Published in final edited form as:

Ageing Res Rev. 2014 September ; 0: 34–42. doi:10.1016/j.arr.2014.03.004.

Non-coding RNA regulation of synaptic plasticity and memory: implications for aging

Laurie R. Earls^{*}, Joby J. Westmoreland, and Stanislav S. Zakharenko^{*}

Department of Development Neurobiology, St. Jude Children's Research Hospital, Memphis TN

Abstract

Advancing age is associated with the loss of cognitive ability and vulnerability to debilitating mental diseases. Although much is known about the development of cognitive processes in the brain, the study of the molecular mechanisms governing memory decline with aging is still in its infancy. Recently, it has become apparent that most of the human genome is transcribed into non-coding RNAs (ncRNAs) rather than protein-coding mRNAs. Multiple types of ncRNAs are enriched in the central nervous system, and this large group of molecules may regulate the molecular complexity of the brain, its neurons, and synapses. Here, we review the current knowledge on the role of ncRNAs in synaptic plasticity, learning, and memory in the broader context of the aging brain and associated memory loss. We also discuss future directions to study the role of ncRNAs in the aging process.

Keywords

Non-coding RNA; microRNA; synaptic plasticity; learning; memory; aging

1. Introduction

Our memories are at the core of who we are as individuals. Loss of memory during normal aging or as the result of disease erodes quality of life. The effect of aging and disease on memory presents a moving target for neuroscience research, requiring not only the investigation of mechanisms that underlie memory but also their study in the context of dynamic developmental and senescent processes. This endeavor has the potential to address many questions important for basic neuroscience and its clinical applications. For example, why do some people's brains age well, whereas others' minds decline too early? Also, why does the brain become vulnerable to certain diseases only at certain ages?

© 2014 Elsevier Ireland Ltd. All rights reserved.

^{*}Correspondence should be addressed to Stanislav S. Zakharenko (stanislav.zakharenko@stjude.org) or Laurie R. Earls (laurie.earls@stjude.org), Department of Developmental Neurobiology, MS 323, St. Jude Children's Research Hospital, 262 Danny Thomas Place, Memphis TN 38105-2794. Tel: 1-901-595-3837.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

We are at the threshold of unraveling the mechanisms of molecular aging in the brain. The discovery of non-coding RNAs (ncRNAs) is changing our view of the mechanisms that influence how neurons form, operate, and age. In recent decades, we have learned much about synaptic plasticity as a mechanism of learning and memory, and we must now incorporate ncRNAs into this knowledge. This review examines the current knowledge about ncRNAs in synaptic plasticity and memory and discusses future directions for the study of ncRNAs in the aging process.

Early studies have revealed that the brain alters its function throughout different developmental stages of an organism. Age-related changes in the expression of genes related to synaptic plasticity may contribute to memory decline and vulnerability to cognitive diseases. For instance, brain-derived neurotrophic factor (BDNF) is involved in mechanisms of long-term potentiation (LTP), a major form of synaptic plasticity (Figurov et al., 1996; Ji et al., 2010). BDNF is downregulated with age in rat hippocampus, and its depletion is associated with a decline in synaptic plasticity (Zeng et al., 2011).

The response of the brain to neurochemicals can also vary with age. For example, an increase in the atypical amino acid D-aspartate is beneficial for memory in adult mice at 4–5 months. However, the same increase is detrimental to memory in older mice (Errico et al., 2008; Errico et al., 2011). These findings suggest that the brain can be responsive to some molecules only during specific developmental windows, and that these molecules may be inconsequential or detrimental during other phases of life.

BDNF and D-aspartate are only two examples of factors that affect memory in an age-dependent manner. The broader picture is a complex epigenetic hierarchy that orchestrates the expression of a multitude of memory-related genes in a dynamic fashion throughout life. Histone acetylation is one example of such age-dependent epigenetic regulation (Day and Sweatt, 2011; Maze et al., 2013; Zovkic et al., 2013). In the young brain, histone acetylation occurs in response to learning, resulting in global changes in gene expression (Korzus et al., 2004; Levenson et al., 2004). However, learning-dependent histone acetylation is reduced in the aged brain, thereby resulting in reduced responsiveness of learning-related genes (Peleg et al., 2010).

One important area of research that has received relatively little attention in neuroscience is the control of neuronal processes by ncRNAs. By recent estimations, ~98% of the human transcriptome consists of predicted non-coding transcripts rather than protein-coding mRNAs (Mattick, 2005; Furuno et al., 2006; Ravasi et al., 2006). Numerous ncRNAs are enriched in the central nervous system (CNS) (Mercer et al., 2008), functionally conserved, and often tissue specific (Cabili et al., 2011; Derrien et al., 2012; Ulitsky et al., 2011), suggesting important roles for ncRNAs in the brain. Emerging evidence suggests that ncRNAs have important roles in synaptic plasticity, learning, and memory. In this review, we summarize recent findings on synaptic plasticity- and memory-related ncRNAs and attempt to integrate some of this research with our limited knowledge of ncRNAs in aging.

2. Classes of ncRNAs

ncRNAs are classified into different groups based on their biogenesis, size, and biologic function. These include microRNAs (miRNAs), long non-coding RNAs (lncRNAs), short non-coding RNAs (sncRNAs), Piwi-interacting RNAs (piRNAs), and circular RNAs (circRNAs). The ultimate function of most ncRNAs studied to date is the regulation of protein expression. However, the mechanisms by which different ncRNAs execute this function range from chromatin modification and DNA methylation to directly inhibiting translation of target mRNAs. miRNAs are ~22 nucleotides (nt) long, directly inhibit protein translation, and cause mRNA degradation through an RNA–protein complex known as the RNA-induced silencing complex (RISC) (Czech and Hannon, 2011). lncRNAs and sncRNAs are distinguished by their sizes, with transcripts longer than 200 nt designated as lncRNAs and those 200 nt or shorter defined as sncRNAs (Ng et al., 2013). Both lncRNAs and sncRNAs exhibit diverse functions (see below) (Barry et al., 2013; Cao et al., 2006; Gong and Maquat, 2011; Jeon and Lee, 2011; Osato et al., 2007). circRNAs, presumably formed as a byproduct of splicing, were originally thought to be degraded, but are now believed to persist and function in the cell. circRNAs have only recently been described, and the extent of their functions remains unknown. However, it has been reported that a subset of circRNAs inhibit miRNAs by acting as endogenous miRNA sponges (Hentze and Preiss, 2013). piRNAs are small (~24–34 nt) ncRNAs that were originally found to be required for germ cell development (Peng and Lin, 2013; Malone and Hannon, 2009a; Malone and Hannon, 2009b; Thomson and Lin, 2009). piRNAs can regulate protein expression by interacting with the epigenetic silencing machinery and by translational regulation of mRNAs (Juliano et al., 2011; Peng and Lin, 2013).

3. Synaptic localization of ncRNAs

A large portion of the miRNAs discovered in mammals is expressed in the CNS (Kosik, 2006). A similar enrichment of lncRNAs in the CNS has also been shown (Qureshi and Mehler, 2012), indicating an important role for ncRNAs in the brain. In addition, many ncRNAs are expressed in specific brain regions and/or at specific times in development (Krichevsky et al., 2003; Miska et al., 2004; Sempere et al., 2004), suggesting that the non-coding transcriptome has an important role in the developmental and functional complexity of the brain. A subset of neuronal miRNAs is enriched in synaptic structures. A microarray comparison of synaptosomes (biochemically isolated synapses) with whole-tissue lysates from adult mouse forebrains detected 178 miRNAs, 37 of which were enriched in synapses (Lugli et al., 2008). However, in juvenile mice, a similar study found only 10 miRNAs to be synaptically enriched (Siegel et al., 2009). Interestingly, miRNAs enriched in synapses of juveniles were different from those enriched in the synapses of adults. This finding suggests that there are distinct synaptic complements of miRNAs at different stages of life. Synaptic enrichment of natural antisense RNAs has also been reported (Smalheiser et al., 2008). These are a subgroup of lncRNAs that are transcribed from the antisense strand of coding or non-coding genes. While these natural antisense RNAs can have multiple regulatory roles in the nucleus, synaptic antisense RNAs most likely work by pairing with and regulating sense transcripts (Faghihi and Wahlestedt, 2009).

Even though many of the identified synaptic ncRNAs are presumed to be postsynaptic, presynaptic localization of miRNAs has also been reported. Using Campenot chambers, which physically isolate the axon from the rest of the neuron, Natera-Naranjo et al. identified 130 miRNAs that localized to the axons of rat superior cervical ganglia neurons (Natera-Naranjo et al., 2010). A similar study profiling the expression of axonal miRNAs from mouse cortical neuronal “ball” cultures showed that approximately 100 miRNAs were localized to axons, several of which were enriched in growth cones (Sasaki et al., 2013).

While many miRNAs are now thought to reside in both the presynaptic and postsynaptic compartments of the synapse, similar nonbiased screening for synaptic localization of other types of ncRNAs has not yet been performed. Because some types of ncRNAs were originally characterized in the nucleus, there is currently an expectation in the field that other ncRNAs are nuclear rather than synaptic. However, early studies on brain cytoplasmic RNAs (BC RNAs) supported a role for ncRNAs at the synapse. BC RNAs are sncRNAs that localize to both axons and dendrites, where they serve as local translational repressors by antagonizing translation initiation factors associated with specific mRNAs (Tiedge et al., 1991; Tiedge et al., 1993; Iacoangeli and Tiedge, 2013). Future studies should seek to identify the full complement of synaptic ncRNAs.

In addition to synaptically localized ncRNAs, synaptic function may be influenced by ncRNAs delivered from exosomes. Exosomes are 30–100 nm microvesicles that bud off from the plasma membrane of various cell types and travel through the extracellular space, where they merge with recipient cells to deliver cargoes that include various types of RNAs (Tetta et al., 2013). Exosomes from neurons are just beginning to be characterized. Rat cortical neuron cultures secrete exosomes in response to neuronal activity (Faure et al., 2006). Studies in fruit flies have shown that both presynaptic and postsynaptic neurons can interact through exosomal release and subsequent endocytosis (Koles et al., 2012; Korkut et al., 2009; Korkut et al., 2013). Furthermore, a recent study has identified various RNA species in neuronal exosomes (Morel et al., 2013), indicating that neuronal exosomes, like exosomes released from other cell types, carry ncRNAs. Together, these data support the possibility that neurons exchange various ncRNAs as part of synaptic transmission and plasticity mechanisms. Determining the ncRNA content and function of neuronal exosomes is another important area for synaptic function research.

Finally, as discussed above, synaptic ncRNAs appear to vary with age. However, limited time points and ncRNAs have been studied, and no study has comprehensively investigated the dynamics of synaptic ncRNAs during aging. Because ncRNAs can have broad effects due to interactions with multiple genes, age-dependent changes in the synaptic ncRNA milieu could have far-reaching consequences for plasticity and memory mechanisms throughout the lifespan. Therefore, determining the synaptic complement of ncRNAs throughout aging will be an important avenue for future research on aging and memory.

4. Synaptic functions of ncRNAs

Because of the morphologic and functional complexity of neurons, they are highly compartmentalized. One of the ways in which this is achieved is by shuttling mRNA

transcripts to specific subcellular areas and translating them locally in those areas. ncRNAs are increasingly being shown to be involved in the spatial and temporal control of mRNA translation. Such control is essential in functionally partitioned neurons, which is possibly why ncRNAs are so highly expressed in the brain. The processes whereby mRNAs are shuttled to the synapse and their synaptic translation is controlled in an activity-dependent manner are critical for proper synaptic function. In this section, we present several examples of the involvement of ncRNAs in synapse-specific translation, plasticity, and memory.

4.1. Synaptic functions of miRNAs

A growing list of miRNAs has been implicated in synaptic function and memory. For example, in cultured neurons, *miR-138* affects the morphology of dendritic spines, small protrusions that are the postsynaptic sites for the majority of excitatory synapses. Dendritic spines are also the sites for the postsynaptic events of synaptic plasticity. *miR-138* is concentrated in synapses, where it regulates dendritic spine size through its translational target acyl protein thioesterase 1 (Apt1) (Siegel et al., 2009). In cultured neurons, a *miR-138/Apt1* duplex associates with the Mov10 component of the RISC, and Mov10 is degraded on dendritic activation, thereby allowing for the temporally precise expression of Apt1 in response to neuronal stimulation (Banerjee et al., 2009). Unfortunately, because cultured neurons are not an ideal model for the aging adult brain, the extent to which *miR-138* controls spine morphology in adult or aged animals remains unclear. Furthermore, dendritic spine size and density may be dynamic in immature neurons, but are more stable in adults and probably have less impact on plasticity mechanisms (Grutzendler et al., 2002; Lang et al., 2004). Therefore, the control of Apt1 by *miR-138* may be relevant for structural synaptic changes in early developmental stages. It remains to be determined whether this pathway is active in more intact preparations and how it is affected by age.

Several groups have provided in vivo evidence in mouse models that *miR-132* is involved in synaptic plasticity, learning, and memory. *miR-132* expression is upregulated in the hippocampus and cortex in response to novel task learning (Hansen et al., 2013a; Mellios et al., 2011) and synaptic plasticity induction (Nudelman et al., 2010; Wibrand et al., 2010). *miR-132* is upregulated through BDNF-dependent and cAMP response element binding protein (CREB)-dependent mechanisms (Impey et al., 2004; Magill et al., 2010; Vo et al., 2005). Viral overexpression of *miR-132* in the perirhinal cortex modulates synaptic plasticity and memory (Scott et al., 2012). In the visual cortex, in vivo blockade of *miR-132* using a miRNA sponge prevents ocular dominance plasticity (Mellios et al., 2011). Importantly, transgenic animals that overexpress *miR-132* at low levels in the hippocampus have enhanced memory capabilities, whereas more robust overexpression of *miR-132* is detrimental to memory (Hansen et al., 2013a). This illustrates that neurons can be highly sensitive to miRNA concentration, with excessive increases in miRNA levels leading to neuronal malfunction, perhaps by inhibiting various transcripts in an “off-target” manner. This may be a consequence of both miRNA overexpression approaches and disease states in which miRNAs are upregulated.

One of the targets of *miR-132* is *MeCP2*, the gene implicated in the pathogenesis of Rett syndrome, a neurologic disorder characterized by cognitive deficiencies (Klein et al., 2007).

miR-132 also regulates p250GAP (Impey et al., 2004; Vo et al., 2005; Wayman et al., 2008), an NMDA receptor (NMDAR)-associated GTPase-activating protein that is involved in maintaining the morphology of dendritic spines (Nakazawa et al., 2008). Many in vivo studies have shown that the loss or gain of *miR-132* is associated with changes in spine morphology, and *miR-132* control of p250GAP is likely responsible for these changes. However, while structural changes have been associated with *miR-132*-related learning and disease, there is not yet evidence that these changes are causally linked. For instance, in Rett syndrome, changes in dendritic spine morphology are highly variable and poorly correlate with behavioral outcomes (Chapleau et al., 2012; Wang et al., 2013). Future studies that extend beyond the structural effects of *miR-132*s and seek to causally link *miR-132* to synaptic plasticity mechanisms will be of interest. Interestingly, a microarray study that compared miRNA levels throughout the lifespan showed that *miR-132* increases with age in the mouse cortex (Eda et al., 2011). Because *miR-132* at very high levels is detrimental to hippocampus-dependent memory, it will be interesting to determine whether the age-dependent increase in *miR-132* contributes to agedependent memory decline.

The expression of three miRNAs, *miR-182*, *miR128b*, and *miR-134*, is regulated in response to the acquisition of fear memory (Griggs et al., 2013; Lin et al., 2011). *miR-134* is a brain-specific miRNA that works upstream of BDNF and CREB to control synaptic plasticity, learning, and memory (Figure 1). *miR-134* expression is controlled in the brain by the deacetylase SIRT1. Brain-specific loss of SIRT1 results in increased expression of *miR-134*, and deficits in learning and synaptic plasticity through the suppression of CREB and BDNF (Gao et al., 2010). SIRT1 is a member of the phylogenetically conserved sirtuin family of proteins, which has long been known to play a role in the longevity of organisms ranging from yeast to humans (Cohen et al., 2004; Howitz et al., 2003; Kaeberlein et al., 1999). Furthermore, either SIRT1 activation or *miR-134* silencing has a neuroprotective effect in a number of age-dependent neurodegenerative processes (Araki et al., 2004; Jimenez-Mateos et al., 2012; Kim et al., 2007). Therefore, the control of *miR-134* via SIRT1 throughout the lifespan is a potentially important topic for research on the aging brain.

A recent study by our group has shown that *mir-185* and *miR-25* act in an age-dependent manner to control synaptic plasticity. The 22q11 deletion syndrome (22q11DS) is the most common microdeletion syndrome in humans and is characterized by cognitive deficits, among other symptoms (Earls and Zakharenko, 2013). One of the genes expressed in the deletion region is the miRNA biosynthesis gene *DGCR8*. In mouse models of 22q11DS, the haploinsufficiency of *Dgcr8* disrupts the expression of multiple miRNAs. Interestingly, the loss of at least two of these miRNAs, *miR-185* and *miR-25*, results in age-dependent effects on synaptic plasticity in the hippocampus. Although these miRNAs are depleted throughout the lifespan in 22q11DS mouse models, they only begin to have effects on LTP in mature (4-month-old) brains. This age-dependent regulation of LTP is accomplished by translational targeting of the sarco/endoplasmic reticulum Ca^{2+} ATPase type 2 (Serca2), a key regulator of synaptic Ca^{2+} and synaptic plasticity (Earls et al., 2012; Earls et al., 2010). It is nevertheless unclear why these miRNAs influence Serca2 translation only at later ages. This finding illustrates the potential for miRNAs to differentially influence physiologic processes throughout the lifespan.

The control of other synaptic plasticity factors by miRNAs is an interesting area for future research on the aging brain. For instance, hippocampal LTP consists of NMDAR-dependent and NMDAR-independent components (Blundon and Zakharenko, 2008). As the brain ages, NMDAR-dependent processes decline, but NMDAR-independent plasticity mechanisms compensate in high-performing brains (Boric et al., 2008). Therefore, miRNAs that control NMDAR-dependent and NMDAR-independent forms of plasticity may be key regulators of age-dependent memory decline. For example, *miR-125b* targets the NR2A subunit of NMDAR (Edbauer et al., 2010). Furthermore, NMDAR subunits in the brain show an age-dependent decline (Magnusson et al., 2002). Whether *miR-125b* or other miRNAs are responsible for age-dependent NMDAR changes is an interesting topic for future research.

Several important pieces of data are missing in our understanding of how miRNAs contribute to age-dependent memory decline. First, we must determine how synaptic miRNAs change throughout aging. miRNA-dependent processes studied in cultured cells and younger synapses may not accurately reflect the pathways that are important later in life and in the intact brain. Second, we need to determine the roles of synaptically localized miRNAs in synaptic plasticity or memory by functional and behavioral approaches. These studies will provide insights into how synaptically located miRNAs contribute to plasticity and memory processes throughout aging.

4.2. lncRNAs in synaptic plasticity and memory

Currently, little is known about the roles of lncRNAs in the CNS. These transcripts were long ignored as spurious transcriptional products or sequencing artifacts. However, it is now widely believed that lncRNAs represent a new class of RNAs with potential biologic functions. lncRNAs are arbitrarily defined as endogenous RNAs longer than 200 nt and without an appreciable (more than 300 nt), predicted open reading frame (ORF) (Ng et al., 2013). However, defining this class of ncRNAs based merely on the absence of a substantial ORF is problematic. For example, the *Tal* gene in *Drosophila* was initially annotated as a 1.5 kb non-coding transcript, but later was shown to encode multiple short peptides important for various developmental processes (Galindo et al., 2007). Furthermore, some RNAs are bi-functional, operating within the cell as a non-transcribed RNA, but also encoding a protein. For example, the steroid receptor RNA activator (SRA) encodes an RNA that regulates nuclear hormone receptor signaling (Redfern et al., 2013) but also encodes the functional protein SRAP (Chooniedass-Kothari et al., 2004). lncRNAs can contain structural components of messenger RNAs (e.g., 5' methyl caps or 3' polyA signals) and may undergo splicing events (Carninci et al., 2005; Kapranov et al., 2007; Qureshi et al., 2010). Those lncRNAs with known functions have been shown to regulate protein expression by several mechanisms, including transcriptional inactivation of chromatin by histone methylation (Silva et al., 2003), transcriptional interference resulting from converging transcription complexes (Osato et al., 2007), modification of alternative splicing (Barry et al., 2013), depletion of mRNA through decay mechanisms (Gong and Maquat, 2011), depletion of endogenous miRNAs (Cesana et al., 2011), and stabilization of mRNAs (Faghihi et al., 2008).

Several years ago, Mercer et al. mined in situ hybridization data from the Allen Brain Atlas to analyze the expression of lncRNAs within the brain (Mercer et al., 2008). The authors showed that most of the lncRNAs examined (849 of 1328) exhibited specific brain region, neuronal cell type, and/or subcellular expression patterns. For example, AK089514, BC051426, and AK037594 are expressed in the hippocampus in anatomically distinct subregions. AK089514 is expressed in the dentate gyrus, while BC051426 is specific to the CA1-CA3 regions, and AK037594 is detected in the dentate gyrus and CA1 regions. Region-specific expression of other lncRNAs was also observed within particular layers of the cortex and cerebellum. Furthermore, the large size of cerebellar Purkinje neurons enabled the authors to localize lncRNAs to specific subcellular compartments such as the nucleus, soma, and proximal dendrites. This region and subregion specificity, along with subcellular compartmentalization, suggests that lncRNA expression is regulated, and thus is not the result of spurious transcription.

The ability of ncRNAs to directly regulate human brain development and function is supported by the identification and characterization of the lncRNA *UBE3A-ATS* in Angelman syndrome (AS). AS is a classical imprinting disease characterized by intellectual disability, speech impairment, and severe developmental delays (Mabb et al., 2011). AS is caused by the deletion of or an inactivating mutation in the maternal E3 ubiquitin ligase (*UBE3A*) gene. In addition, the paternal allele of *UBE3A* undergoes silencing due to expression of the lncRNA *UBE3A-ATS*. In the AS mutant mouse model, there is a deficit in hippocampal LTP (Jiang et al., 1998). Similar plasticity deficits are likely responsible for the learning disabilities observed in AS patients. This important role for *UBE3A-ATS* in neuronal function highlights the possible role of lncRNAs in memory formation and necessitates further investigation on the role of other lncRNAs in these processes.

In addition to being regulated by miRNAs, BDNF is also regulated by at least one lncRNA. *BDNF-AS* is a lncRNA that is oriented on the opposite DNA strand from *BDNF* (Pruunsild et al., 2007) and inhibits transcription of *BDNF* most likely by regulating the histone methyltransferase PRC2 (Modarresi et al., 2012).

The lncRNA *Gomafu* is widely expressed throughout the brain (Mercer et al., 2008) and has been recently shown to regulate alternative splicing of the schizophrenia-associated genes *DISC1* and *ERBB4* (Barry et al., 2013). Because the loss of *ErbB4* in mice enhances LTP in the hippocampus, it will be important to determine whether *Gomafu*-regulated splice isoforms affect normal synaptic plasticity (Pitcher et al., 2008; Shamir et al., 2012).

The region specificity and subcellular localization of lncRNA expression suggest that lncRNAs play pivotal biologic functions within the brain. Electrophysiologic and behavioral assays in knockout mice deficient in specific lncRNAs are required to elucidate the functions of lncRNAs in memory formation and maintenance. Moreover, comparative global analyses of RNAs, histone modifications, and DNA methylations may be necessary to identify the genetic targets of some lncRNAs. The findings that some lncRNAs are enriched in specific subcellular regions, such as dendrites or axons, provoke many interesting questions. What factor(s) are required for trafficking lncRNAs to the site where they function? What mechanisms govern their expression? Does the expression, localization,

or function of lncRNAs depend on neuronal activity? Does the expression of lncRNAs change during aging? The answers to these questions will shed light on the role of these biologically significant RNAs.

4.3. piRNAs in the CNS

piRNAs were originally described in germ cells, where they play a crucial role in the development and maintenance of germline cells in flies, worms, and mammals (Girard et al., 2006; Peng and Lin, 2013). While mature piRNAs and mature miRNAs are similar in size, they differ in nearly every other aspect. For example, piRNAs, are far more abundant than miRNAs, with ~2000 mature miRNAs having been discovered compared to over 60,000 mature piRNAs identified in the mouse (Beyret et al., 2012). piRNAs are also distinguished by their interactome, requiring RNA-binding proteins of the Piwi subfamily (Meister, 2013), whereas miRNAs associate with the closely related Argonaute subfamily. Also, whereas miRNA transcripts are hairpins recognized and cleaved by double-stranded RNA-binding proteins, piRNAs are transcribed as long (up to 120 kb) linear precursors, and are cleaved into 24–34 nt mature forms by a yet-unidentified mechanism (Saxe and Lin, 2011). Finally, unlike miRNAs, which bind to mRNAs to suppress translation, piRNAs regulate gene expression by at least three distinct mechanisms: epigenetic silencing of gene transcription by histone modifications and DNA methylation, and regulation of protein translation (Juliano et al., 2011; Peng and Lin, 2013).

Are piRNAs expressed in the nervous system? If so, do they have a biological function? Recent work from several labs has shown piRNA expression in the CNS of snails, flies, and mammals (Lee et al., 2011; Rajasethupathy et al., 2012; Yan et al., 2011). Sequencing of a library generated from neurons from the snail *Aplysia californica* identified 372 distinct piRNA clusters (Rajasethupathy et al., 2012). The study further showed that several piRNAs increased their expression in response to the neurotransmitter serotonin, and that some piRNAs enhance the serotonin-dependent facilitation of synaptic transmission. Furthermore, piRNAs caused the downregulation of Creb2, a transcriptional repressor of memory by methylation of its promoter (Rajasethupathy et al., 2012).

In another report, deep sequencing of small RNA libraries from the mouse hippocampus identified hundreds of piRNAs (Lee et al., 2011). In addition, the subcellular localization of four piRNAs in cultured hippocampal neurons was examined by in situ hybridization, with three piRNAs localized to the cell body and one piRNA localized to the soma and neurites. Furthermore, Miwi (a Piwi-family member) and mmu_piR_000935 (also known as DQ541777) localized to discrete puncta along the neurite. Although the authors showed that mmu_piR_000935 is required in vitro to regulate dendritic spine size (8% reduction when the piRNA is depleted), these results need to be validated in vivo. In vitro studies using antisense antagonomers in cultured neurons may help elucidate the role of specific piRNAs. However, further development of such tools is necessary to validate their function in synaptic transmission and memory in vivo.

4.4. circRNAs in the synapse

Several independent laboratories have recently reported a new class of ncRNAs called circRNAs. Although their function was unknown, evidence for the presence of circRNAs surfaced more than 20 years ago with the identification of the testis-localized circRNA *Sry* (Capel et al., 1993). More recently, circRNAs have gained wider appeal as their functional relevance has become apparent. The details of circRNA biogenesis are currently unknown. It is proposed that circRNAs are a product of alternative splicing events, as the 5' end of a preceding exon (splice acceptor) is joined with the 3' end of a downstream exon (splice donor) by a yet-unidentified mechanism (Howitz et al., 2003).

Although few circRNAs have been studied in detail, some circRNAs have been shown to act as endogenous miRNA sponges, binding and sequestering miRNAs. For example, the circRNA *ciRS-7* contains more than 60 binding sites for the miRNA *miR-7*. *miR-7* and *ciRS-7* are widely expressed in the embryonic and adult brain in an overlapping pattern, which would be expected if *ciRS-7* regulates *miR-7* (Hansen et al., 2013b). Each brain region expresses different levels of *ciRS-7* (Hansen et al., 2013b; Memczak et al., 2013) suggesting region-specialized regulation of *miR-7* by *ciRS-7*. This circRNA–miRNA interaction is thought to be required for normal brain morphogenesis (Memczak et al., 2013).

As the circRNA field progresses and with the development of more sophisticated bioinformatic tools to identify circRNAs from RNA-sequencing experiments, the extent to which circRNAs regulate known plasticity-related miRNAs can be experimentally assessed. Furthermore, the potential modulation of miRNA function by circRNAs is an additional layer of regulation of protein expression. This complicates the study of miRNAs in aging by simply measuring miRNA expression levels. A given miRNA may be expressed at constant levels, but its availability to interact with targets may depend on its sequestration by circRNAs. Since there is evidence of developmental regulation of circRNAs in worms (Memczak et al., 2013), it is necessary to study the expression of circRNAs in both young and older mammalian brains, as circRNAs may be novel players in age-associated memory decline.

5. Conclusions and future directions

Until recently, nearly all efforts in molecular neuroscience were focused on protein-coding genes. It is now appreciated that protein-coding RNAs constitute only a fraction of the transcriptome, and ncRNAs are emerging as important, unexplored regulators of various cellular processes. ncRNAs can affect histone methylation, transcription, transcript splicing, translation, mRNA stability, and the activity of other ncRNAs (e.g., circRNA/miRNA interaction) (Figure 2). Therefore, a multilayered network of ncRNAs can control a given protein-coding gene at multiple points. Although current research on ncRNA function has mainly focused on ncRNA roles as regulators of coding genes, future studies may reveal other diverse roles of ncRNAs in the cell.

Thus far, little is known about the roles that ncRNAs play in age-dependent memory decline. There are several possible scenarios for age-dependent control of memory by

ncRNAs. piRNAs may be involved in age-dependent histone control of complex networks of memory-related genes. Antisense lncRNAs might target transcription of specific genes important for memory in an age-dependent fashion. miRNAs also have the potential to affect complex networks of genes, providing translational control and affecting mRNA stability. Because many miRNAs show developmental patterns of expression, it is not unreasonable to imagine scenarios where age-dependent changes occur in the miRNA constitution of cells, which in turn affect the protein environment. The targeting of Serca2 protein levels represents one of the first known examples of age-dependent regulation of a target by miRNAs (Earls et al., 2012). The control of miRNAs by aging genes such as *SIRT1* and interactions of miRNAs with the age-dependent NMDAR pathway suggest that many more miRNA pathways may be critical for the aging brain. In addition, the complement of miRNAs may differ not only with age but also in individual cells and subcellular regions, adding further variables for future research.

These and other scenarios for ncRNAs in aging represent future areas of research on the molecular neuroscience of aging. To begin to understand the role of ncRNAs in aging and memory, we must first obtain several important pieces of information. First, we need a comprehensive profile of ncRNAs that are expressed in the brain, their cell-type specificities, and subcellular localizations. Next, we need to determine how ncRNA expression and localization change with age. Because ncRNA activity can be modulated through binding to other factors, such as the modulation of miRNAs by circRNAs, we must ask how different classes of ncRNAs are differentially modulated with age. Assays with a functional readout will therefore be essential additions to expression studies. We also need to determine how ncRNA function is affected by nucleotide editing events, which can be age-dependent (Ekdahl et al., 2012; Gu et al., 2012; Peng et al., 2012). Answering these and other relevant questions will begin to provide a framework for a holistic understanding of memory and its age-dependent decline.

Acknowledgments

This work was supported in part by the National Institute of Mental Health of the National Institutes of Health (award numbers R01 MH095810, R01 MH097742 to S.S.Z. and K99 MH101240 to L.R.E.) and by the American Lebanese Syrian Associated Charities. We thank Dr. Jamy Peng for constructive comments, Dr. Vani Shanker for editing the manuscript, and Roderick Thompson for assistance with figures. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health or other granting agencies.

References

- Araki T, Sasaki Y, Milbrandt J. Increased nuclear NAD biosynthesis and SIRT1 activation prevent axonal degeneration. *Science*. 2004; 305:1010–1013. [PubMed: 15310905]
- Banerjee S, Neveu P, Kosik KS. A coordinated local translational control point at the synapse involving relief from silencing and MOV10 degradation. *Neuron*. 2009; 64:871–884. [PubMed: 20064393]
- Barry G, Briggs JA, Vanichkina DP, Poth EM, Beveridge NJ, Ratnu VS, Nayler SP, Nones K, Hu J, Bredy TW, Nakagawa S, Rigo F, Taft RJ, Cairns MJ, Blackshaw S, Wolvetang EJ, Mattick JS. The long non-coding RNA Gomaflu is acutely regulated in response to neuronal activation and involved in schizophrenia-associated alternative splicing. *Mol. Psychiatry*. 2013 Epub ahead of print 04/30/2013.

- Beyret E, Liu N, Lin H. piRNA biogenesis during adult spermatogenesis in mice is independent of the ping-pong mechanism. *Cell Res.* 2012; 22:1429–1439. [PubMed: 22907665]
- Blundon JA, Zakharenko SS. Dissecting the components of long-term potentiation. *Neuroscientist.* 2008; 14:598–608. [PubMed: 18940785]
- Boric K, Munoz P, Gallagher M, Kirkwood A. Potential adaptive function for altered long-term potentiation mechanisms in aging hippocampus. *J.Neurosci.* 2008; 28:8034–8039. [PubMed: 18685028]
- Cabili MN, Trapnell C, Goff L, Koziol M, Tazon-Vega B, Regev A, Rinn JL. Integrative annotation of human large intergenic noncoding RNAs reveals global properties and specific subclasses. *Genes Dev.* 2011; 25:1915–1927. [PubMed: 21890647]
- Cao X, Yeo G, Muotri AR, Kuwabara T, Gage FH. Noncoding RNAs in the mammalian central nervous system. *Annu.Rev.Neurosci.* 2006; 29:77–103. [PubMed: 16776580]
- Capel B, Swain A, Nicolis S, Hacker A, Walter M, Koopman P, Goodfellow P, Lovell-Badge R. Circular transcripts of the testis-determining gene Sry in adult mouse testis. *Cell.* 1993; 73:1019–1030. [PubMed: 7684656]
- Carninci P, Kasukawa T, Katayama S, Gough J, Frith MC, Maeda N, Oyama R, Ravasi T, Lenhard B, Wells C, Kodzius R, Shimokawa K, Bajic VB, Brenner SE, Batalov S, Forrest AR, Zavolan M, Davis MJ, Wilming LG, Aidinis V, Allen JE, Ambesi-Impimbato A, Apweiler R, Aturaliya RN, Bailey TL, Bansal M, Baxter L, Beisel KW, Bersano T, Bono H, Chalk AM, Chiu KP, Choudhary V, Christoffels A, Clutterbuck DR, Crowe ML, Dalla E, Dalrymple BP, de BB, Della GG, di BD, Down T, Engstrom P, Fagiolini M, Faulkner G, Fletcher CF, Fukushima T, Furuno M, Futaki S, Gariboldi M, Georgii-Hemming P, Gingeras TR, Gojobori T, Green RE, Gustincich S, Harbers M, Hayashi Y, Hensch TK, Hirokawa N, Hill D, Huminecki L, Iacono M, Ikeo K, Iwama A, Ishikawa T, Jakt M, Kanapin A, Katoh M, Kawasaki Y, Kelso J, Kitamura H, Kitano H, Kollias G, Krishnan SP, Kruger A, Kummerfeld SK, Kurochkin IV, Lareau LF, Lazarevic D, Lipovich L, Liu J, Liuni S, McWilliam S, Madan BM, Madera M, Marchionni L, Matsuda H, Matsuzawa S, Miki H, Mignone F, Miyake S, Morris K, Mottagui-Tabar S, Mulder N, Nakano N, Nakauchi H, Ng P, Nilsson R, Nishiguchi S, Nishikawa S, Nori F, Ohara O, Okazaki Y, Orlando V, Pang KC, Pavan WJ, Pavesi G, Pesole G, Petrovsky N, Piazza S, Reed J, Reid JF, Ring BZ, Ringwald M, Rost B, Ruan Y, Salzberg SL, Sandelin A, Schneider C, Schonbach C, Sekiguchi K, Semple CA, Seno S, Sessa L, Sheng Y, Shibata Y, Shimada H, Shimada K, Silva D, Sinclair B, Sperling S, Stupka E, Sugiura K, Sultana R, Takenaka Y, Taki K, Tammoja K, Tan SL, Tang S, Taylor MS, Tegner J, Teichmann SA, Ueda HR, van NE, Verardo R, Wei CL, Yagi K, Yamanishi H, Zabarovsky E, Zhu S, Zimmer A, Hide W, Bult C, Grimmond SM, Teasdale RD, Liu ET, Brusic V, Quackenbush J, Wahlestedt C, Mattick JS, Hume DA, Kai C, Sasaki D, Tomaru Y, Fukuda S, Kanamori-Katayama M, Suzuki M, Aoki J, Arakawa T, Iida J, Imamura K, Itoh M, Kato T, Kawaji H, Kawagashira N, Kawashima T, Kojima M, Kondo S, Konno H, Nakano K, Ninomiya N, Nishio T, Okada M, Plessy C, Shibata K, Shiraki T, Suzuki S, Tagami M, Waki K, Watahiki A, Okamura-Oho Y, Suzuki H, Kawai J, Hayashizaki Y. The transcriptional landscape of the mammalian genome. *Science.* 2005; 309:1559–1563. [PubMed: 16141072]
- Cesana M, Cacchiarelli D, Legnini I, Santini T, Sthandier O, Chinappi M, Tramontano A, Bozzoni I. A long noncoding RNA controls muscle differentiation by functioning as a competing endogenous RNA. *Cell.* 2011; 147:358–369. [PubMed: 22000014]
- Chapleau CA, Boggio EM, Calfa G, Percy AK, Giustetto M, Pozzo-Miller L. Hippocampal CA1 pyramidal neurons of Mecp2 mutant mice show a dendritic spine phenotype only in the presymptomatic stage. *Neural Plast.* 2012; 2012:976164. [PubMed: 22919518]
- Chooniedass-Kothari S, Emberley E, Hamedani MK, Troup S, Wang X, Czosnek A, Hube F, Mutawe M, Watson PH, Leygue E. The steroid receptor RNA activator is the first functional RNA encoding a protein. *FEBS Lett.* 2004; 566:43–47. [PubMed: 15147866]
- Cohen HY, Miller C, Bitterman KJ, Wall NR, Hekking B, Kessler B, Howitz KT, Gorospe M, de CR, Sinclair DA. Calorie restriction promotes mammalian cell survival by inducing the SIRT1 deacetylase. *Science.* 2004; 305:390–392. [PubMed: 15205477]
- Czech B, Hannon GJ. Small RNA sorting: matchmaking for Argonautes. *Nat.Rev.Genet.* 2011; 12:19–31. [PubMed: 21116305]

- Day JJ, Sweatt JD. Epigenetic mechanisms in cognition. *Neuron*. 2011; 70:813–829. [PubMed: 21658577]
- Derrien T, Johnson R, Bussotti G, Tanzer A, Djebali S, Tilgner H, Guernec G, Martin D, Merkel A, Knowles DG, Lagarde J, Veeravalli L, Ruan X, Ruan Y, Lassmann T, Carninci P, Brown JB, Lipovich L, Gonzalez JM, Thomas M, Davis CA, Shiekhata R, Gingeras TR, Hubbard TJ, Notredame C, Harrow J, Guigo R. The GENCODE v7 catalog of human long noncoding RNAs: analysis of their gene structure, evolution, and expression. *Genome Res*. 2012; 22:1775–1789. [PubMed: 22955988]
- Earls LR, Bayazitov IT, Fricke RG, Berry RB, Illingworth E, Mittleman G, Zakharenko SS. Dysregulation of presynaptic calcium and synaptic plasticity in a mouse model of 22q11 deletion syndrome. *J Neurosci*. 2010; 30:15843–15855. [PubMed: 21106823]
- Earls LR, Fricke RG, Yu J, Berry RB, Baldwin LT, Zakharenko SS. Age-dependent microRNA control of synaptic plasticity in 22q11 deletion syndrome and schizophrenia. *J Neurosci*. 2012; 32:14132–14144. [PubMed: 23055483]
- Earls LR, Zakharenko SS. A Synaptic Function Approach to Investigating Complex Psychiatric Diseases. *Neuroscientist*. 2013 Epub ahead of print 11/14/2013.
- Eda A, Takahashi M, Fukushima T, Hohjoh H. Alteration of microRNA expression in the process of mouse brain growth. *Gene*. 2011; 485:46–52. [PubMed: 21718763]
- Edbauer D, Neilson JR, Foster KA, Wang CF, Seeburg DP, Batterton MN, Tada T, Dolan BM, Sharp PA, Sheng M. Regulation of synaptic structure and function by FMRP-associated microRNAs miR-125b and miR-132. *Neuron*. 2010; 65:373–384. [PubMed: 20159450]
- Ekdahl Y, Farahani HS, Behm M, Lagergren J, Ohman M. A-to-I editing of microRNAs in the mammalian brain increases during development. *Genome Res*. 2012; 22:1477–1487. [PubMed: 22645261]
- Errico F, Nistico R, Napolitano F, Oliva AB, Romano R, Barbieri F, Florio T, Russo C, Mercuri NB, Usiello A. Persistent increase of D-aspartate in D-aspartate oxidase mutant mice induces a precocious hippocampal age-dependent synaptic plasticity and spatial memory decay. *Neurobiol Aging*. 2011; 32:2061–2074. [PubMed: 20031274]
- Errico F, Nistico R, Palma G, Federici M, Affuso A, Brilli E, Topo E, Centonze D, Bernardi G, Bozzi Y, D'Aniello A, Di LR, Mercuri NB, Usiello A. Increased levels of d-aspartate in the hippocampus enhance LTP but do not facilitate cognitive flexibility. *Mol Cell Neurosci*. 2008; 37:236–246. [PubMed: 17981050]
- Faghihi MA, Modarresi F, Khalil AM, Wood DE, Sahagan BG, Morgan TE, Finch CE, St LG III, Kenny PJ, Wahlestedt C. Expression of a noncoding RNA is elevated in Alzheimer's disease and drives rapid feed-forward regulation of beta-secretase. *Nat Med*. 2008; 14:723–730. [PubMed: 18587408]
- Faghihi MA, Wahlestedt C. Regulatory roles of natural antisense transcripts. *Nat Rev Mol Cell Biol*. 2009; 10:637–643. [PubMed: 19638999]
- Faure J, Lachenal G, Court M, Hirrlinger J, Chatellard-Causse C, Blot B, Grange J, Schoehn G, Goldberg Y, Boyer V, Kirchhoff F, Raposo G, Garin J, Sadoul R. Exosomes are released by cultured cortical neurones. *Mol Cell Neurosci*. 2006; 31:642–648. [PubMed: 16446100]
- Figurov A, Pozzo-Miller LD, Olafsson P, Wang T, Lu B. Regulation of synaptic responses to high-frequency stimulation and LTP by neurotrophins in the hippocampus. *Nature*. 1996; 381:706–709. [PubMed: 8649517]
- Furuno M, Pang KC, Ninomiya N, Fukuda S, Frith MC, Bult C, Kai C, Kawai J, Carninci P, Hayashizaki Y, Mattick JS, Suzuki H. Clusters of internally primed transcripts reveal novel long noncoding RNAs. *PLoS Genet*. 2006; 2:e37. [PubMed: 16683026]
- Galindo MI, Pueyo JI, Fouix S, Bishop SA, Couso JP. Peptides encoded by short ORFs control development and define a new eukaryotic gene family. *PLoS Biol*. 2007; 5:e106. [PubMed: 17439302]
- Gao J, Wang WY, Mao YW, Graff J, Guan JS, Pan L, Mak G, Kim D, Su SC, Tsai LH. A novel pathway regulates memory and plasticity via SIRT1 and miR-134. *Nature*. 2010; 466:1105–1109. [PubMed: 20622856]

- Girard A, Sachidanandam R, Hannon GJ, Carmell MA. A germline-specific class of small RNAs binds mammalian Piwi proteins. *Nature*. 2006; 442:199–202. [PubMed: 16751776]
- Gong C, Maquat LE. IncRNAs transactivate STAU1-mediated mRNA decay by duplexing with 3' UTRs via Alu elements. *Nature*. 2011; 470:284–288. [PubMed: 21307942]
- Griggs EM, Young EJ, Rumbaugh G, Miller CA. MicroRNA-182 regulates amygdala-dependent memory formation. *J.Neurosci*. 2013; 33:1734–1740. [PubMed: 23345246]
- Grutzendler J, Kasthuri N, Gan WB. Long-term dendritic spine stability in the adult cortex. *Nature*. 2002; 420:812–816. [PubMed: 12490949]
- Gu T, Buaas FW, Simons AK, Ackert-Bicknell CL, Braun RE, Hibbs MA. Canonical A-to-I and C-to-U RNA editing is enriched at 3'UTRs and microRNA target sites in multiple mouse tissues. *PLoS.One*. 2012; 7:e33720. [PubMed: 22448268]
- Hansen KF, Karelina K, Sakamoto K, Wayman GA, Impey S, Obrietan K. miRNA-132: a dynamic regulator of cognitive capacity. *Brain Struct.Funct*. 2013a; 218:817–831. [PubMed: 22706759]
- Hansen TB, Jensen TI, Clausen BH, Bramsen JB, Finsen B, Damgaard CK, Kjems J. Natural RNA circles function as efficient microRNA sponges. *Nature*. 2013b; 495:384–388. [PubMed: 23446346]
- Hentze MW, Preiss T. Circular RNAs: splicing's enigma variations. *EMBO J*. 2013; 32:923–925. [PubMed: 23463100]
- Howitz KT, Bitterman KJ, Cohen HY, Lamming DW, Lavu S, Wood JG, Zipkin RE, Chung P, Kisielewski A, Zhang LL, Scherer B, Sinclair DA. Small molecule activators of sirtuins extend *Saccharomyces cerevisiae* lifespan. *Nature*. 2003; 425:191–196. [PubMed: 12939617]
- Iacoangeli A, Tiedge H. Translational control at the synapse: role of RNA regulators. *Trends Biochem.Sci*. 2013; 38:47–55. [PubMed: 23218750]
- Impey S, McCorkle SR, Cha-Molstad H, Dwyer JM, Yochum GS, Boss JM, McWeeney S, Dunn JJ, Mandel G, Goodman RH. Defining the CREB regulon: a genome-wide analysis of transcription factor regulatory regions. *Cell*. 2004; 119:1041–1054. [PubMed: 15620361]
- Jeon Y, Lee JT. YY1 tethers Xist RNA to the inactive X nucleation center. *Cell*. 2011; 146:119–133. [PubMed: 21729784]
- Ji Y, Lu Y, Yang F, Shen W, Tang TT, Feng L, Duan S, Lu B. Acute and gradual increases in BDNF concentration elicit distinct signaling and functions in neurons. *Nat.Neurosci*. 2010; 13:302–309. [PubMed: 20173744]
- Jiang YH, Armstrong D, Albrecht U, Atkins CM, Noebels JL, Eichele G, Sweatt JD, Beaudet AL. Mutation of the Angelman ubiquitin ligase in mice causes increased cytoplasmic p53 and deficits of contextual learning and long-term potentiation. *Neuron*. 1998; 21:799–811. [PubMed: 9808466]
- Jimenez-Mateos EM, Engel T, Merino-Serrais P, McKiernan RC, Tanaka K, Mouri G, Sano T, O'Tuathaigh C, Waddington JL, Prenter S, Delanty N, Farrell MA, O'Brien DF, Conroy RM, Stallings RL, DeFelipe J, Henshall DC. Silencing microRNA-134 produces neuroprotective and prolonged seizure-suppressive effects. *Nat.Med*. 2012; 18:1087–1094. [PubMed: 22683779]
- Juliano C, Wang J, Lin H. Uniting germline and stem cells: the function of Piwi proteins and the piRNA pathway in diverse organisms. *Annu.Rev.Genet*. 2011; 45:447–469. [PubMed: 21942366]
- Kaerberlein M, McVey M, Guarente L. The SIR2/3/4 complex and SIR2 alone promote longevity in *Saccharomyces cerevisiae* by two different mechanisms. *Genes Dev*. 1999; 13:2570–2580. [PubMed: 10521401]
- Kapranov P, Cheng J, Dike S, Nix DA, Duttagupta R, Willingham AT, Stadler PF, Hertel J, Hackermuller J, Hofacker IL, Bell I, Cheung E, Drenkow J, Dumais E, Patel S, Helt G, Ganesh M, Ghosh S, Piccolboni A, Sementchenko V, Tammana H, Gingeras TR. RNA maps reveal new RNA classes and a possible function for pervasive transcription. *Science*. 2007; 316:1484–1488. [PubMed: 17510325]
- Kim D, Nguyen MD, Dobbin MM, Fischer A, Sananbenesi F, Rodgers JT, Delalle I, Baur JA, Sui G, Armour SM, Puigserver P, Sinclair DA, Tsai LH. SIRT1 deacetylase protects against neurodegeneration in models for Alzheimer's disease and amyotrophic lateral sclerosis. *EMBO J*. 2007; 26:3169–3179. [PubMed: 17581637]

- Klein ME, Lioy DT, Ma L, Impey S, Mandel G, Goodman RH. Homeostatic regulation of MeCP2 expression by a CREB-induced microRNA. *Nat.Neurosci.* 2007; 10:1513–1514. [PubMed: 17994015]
- Koles K, Nunnari J, Korkut C, Barria R, Brewer C, Li Y, Leszyk J, Zhang B, Budnik V. Mechanism of evenness interrupted (Evi)-exosome release at synaptic boutons. *J.Biol.Chem.* 2012; 287:16820–16834. [PubMed: 22437826]
- Korkut C, Ataman B, Ramachandran P, Ashley J, Barria R, Gherbesi N, Budnik V. Trans-synaptic transmission of vesicular Wnt signals through Evi/Wntless. *Cell.* 2009; 139:393–404. [PubMed: 19837038]
- Korkut C, Li Y, Koles K, Brewer C, Ashley J, Yoshihara M, Budnik V. Regulation of postsynaptic retrograde signaling by presynaptic exosome release. *Neuron.* 2013; 77:1039–1046. [PubMed: 23522040]
- Korzus E, Rosenfeld MG, Mayford M. CBP histone acetyltransferase activity is a critical component of memory consolidation. *Neuron.* 2004; 42:961–972. [PubMed: 15207240]
- Kosik KS. The neuronal microRNA system. *Nat.Rev.Neurosci.* 2006; 7:911–920. [PubMed: 17115073]
- Krichevsky AM, King KS, Donahue CP, Khrapko K, Kosik KS. A microRNA array reveals extensive regulation of microRNAs during brain development. *RNA.* 2003; 9:1274–1281. [PubMed: 13130141]
- Lang C, Barco A, Zablow L, Kandel ER, Siegelbaum SA, Zakharenko SS. Transient expansion of synaptically connected dendritic spines upon induction of hippocampal long-term potentiation. *Proc.Natl.Acad.Sci.U.S.A.* 2004; 101:16665–16670. [PubMed: 15542587]
- Lee EJ, Banerjee S, Zhou H, Jammalamadaka A, Arcila M, Manjunath BS, Kosik KS. Identification of piRNAs in the central nervous system. *RNA.* 2011; 17:1090–1099. [PubMed: 21515829]
- Levenson JM, O'Riordan KJ, Brown KD, Trinh MA, Molfese DL, Sweatt JD. Regulation of histone acetylation during memory formation in the hippocampus. *J.Biol.Chem.* 2004; 279:40545–40559. [PubMed: 15273246]
- Lin Q, Wei W, Coelho CM, Li X, Baker-Andresen D, Dudley K, Ratnu VS, Boskovic Z, Kobor MS, Sun YE, Bredy TW. The brain-specific microRNA miR-128b regulates the formation of fear-extinction memory. *Nat.Neurosci.* 2011; 14:1115–1117. [PubMed: 21841775]
- Lugli G, Torvik VI, Larson J, Smalheiser NR. Expression of microRNAs and their precursors in synaptic fractions of adult mouse forebrain. *J.Neurochem.* 2008; 106:650–661. [PubMed: 18410515]
- Mabb AM, Judson MC, Zylka MJ, Philpot BD. Angelman syndrome: insights into genomic imprinting and neurodevelopmental phenotypes. *Trends Neurosci.* 2011; 34:293–303. [PubMed: 21592595]
- Magill ST, Cambronne XA, Luikart BW, Lioy DT, Leighton BH, Westbrook GL, Mandel G, Goodman RH. microRNA-132 regulates dendritic growth and arborization of newborn neurons in the adult hippocampus. *Proc.Natl.Acad.Sci.U.S.A.* 2010; 107:20382–20387. [PubMed: 21059906]
- Magnusson KR, Nelson SE, Young AB. Age-related changes in the protein expression of subunits of the NMDA receptor. *Brain Res.Mol.Brain Res.* 2002; 99:40–45. [PubMed: 11869807]
- Malone CD, Hannon GJ. Molecular evolution of piRNA and transposon control pathways in *Drosophila*. *Cold Spring Harb.Symp.Quant.Biol.* 2009a; 74:225–234. [PubMed: 20453205]
- Malone CD, Hannon GJ. Small RNAs as guardians of the genome. *Cell.* 2009b; 136:656–668. [PubMed: 19239887]
- Mattick JS. The functional genomics of noncoding RNA. *Science.* 2005; 309:1527–1528. [PubMed: 16141063]
- Maze I, Noh KM, Allis CD. Histone regulation in the CNS: basic principles of epigenetic plasticity. *Neuropsychopharmacology.* 2013; 38:3–22. [PubMed: 22828751]
- Meister G. Argonaute proteins: functional insights and emerging roles. *Nat.Rev.Genet.* 2013; 14:447–459. [PubMed: 23732335]
- Mellios N, Sugihara H, Castro J, Banerjee A, Le C, Kumar A, Crawford B, Strathmann J, Tropea D, Levine SS, Edbauer D, Sur M. miR-132, an experience-dependent microRNA, is essential for visual cortex plasticity. *Nat.Neurosci.* 2011; 14:1240–1242. [PubMed: 21892155]

- Memczak S, Jens M, Elefsinioti A, Torti F, Krueger J, Rybak A, Maier L, Mackowiak SD, Gregersen LH, Munschauer M, Loewer A, Ziebold U, Landthaler M, Kocks C, le NF, Rajewsky N. Circular RNAs are a large class of animal RNAs with regulatory potency. *Nature*. 2013; 495:333–338. [PubMed: 23446348]
- Mercer TR, Dinger ME, Sunkin SM, Mehler MF, Mattick JS. Specific expression of long noncoding RNAs in the mouse brain. *Proc.Natl.Acad.Sci.U.S.A.* 2008; 105:716–721. [PubMed: 18184812]
- Miska EA, Alvarez-Saavedra E, Townsend M, Yoshii A, Sestan N, Rakic P, Constantine-Paton M, Horvitz HR. Microarray analysis of microRNA expression in the developing mammalian brain. *Genome Biol.* 2004; 5:R68. [PubMed: 15345052]
- Modarresi F, Faghihi MA, Lopez-Toledano MA, Fatemi RP, Magistri M, Brothers SP, van der Brug MP, Wahlestedt C. Inhibition of natural antisense transcripts in vivo results in gene-specific transcriptional upregulation. *Nat.Biotechnol.* 2012; 30:453–459. [PubMed: 22446693]
- Morel L, Regan M, Higashimori H, Ng SK, Esau C, Vidensky S, Rothstein J, Yang Y. Neuronal exosomal miRNA-dependent translational regulation of astroglial glutamate transporter GLT1. *J.Biol.Chem.* 2013; 288:7105–7116. [PubMed: 23364798]
- Nakazawa T, Kuriu T, Tezuka T, Umemori H, Okabe S, Yamamoto T. Regulation of dendritic spine morphology by an NMDA receptor-associated Rho GTPase-activating protein, p250GAP. *J.Neurochem.* 2008; 105:1384–1393. [PubMed: 18331582]
- Natera-Naranjo O, Aschrafi A, Gioio AE, Kaplan BB. Identification and quantitative analyses of microRNAs located in the distal axons of sympathetic neurons. *RNA*. 2010; 16:1516–1529. [PubMed: 20584895]
- Ng SY, Lin L, Soh BS, Stanton LW. Long noncoding RNAs in development and disease of the central nervous system. *Trends Genet.* 2013; 29:461–468. [PubMed: 23562612]
- Nudelman AS, DiRocco DP, Lambert TJ, Garelick MG, Le J, Nathanson NM, Storm DR. Neuronal activity rapidly induces transcription of the CREB-regulated microRNA-132, in vivo. *Hippocampus*. 2010; 20:492–498. [PubMed: 19557767]
- Osato N, Suzuki Y, Ikeo K, Gojobori T. Transcriptional interferences in cis natural antisense transcripts of humans and mice. *Genetics*. 2007; 176:1299–1306. [PubMed: 17409075]
- Peleg S, Sananbenesi F, Zovoilis A, Burkhardt S, Bahari-Javan S, Agis-Balboa RC, Cota P, Wittmann JL, Gogol-Doering A, Opitz L, Salinas-Riester G, Dettenhofer M, Kang H, Farinelli L, Chen W, Fischer A. Altered histone acetylation is associated with age-dependent memory impairment in mice. *Science*. 2010; 328:753–756. [PubMed: 20448184]
- Peng JC, Lin H. Beyond transposons: the epigenetic and somatic functions of the Piwi-piRNA mechanism. *Curr.Opin.Cell Biol.* 2013; 25:190–194. [PubMed: 23465540]
- Peng Z, Cheng Y, Tan BC, Kang L, Tian Z, Zhu Y, Zhang W, Liang Y, Hu X, Tan X, Guo J, Dong Z, Liang Y, Bao L, Wang J. Comprehensive analysis of RNA-Seq data reveals extensive RNA editing in a human transcriptome. *Nat.Biotechnol.* 2012; 30:253–260. [PubMed: 22327324]
- Pitcher GM, Beggs S, Woo RS, Mei L, Salter MW. ErbB4 is a suppressor of long-term potentiation in the adult hippocampus. *Neuroreport*. 2008; 19:139–143. [PubMed: 18185097]
- Pruunsild P, Kazantseva A, Aid T, Palm K, Timmusk T. Dissecting the human BDNF locus: bidirectional transcription, complex splicing, and multiple promoters. *Genomics*. 2007; 90:397–406. [PubMed: 17629449]
- Qureshi IA, Mattick JS, Mehler MF. Long non-coding RNAs in nervous system function and disease. *Brain Res.* 2010; 1338:20–35. [PubMed: 20380817]
- Qureshi IA, Mehler MF. Emerging roles of non-coding RNAs in brain evolution, development, plasticity and disease. *Nat.Rev.Neurosci.* 2012; 13:528–541. [PubMed: 22814587]
- Rajasethupathy P, Antonov I, Sheridan R, Frey S, Sander C, Tuschl T, Kandel ER. A role for neuronal piRNAs in the epigenetic control of memory-related synaptic plasticity. *Cell*. 2012; 149:693–707. [PubMed: 22541438]
- Ravasi T, Suzuki H, Pang KC, Katayama S, Furuno M, Okunishi R, Fukuda S, Ru K, Frith MC, Gongora MM, Grimmond SM, Hume DA, Hayashizaki Y, Mattick JS. Experimental validation of the regulated expression of large numbers of non-coding RNAs from the mouse genome. *Genome Res.* 2006; 16:11–19. [PubMed: 16344565]

- Redfern AD, Colley SM, Beveridge DJ, Ikeda N, Epis MR, Li X, Foulds CE, Stuart LM, Barker A, Russell VJ, Ramsay K, Kobelke SJ, Li X, Hatchell EC, Payne C, Giles KM, Messineo A, Gatignol A, Lanz RB, O'Malley BW, Leedman PJ. RNA-induced silencing complex (RISC) Proteins PACT, TRBP, and Dicer are SRA binding nuclear receptor coregulators. *Proc.Natl.Acad.Sci.U.S.A.* 2013; 110:6536–6541. [PubMed: 23550157]
- Sasaki Y, Gross C, Xing L, Goshima Y, Bassell GJ. Identification of axon-enriched microRNAs localized to growth cones of cortical neurons. *Dev.Neurobiol.* 2013 Epub ahead of print 04/29/2013.
- Saxe JP, Lin H. Small noncoding RNAs in the germline. *Cold Spring Harb.Perspect.Biol.* 2011; 3:a002717. [PubMed: 21669983]
- Scott HL, Tamagnini F, Narduzzo KE, Howarth JL, Lee YB, Wong LF, Brown MW, Warburton EC, Bashir ZI, Uney JB. MicroRNA-132 regulates recognition memory and synaptic plasticity in the perirhinal cortex. *Eur.J.Neurosci.* 2012; 36:2941–2948. [PubMed: 22845676]
- Sempere LF, Freemantle S, Pitha-Rowe I, Moss E, Dmitrovsky E, Ambros V. Expression profiling of mammalian microRNAs uncovers a subset of brain-expressed microRNAs with possible roles in murine and human neuronal differentiation. *Genome Biol.* 2004; 5:R13. [PubMed: 15003116]
- Shamir A, Kwon OB, Karavanova I, Vullhorst D, Leiva-Salcedo E, Janssen MJ, Buonanno A. The importance of the NRG-1/ErbB4 pathway for synaptic plasticity and behaviors associated with psychiatric disorders. *J.Neurosci.* 2012; 32:2988–2997. [PubMed: 22378872]
- Siegel G, Obernosterer G, Fiore R, Oehmen M, Bicker S, Christensen M, Khudayberdiev S, Leuschner PF, Busch CJ, Kane C, Hubel K, Dekker F, Hedberg C, Rengarajan B, Drepper C, Waldmann H, Kauppinen S, Greenberg ME, Draguhn A, Rehmsmeier M, Martinez J, Schratt GM. A functional screen implicates microRNA-138-dependent regulation of the depalmitoylation enzyme APT1 in dendritic spine morphogenesis. *Nat.Cell Biol.* 2009; 11:705–716. [PubMed: 19465924]
- Silva J, Mak W, Zvetkova I, Appanah R, Nesterova TB, Webster Z, Peters AH, Jenuwein T, Otte AP, Brockdorff N. Establishment of histone h3 methylation on the inactive X chromosome requires transient recruitment of Eed-Enx1 polycomb group complexes. *Dev.Cell.* 2003; 4:481–495. [PubMed: 12689588]
- Smalheiser NR, Lugli G, Torvik VI, Mise N, Ikeda R, Abe K. Natural antisense transcripts are co-expressed with sense mRNAs in synaptoneurosomes of adult mouse forebrain. *Neurosci.Res.* 2008; 62:236–239. [PubMed: 18812194]
- Tetta C, Ghigo E, Silengo L, Deregibus MC, Camussi G. Extracellular vesicles as an emerging mechanism of cell-to-cell communication. *Endocrine.* 2013; 44:11–19. [PubMed: 23203002]
- Thomson T, Lin H. The biogenesis and function of PIWI proteins and piRNAs: progress and prospect. *Annu.Rev.Cell Dev.Biol.* 2009; 25:355–376. [PubMed: 19575643]
- Tiedge H, Chen W, Brosius J. Primary structure, neural-specific expression, and dendritic location of human BC200 RNA. *J.Neurosci.* 1993; 13:2382–2390. [PubMed: 7684772]
- Tiedge H, Freneau RT Jr, Weinstock PH, Arancio O, Brosius J. Dendritic location of neural BC1 RNA. *Proc.Natl.Acad.Sci.U.S.A.* 1991; 88:2093–2097. [PubMed: 1706516]
- Ulitisky I, Shkumatava A, Jan CH, Sive H, Bartel DP. Conserved function of lincRNAs in vertebrate embryonic development despite rapid sequence evolution. *Cell.* 2011; 147:1537–1550. [PubMed: 22196729]
- Vo N, Klein ME, Varlamova O, Keller DM, Yamamoto T, Goodman RH, Impey S. A cAMP-response element binding protein-induced microRNA regulates neuronal morphogenesis. *Proc.Natl.Acad.Sci.U.S.A.* 2005; 102:16426–16431. [PubMed: 16260724]
- Wang IT, Reyes AR, Zhou Z. Neuronal morphology in MeCP2 mouse models is intrinsically variable and depends on age, cell type, and Mecp2 mutation. *Neurobiol.Dis.* 2013; 58:3–12. [PubMed: 23659895]
- Wayman GA, Davare M, Ando H, Fortin D, Varlamova O, Cheng HY, Marks D, Obrietan K, Soderling TR, Goodman RH, Impey S. An activity-regulated microRNA controls dendritic plasticity by down-regulating p250GAP. *Proc.Natl.Acad.Sci.U.S.A.* 2008; 105:9093–9098. [PubMed: 18577589]
- Wibrand K, Panja D, Tiron A, Ofte ML, Skaftnesmo KO, Lee CS, Pena JT, Tuschl T, Bramham CR. Differential regulation of mature and precursor microRNA expression by NMDA and

- metabotropic glutamate receptor activation during LTP in the adult dentate gyrus in vivo. *Eur.J.Neurosci.* 2010; 31:636–645. [PubMed: 20384810]
- Yan Z, Hu HY, Jiang X, Maierhofer V, Neb E, He L, Hu Y, Hu H, Li N, Chen W, Khaitovich P. Widespread expression of piRNA-like molecules in somatic tissues. *Nucleic Acids Res.* 2011; 39:6596–6607. [PubMed: 21546553]
- Zeng Y, Tan M, Kohyama J, Sneddon M, Watson JB, Sun YE, Xie CW. Epigenetic enhancement of BDNF signaling rescues synaptic plasticity in aging. *J.Neurosci.* 2011; 31:17800–17810. [PubMed: 22159096]
- Zovkic IB, Guzman-Karlsson MC, Sweatt JD. Epigenetic regulation of memory formation and maintenance. *Learn.Mem.* 2013; 20:61–74. [PubMed: 23322554]

Figure 1. *miR-132* and *miR-134* in age-related memory pathways

The sirtuin SIRT1 silences the *miR-134* promoter either by deacetylating the histones, which allows for formation of higher-order chromatin structure (shown), or by targeting acetyl groups on other unknown proteins. In the absence of SIRT1, *miR-134* transcription increases (Gao et al., 2010), and *miR-134* suppresses translation of the transcription factor Creb by binding to seed sites in its 3' UTR. Creb mediates the transcription of *Bdnf* and *miR-132* (Impey et al., 2004), both of which positively influence synaptic plasticity, memory, and

dendritic spine structure. Several components of this pathway are influenced by aging or disease and might play important roles in age-associated memory loss.

Figure 2. Mechanisms used by ncRNAs to regulate protein expression

In the nucleus, piRNAs inhibit gene transcription by histone methylation (red flags) and DNA methylation at CpG islands (solid purple circles) (Rajasethupathy et al., 2012; Peng and Lin, 2013). Histone methylation is also induced by lncRNAs (Jeon and Lee, 2011). In addition, lncRNAs can regulate alternative splicing (Barry et al., 2013) and inhibit mRNA transcript elongation (Osato et al., 2007). In the cytoplasm, *BC1* and *BC200* RNAs interact with translation initiation factors to repress protein translation (Iacoangeli and Tiedge, 2013). miRNAs repress mRNA translation and induce mRNA degradation (Czech and Hannon, 2011). miRNA function is inhibited by circRNAs, which are endogenous sponges for miRNAs (Hansen et al., 2013b; Memczak et al., 2013). The formation of a nucleotide duplex between the mRNA 3' UTR and lncRNAs leads to the degradation of mRNAs (Gong and Maquat, 2011).