the interaction network between teneurin, latrophilin and several other proteins at the synapse.

Figure shows the diagram of TEN, LPHN, FLRT3 and UNC5 at a synaptic junction. Teneurin and FLRT are localized on the presynaptic cell membrane, while LPHN and UNC5 are localized on the postsynaptic membrane. Conditional genetic KOs established that in cultured neurons and *in vivo*, at least Lphn2 is exclusively postsynaptic [47]. Teneurin-2 is positioned presynaptically because it forms a *trans*-cellular junction with postsynaptic latrophilins [27] and also because it induces postsynaptic specializations in the artificial synapse formation assay [37]. FLRT3 is also positioned presynaptically because it forms

trans-cellular complexes with Lphn2 [72]. Note that latrophilins were originally thought to be presynaptic and FLRTs postsynaptic based on RNAi data [50].

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Figure 2: TENs structure have similarity to bacterial toxins.

(a) Domain composition of human TEN2. Extracellular domains are colored grey, yellow, green, blue and magenta for domains 1-5, respectively; transmembrane region (TM) in brown. Domain numbers and descriptions are indicated in black above scheme. (b) Representative TEN2 dimers from negative stain EM micrographs of TEN2 ECR(1-5) indicating *cis*-dimerization through the EGF repeats (domain 1). Yellow arrows indicate EGF repeat *cis*-dimerization. (c-e) Cryo-EM structure of TEN2 ECR(2-5) from different views. (f and g) The TEN2 domain 4 (blue) assembles into a hollow barrel that resembles

the BC components of bacterial Tc-toxins (TcC-toxin, orange, PDB: 4O9X). Side (f) and top (g) views indicate similar radial dimensions with a significantly enlarged volume in the bacterial toxin barrels compared to TEN. **(h)** Multiple sequence alignment of TENs with bacterial Tc-toxins reveals similarity with an aspartate protease domain localized within the barrel. Conserved residues that were shown to mediate catalytic activity in bacteria are highlighted by arrows. Putative cleavage site is shown in red dashed line. Corresponding residue numbers in TEN2 and in TcC are shown in blue and orange, respectively. **(i)** Three-dimensional representation of TEN residues with suspected catalytic activity. The cleavage site is indicated by arrow. Figure modified from reference [37].

Figure 3: Model for alternative-splice-dependent functions of TEN2.

The model depicts how alternative splicing acts as a molecular switch to determine which adhesion partner TEN2 binds to, and, accordingly, which cellular functions TEN2 mediates. On the left, the TEN2 variant lacking the β -propeller splice insert (empty star) interacts with LPHN and modulates cAMP levels in the neighboring cell. On the right, TEN2 variant including the splice insert (full star) is unable to interact with LPHN, but it forms *trans*-homodimers to mediate neural circuit-wiring and induce inhibitory synapses by interacting with unknown ligands. The left and right sides of the TEN2 dimer represent various cell-cell junctions and inhibitory synapses, respectively. The teneurin structure[37] (PDB ID: 6CMX), membranes and distance between synaptic membranes are drawn to scale. The molecules on the post-synaptic side are drawn as schemes and are not to scale. Alternative splice site is shown by red star.

Figure 4: Latrophilin and its heterotetrameric and trimeric complexes with FLRTs and UNC5s.

(a) Schematic domain diagram of LPHNs as a model aGPCR. All aGPCRs have a GAIN domain, a TM domain, and variable other domains. The GAIN domain is coloured pink, HormR domain light blue, olfactomedin domain yellow and Lectin domain gray. The cleaved last beta-strand of the GAIN domain is a tethered agonist. Red line indicates tethered agonist peptide and red star indicates cleavage site. (b-d) Cartoon representation of the crystal structure of the HormR and GAIN domains of LPHN1 (PDB: 4DLQ) (b), Lectin and olfactomedin domain of LPHN3 (PDB: 5AFB) (c), and FLRT3 dimer (PDB: 5CMN) (d). The protomers of the FLRT3 dimer are colored orange and cyan. (e-f) Model for possible rearrangement of interactions involving LPHN at the synapse. (e) Structures of LPHN3/FLRT3 complex (PDB: 5CMN, 4YEB) show two LPHN3 molecules (one LPHN is colored pink, the other LPHN is colored as in a) bind to a FLRT dimer (colored orange and cyan) and form a heterotetramer between the two synaptic membranes. (f) In the presence of UNC5, UNC5 binds to the FLRT dimerization interface on FLRT with high affinity and breaks the weak FLRT dimer. FLRT, LPHN and UNC5 form a trimeric complex [72] (PDB IDs: 5CMN, 4V2C, 5FTU). The physiological consequences of this regulatory interactions

are yet to be studied. The TM domain of LPHN is modeled using the corticotrophin-releasing hormone-receptor structure (PDB: 4K5Y, green).

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Figure 5. High resolution structures of the trans-synaptic teneurin, latrophilin, FLRT and UNC5 Complex.

Structures determined for Lphn3 (pink), TEN2 (green) FLRT3 (orange) and Unc5 (blue), are placed into synaptic cleft of an excitatory synapse. Interestingly, presynaptic FLRT3 can bind simultaneously to both postsynaptic Lphn3 and postsynaptic Unc5, as shown by biochemistry and crystallography of these complexes [72]. Arrows indicate interactions. It is unknown whether Lphn3 can simultaneously bind to FLRT3 and to teneurins, or whether these interactions are mutually exclusive, but in either case the various binding reactions suggest that a *trans*-synaptic interaction network anchored on latrophilins and teneurins may form the basis for an extensive signaling machinery at the synapse. Structures are from Ref. [72] for the FLRT3/Unc5/Lphn3 complex, [54] for the Lphn3 GAIN/hormone binding domain fragment, and [37] for teneurin (PDB IDs: teneurin-2, 6CMX; Lphn3, 4DLQ [GAIN and hormone-binding domains], 5CMN/5AFB [lectin and olfactomedin domains] and 4K5Y [transmembrane GPCR domain of the corticotrophin releasing hormone receptor]; FLRT, 5CMN; Unc5, 5FTT). The model is drawn to scale with the exception of EGF repeats of TEN that span a longer distance and the stalk region of LPHN. The interaction network likely spans the synaptic cleft in 3D rather than 2D that this figure is limited to.