AUTHOR QUERY FORM

ELSEVIER	**Journal:** TRMOME **Article Number:** 719	**Please e-mail or fax your responses and any corrections to:** **E-mail:** corrections.esnl@elsevier.thomsondigital.com **Fax:** + 31 20 485 2521

Dear Author,

Please check your proof carefully and mark all corrections at the appropriate place in the proof (e.g., by using on-screen annotation in the PDF file) or compile them in a separate list. To ensure fast publication of your paper please return your corrections within 48 hours.

For correction or revision of any artwork, please consult http://www.elsevier.com/artworkinstructions.

Please note that the figure resolution in these proofs is lower than the original figures to avoid problems with emailing large file sizes. The figure resolution in the final publication will be equivalent to the original figure submission.

Any queries or remarks that have arisen during the processing of your manuscript are listed below and highlighted by flags in the proof. Click on the 'Q' link to go to the location in the proof.

Location in article	Query / Remark: **click on the Q link to go** **Please insert your reply or correction at the corresponding line in the proof**
Q1	References should be cited in the order Text-Figures-Tables-Boxes. [61] was changed to [59] here. See change to reference in Box 1 and Box 2 also. Please check all citations carefully.
Q2	See comment for Figure 1 legend regarding renumbering of citations 59, 60, 61.
Q3	See comment for Figure 1 legend regarding renumbering of citations 59, 60, 61.

Thank you for your assistance.

Opinion

Cell PRESS

NeurimmiRs: microRNAs in the neuroimmune interface

Hermona Soreq and Yochai Wolf

Institute of Life Sciences and Edmond and Lily Safra Center of Brain Sciences, Hebrew University of Jerusalem, Edmond J. Safra Campus, Jerusalem 91904, Israel

Recent reports of microRNA (miR) modulators of both neuronal and immune processes (here termed NeurimmiRs) predict therapeutic potential for manipulating NeurimmiR levels in diseases affecting both the immune system and higher brain functions, such as Alzheimer's disease (AD), Parkinson's disease (PD), multiple sclerosis (MS) and anxiety-related disorders. In our opinion, NeurimmiRs that function within both the nervous and the immune systems, such as miR-132 and miR-124, might act as 'negotiators' between these two interacting compartments. We suggest that NeurimmiRs primarily target transcriptional or other regulatory genes, which enables modulation of both immune and cognitive processes through direct or indirect alterations of neuron–glia and/or brain-to-body signaling. Thus, manipulating NeurimmiR control over the immune contributions to cognitive pathways might offer new therapeutic targets.

> 'A healthy body is the guest-chamber of the soul; a sick, its prison.'
>
> Francis Bacon

MicroRNAs can modulate both neuronal and immune functions

MicroRNAs (miRs) are increasingly appreciated as being involved in all aspects of cellular functioning in a wide range of eukaryotic organisms [1]. However, although most miRs have multiple predicted targets, the majority of current miR studies focus on experimental validation of only one target. To the best of our knowledge, no hierarchy of miR–target interactions has yet been published for any miR. Furthermore, numerous mRNAs carry multiple putative binding sites for many miRs, but the experimental validation of miR–target relationships is rather laborious. Therefore, most studies have validated only one of those miRs experimentally. Also, whether or not any of those miR–target interactions override the others, and the underlying principles for such putative competition, remain unknown (Box 1).

The brain modulates immune functions using neurotransmitters, neuropeptides and hormones, senses peripheral inflammation by detecting inflammatory agents (such as proinflammatory cytokines and other immune regulators), and can potentiate or counteract inflammation in a 'top-down' effect [2]. Reciprocally, the immune system can affect the brain in a 'bottom-up' manner, and converging lines of evidence highlight the role of neuroinflammation in both psychiatric and neurodegenerative diseases [3]. Of particular interest are microglia, myeloid cells of the monocyte/macrophage lineage that reside within the central nervous system (CNS) and thus are prime candidates for mediating communication between the two systems (Box 2). Furthermore, 'classic' immune proteins, such as MHC class I and complement factors, are considered to play new roles within the nervous system [4], suggesting that these interacting compartments might also share common miRs.

In the mammalian immune system, miRs control differentiation as well as innate and adaptive immune responses [5]. In the central nervous system, miRs are involved in diverse functions, including neuronal development, plasticity and physiological reactions [6]. A subset of these miRs, here designated NeurimmiRs, notably affects both immune and neuronal functions. Given that the validation of many miR targets is far from complete, appreciating the full neuroimmune impact of NeurimmiRs is in its infancy. Nevertheless, outlining the existence and assessing the scope of NeurimmiRs involved in neuroinflammatory processes can assist in evaluating the therapeutic potential of manipulating their levels in 'classic' neuroimmune disorders, neurodegenerative diseases and psychiatric syndromes. Below, we suggest several putative candidates for this new class of miRs and focus on one of them, miR-132, as an exemplar NeurimmiR with far-reaching implications.

We suggest that the co-occurrence of NeurimmiRs in the brain and peripheral organs has a function in the crosstalk between these two compartments, both locally (i.e. brain inflammation) or in a broad context (regulation of inflammation via the autonomous nervous system). Many miRs have a broad affect within a certain tissue owing to their regulation of transcription factors and other regulatory elements. Therefore, various scenarios might exist in which the expression of a miRNA 'shared' by two tissues is altered. This in turn might alter gene expression in either one or both of these compartments. Correspondingly, impaired miR levels were found in many nervous system diseases [7]. Also, miRs are involved in hematopoiesis [5] and in the immune components of diverse neural diseases such as epilepsy or Alzheimer's disease (AD) [8,9]. We also propose that immune insults change the levels of NeurimmiRs. Owing to the regulatory roles of these miRs, such changes often alter inflammation-responsive transcripts within the brain [e.g. multiple sclerosis (MS)] [10],

Corresponding author: Soreq, H. (soreq@cc.huji.ac.il).

Box 1. The biogenesis and functions of miRs

MiRs are short non-protein-coding RNAs with both conserved and species-specific members. MiRs regulate transcription, mRNA stability and translation and are critical for the expression and epigenetic control of at least one-half to two-thirds of all known genes and biochemical pathways [33]. miR precursors (stem-and-loop molecules generated from a primary transcript) are cleaved to mature 22-base pair double-stranded molecules, with one strand usually guiding the complex to a partially complementary sequence often found in the 3′-untranslated region (3′-UTR) of target genes. The miR 5′-end 'seed' region determines target specificity and dictates mRNA degradation and translational repression, whereas mRNA cleavage is dependent on perfect complementary base-pairing at positions 10/11 as well as on target sites located at the center of the miR [55]. Classically, every miR can bind to its mRNA targets by incomplete base pairing, with only the short, approximately seven nucleotides long 'seed' region displaying full complementation; therefore, each miR can potentially target hundreds of transcripts and achieve network-level regulation of gene expression [60]. MiR regulation takes place post-transcriptionally, and is thought to perform rapid fine-tuning rather than total silencing or induction of targets ('on' and 'off' switching). Furthermore, repeated binding motifs enable one miR to target several mRNAs involved in a particular pathway and achieve gene-network-level regulation. Although a particular miR often affects only a few targets in specific cell types, the potential multiplicity of gene targeting makes assessing and proving that a specific gene or pathway is indeed a target of a particular miR an elusive and nontrivial task. A major disadvantage in miR research is the difficulty in validating putative target mRNAs identified for a specific miR; a variety of online algorithms that utilize different approaches to predict miR–mRNA interactions exist, but the predicted targets for each miR might be dissimilar and largely non-overlapping and often exclude many experimentally proven and validated targets. Thus, current bioinformatics tools, as well as the understanding of miR biology itself, are still in their infancy. In many cases, a target-based search for complementary miRs [34] might be more useful than a miR screen for targets.

or in peripheral leukocytes. In the periphery, this would alter complement factors, adhesion molecules, reactive oxygen species and proinflammatory cytokines, facilitate crossing of the blood–brain-barrier (BBB) and influence a broad spectrum of neuronal processes such as neurotransmission, synapse morphology and cognition [11].

NeurimmiR-modulated processes link immune and neuronal functions

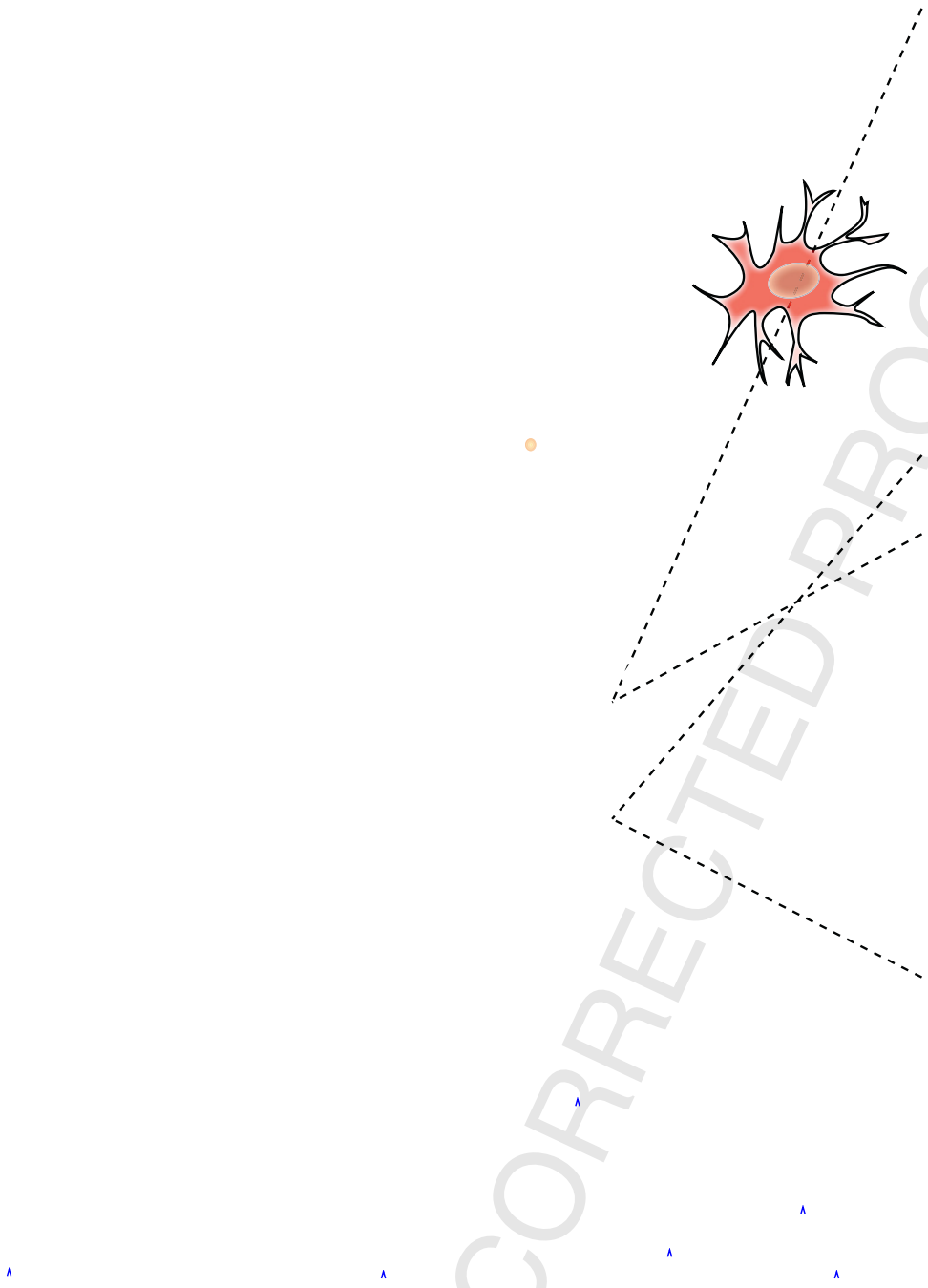
Several NeurimmiR-related processes involve measurable developmental or deterioration changes of neuronal and hematopoietic pathways. Figure 1 schematically presents how NeurimmiR functioning might be viewed as operating within and between the neural and immune worlds, with each interaction affecting both cognitive and immune features in a multi-layered fashion. A prominent example is the brain-enriched miR-124, which enhances neuronal maturation by targeting the transcription factor sox9 [12], inhibits the neuronal transcription regulator complex REST involved in the neurodevelopment disorder Rett syndrome [13], modulates serotonin-dependent long-term potentiation (LTP) in sensory-motor neurons [14] and is involved in regulating hippocampal-induced plasticity under chronic cocaine intake [15]. In the hematopoietic compartment, miR-124 is exclusively expressed in microglia and CNS macrophages where it supposedly targets the transcription factor C/EBP-α and therefore also its downstream gene, the myeloid cell-associated transcription factor Pu.1. A recent study reports a miR-124-mediated microglial phenotype switch from an inflammatory ($CD45^{hi}MHCII^{+}$) to a quiescent state ($CD45^{lo}MHCII^{-}$). This successfully inhibits the onset of experimental autoimmune encephalomyelitis (EAE), a rodent model of MS [16]. Furthermore, miR-124 targets glucocorticoid receptors, an important component of stress responses and thus might contribute to controlling stress disorders [17]. As is often the case, none of these studies addresses the miR-124 targets studied by others, so that comparative analysis of the different processes in which this miR might be involved is virtually impossible at the present time.

Another brain-enriched REST-targeting NeurimmiR, miR-9, shows decreases in Huntington's disease [18]. By

Box 2. Neuroinflammation, cognition and the anti-inflammatory reflex

Neuroinflammation is an immune response initiated by the innate immunity arm of the CNS, and sometimes mediated by the adaptive arm, which involves increases in proinflammatory cytokines, chemokines, complement factors, reactive oxygen species and other agents, secreted by either resident or infiltrating leukocytes. In human patients, neuroinflammation is triggered by injury and aging, and peripheral immune challenges emerge as being involved in a wide range of CNS diseases such as AD, MS, PD and ALS [3]. Proinflammatory cytokines affect hippocampal LTP, neuronal differentiation and neurogenesis. Elevated levels of such cytokines have been found in the CNS of aged and post-traumatic stress disorder (PTSD) patients, particularly in brain regions associated with learning, memory and cognition. Compatible with this, inflammation affects emotions and mood, causing a depression-like state. Moreover, systemic inflammation increases cytokine production, most importantly IL-1β, which is capable of modulating the HPA axis, thereby enhancing anxiety-initiating body-to-brain signals [2]. The brain's immune system consists of various subsets of macrophage-associated cells, most notably the microglia [61]. Experimental exposure to proinflammatory cytokines such as IL-1β, protein aggregates such as Aβ or self antigens such as the MOG peptide induces activation of astrocytes and microglia, leading to further expression of cytokines and leukocyte recruitment in a feed-forward loop. Following peripheral LPS injection, such increases are accompanied by impaired cognitive functions (e.g. hippocampus-dependent learning tasks) and altered neuronal differentiation. Brain-infiltrating leukocytes secrete proinflammatory cytokines, reactive oxygen species and others that might both be detrimental to neurons and activate surrounding resting microglia, suggesting involvement in disease initiation and progression by the interlocked neuroimmune communication pathways.

The ascending fibers of the vagus nerve can signal the presence of peripheral inflammation to the brain, through cytokine receptors expressed by parasympathetic ganglia cells [2]. Reciprocally, macrophages and dendritic cells, T cells and B cells can react to ACh released from the descending fibers of the vagus nerve or from internal or peripheral sources, such as the splenic nerve. In cells exposed to inflammatory insults, NF-κ-B translocates into the nucleus, and downstream inflammatory products are expressed and secreted. Cholinergic activation of the nicotinic α7 receptor interferes with this translocation and limits the secretion of proinflammatory cytokines. Overall, ACh has an anti-inflammatory effect, dubbed 'the cholinergic anti-inflammatory reflex' [2]. By contrast, extensive ACh hydrolysis by AChE exerts a proinflammatory output; thus, counteracting AChE activity by miR-132 exerts a net anti-inflammatory effect [34].



regulating the transcription factor FoxP1, miR-9 promotes spinal columnar organization [19]. Correspondingly, miR-9 is downregulated in an animal model of spinal muscular atrophy (SMA) [20]. By destabilizing a splice variant of the large-conductance calcium- and voltage-activated potassium channel (BK) [21] miR-9 was proposed to modulate neuroadaptation to alcohol. In myeloid cells, miR-9 targets the vital inflammatory transcription factor NF-κ-B1 and is upregulated following exposure to activators of the innate immune toll-like receptors (TLRs) or proinflammatory cytokines [22]. Similarly bifunctional is miR-125b, which modulates the morphology and excitability of neurons through its Fragile X mental retardation protein (FMRP) target [23], and was recently shown, in an exceptionally wide-scoped study, to promote neurite growth through its capacity to target multiple transcripts. MiR-125b also modulates lymphoid cell differentiation and targets the mRNA of the critical proinflammatory cytokine TNF-α, suggesting a significant role in the regulation and resolution of inflammation [24].

Following peripheral or central immune insults, immune cell changes in NeurimmiR levels might influence neuronal functions, whether directly by suppressing genes within neurons or indirectly by influencing the functioning of immune or supporting glial cells. For example, miR-155 regulates T cell and B cell differentiation as well as

dendritic cell function by targeting the SHIP-1 inositol phosphatase and controlling IL-1 signaling [25]. In a human-derived neuronal cell line, the mature MiR-155 can also target the methyl-CpG binding protein 2 (MeCP2), a co-activator of REST, and is deregulated in Down and Rett's syndrome and other neurodevelopmental diseases [26]. Mice that are null for *miR155* (*miR155*$^{-/-}$) show significantly reduced numbers of encephalogenic CD4$^+$ Th17 cells, an inflammatory T cell subset known to have a significant role in MS and react to the neuroinflammatory stimulus of the MOG peptide by increased proliferation of CD4$^+$ Th17 cells. Also, *miR155*$^{-/-}$ CD4$^+$ T cells failed to passively induce the EAE experimental model of MS when transferred to lymphocyte-deficient animals [25]. In addition, impaired regulation of SHIP-1 and the suppressor of cytokine signaling 1 (SOCS1) in *miR155*$^{-/-}$ dendritic cells reduces the Th17 regulating cytokines IL-6 and IL-23. A similar role was proposed for miR-326, which regulates the Th17 cells by targeting the transcription factor Est-1 [27]. Both miRs were overexpressed in astrocytes derived from MS lesions in human patients and were suggested to modulate phagocytosis of neighboring neurons by regulating the macrophage/microglial SIRPα protein [28]. By deregulating Th17 and myeloid cell functions, miR-155 and miR-326 emerge as MS-associated NeurimmiRs.

Other NeurimmiR examples include the NF-κ-B-dependent miR-146a, which is upregulated in the brain of AD patients and might enhance inflammation by targeting the inflammation-repressing complement factor H (CFH) [9]. MiR-146a is also upregulated in the hippocampus of both temporal lobe epilepsy patients and a rat epilepsy model [8]. Under HIV-1 infection, miR-146a elevation in human microglia supports the fine-tuning of immune functions by targeting the chemokine CCL8/MCP-2 [29]. Within the TLR innate immune pathway, miR146a suppresses the TRAF6 and IRAK1/2 proteins, potentially enhancing RIG-I mediated interferon-1 (IFN-1) production [5] and suggesting multiple routes contributing to microglial function.

Of note, different NeurimmiRs show overlapping patterns of immune-related changes: LPS-exposed human monocytes show upregulation of both miR-146a and miR-155, whereas TNF-α exposure upregulates miR-155 and miR-125b [24]. Similar to miR-155, miR-146a is also associated with MeCP2 because this miR is downregulated in the Rett syndrome model (*MeCP2*$^{-/-}$ animals) [30]. Furthermore, MecP2 regulates the expression of miRs [31] and miR-146a can downregulate the IL-1 signaling protein IRAK1 both in neurons and in myeloid cells, suggesting its involvement in the neuroimmune crosstalk by IL-1. The evidence accumulated so far is compatible with the theory that a single NeurimmiR affects a pathway of inter-related transcripts, all involved in a cellular process, rather than single proteins. Because of the tendency of miR studies to focus on one protein–one miR interaction, the global miR effect on different pathways is often overlooked. One such example is miR-1, which was shown to coregulate several cholinergic-signaling related proteins in the neuromuscular junction of *Caenorhabditis elegans* [32]. Others proposed that the capacity of several miRs to target key regulatory transcripts associated with the same process ensures tight regulation [33]. Thus, the complete scope of NeurimmiRs functioning in the neuroimmune interface appears to be central.

Inflammation controlling NeurimmiRs are implicated in multiple syndromes

Inflammation is primarily aimed at fighting invading pathogens and is terminated by reciprocal brain-to-body signals. Most elderly individuals show excessive inflammation, suggesting failure of their natural control mechanisms [2]. In the mammalian brain, aging is associated with miR changes which have been associated with increases in inflammatory markers in astrocytes, microglia and neurons, as well as with reduced neurogenesis, increased neuron death and neurodegenerative processes [3,7]. The causes for the heightened inflammatory state in advanced age or the impact of inflammation on aging in general are not well understood, but NeurimmiRs that target transcripts involved in the neuroimmune interface (e.g. miR-125b, miR-132 and miR-146a) are probably involved, which makes them promising targets for interference with the hope of reducing age-associated inflammation.

Recent findings demonstrate that small RNAs functioning within the neuroimmune interface play a significant role in controlling the bidirectional communication line between neuronal circuits and inflammatory processes [34]. For example, in Parkinson's disease (PD), where neuroinflammation is a central component, mutated LRRK2 globally interferes with miR-mediated processes [35], whereas Let-7 targets the PD hallmark fibril-forming protein α-synuclein [36]. In amyotrophic lateral sclerosis (ALS), upregulating miR-206 was shown to reinnervate damaged neuromuscular junctions by targeting histone deacetylase 4 [37]. Specifically, NeurimmiR alterations under peripheral or central inflammation are compatible with the hypothesis that these changes contribute to the emergence of several neurodegenerative and neuropsychiatric conditions (Table 1). Peripheral immune insults and/or BBB promiscuity [11] induce the secretion and brain penetration of inflammatory agents and leukocytes, which would consequently modify NeurimmiRs expression in the CNS. Although the direct association of such miR changes with neuronal function and malfunction is yet to be confirmed, NeurimmiRs are primarily adapted for such body-to-brain signaling in the rapidly growing field of neuroimmunity. For exploring the relevance of this phenomenon, we can turn to the well-investigated brain-enriched miR-132, which was implicated with body-to-brain communication [34], as a case study.

MiR-132 transcription is controlled both by the cyclic-AMP response element binding protein (CREB) [34], a transcription factor classically involved in learning and memory as well as in neural growth, and by the neuronal growth factor BDNF that is associated with cholinergic functioning. MiR-132 transfection suppresses the Rho family GTPase activating protein p250GAP while enhancing neurite sprouting in cortical neurons, compatible with miR-132 upregulation during postnatal development and with the attenuation of neurite growth by miR-132-targeted antisense oligonucleotides [38]. MiR-132 was also predicted to target several ion channels, and thus affect cell excitability; correspondingly, overexpressed pri-miR-132

Table 1. Relevance of NeurimmiRs for various neuropathologies

miRNA	Neuropathology	Direction of expression change	Validated target	Physiological outcome	Refs
miR-9	Huntington's disease	Downregulation	REST	Disrupted regulation of neuronal gene expression	[18]
	SMA	Downregulation	Heavy neurofilament subunit	Disrupted axonal cytoskeleton	[20]
	AD	Downregulation	?	?	[48]
miR-124	EAE	Downregulation	C/EBP-α	Over-activation of microglia and infiltrating macrophages	[16]
miR-125b	AD	Upregulated	?	?	[48]
miR-132	Huntington's disease	Upregulation	?	?	[18]
	AD	Downregulation	?	?	[48]
	Cerebral ischemia	Downregulation (following ischemic preconditioning)	MeCP2	Neuroprotection against subsequent ischemic insults	[53]
	Schizophrenia/bipolar disorder	Upregulation	?	?	[51]
miR-146a	AD	Upregulation	CFH	Enhanced inflammatory response in neocortex and hippocampus of AD patients	[9]
	Epilepsy	Upregulation	?	?	[8]
	HIV-encephalitis	Upregulation	MCP-2	Regulation of excessive inflammatory response during reaction to viral infection	[29]
	Rett syndrome	Downregulation	IRAK-1	?	[30]
miR-155	Down's syndrome	Multiple copies[a]	MeCP2	Broad disruption of gene expression as a result of aberrant expression of transcription factors downstream of MeCP2[b]	[26]
	MS/EAE	Upregulation	CD47, 73P1	Aberrant 'don't eat me signal' to macrophages/ microglia; aberrant regulation of Th17 subset via dentritic cell signaling	[25,28]
miR-212	Schizophrenia/bipolar disorder	Upregulation	?	?	[51]
	AD	Downregulation	?	?	[48]
miR-326	MS/EAE	Upregulation	CD47, Est-1	Aberrant 'don't eat me signal' to macrophages/ microglia; deregulated shift of Th0 cells to Th17 cells	[27,28]

[a]MiR-155 resides within chromosome 21, which is tripled in Down's syndrome.

[b]MeCP2 is also relevant for Rett's syndrome, although the miRNA is not yet reported to be directly involved in the disease.

potentiates glutamate, NMDA, or K^+-mediated depolarization of cultured neurons, suggesting global involvement in regulating neurotransmission and plasticity. However, involvement of other putative miR-132 targets has not been tested in these experiments, which leaves the detailed mechanisms underlying the neuronal growth effects of miR-132 incompletely understood.

In transfected cultured neurons, miR-132 leads to wider and shorter dendritic spines, accompanied by increased miniature excitatory postsynaptic currents [23]. In newborn hippocampal neurons, Cre-lox mediated deletion of miR-132 decreases dendritic length and arborization in adult mice [39], supporting a role for miR-132 in neuronal differentiation, synaptogenesis and maintenance. Conversely, cells transfected with the TNF-α-inducible miR-125b extend long and thin spines and show reduced excitatory postsynaptic currents. Both miR-132 and miR-125b were co-immunoprecipitated with FMRP, and FMRP knockdown abolished the morphological effects of both miR-132 and miR-125b. This suggested that miR-132 and miR-125b balance each other's effects through competitive interaction with FMRP [23]. Compatible with the stress-associated alteration of cholinergic signaling, miR-132 was upregulated in the hippocampus of chronically stressed rats [40]. Another miR-132 target is the light-induced transcription regulatory factor X 4 (RFX4), abundant in the suprachiasmatic nucleus (SCN) of the hypothalamus, which regulates biological clocks and rhythms. MiR-132 levels are higher during the light part of the circadian cycle in the SCN [41]. Infused antisense oligonucleotide against miR-132 (AntagoMir) impaired the resetting of the circadian clock while elevating RFX4. In this case as well, no other possible targets were measured.

Cultured human monocytes react to LPS by overexpressing the NeurimmiRs miR-146a, miR-155 and miR-132 [5]. Correspondingly, all three miRs are increased in the autoimmune disease rheumatoid arthritis [42]. In human lymphatic endothelial cells infected with Kaposi's sarcoma-associated virus, as well as in viral or bacterial-infected brain tissue, miR-132 induces transcriptional arrest by downregulating the CREB co-activator p300 [43]. This inhibits both inflammatory cytokines and antiviral genes, indicating that stress-induced facilitation of anti-inflammatory protection, which would also support cognitive alertness, might take its toll by compromising anti-viral immune functions. Because p300 is also a coactivator of the developing brain, this process might further intensify the damaging effects of viral infection in neuronal development.

At the body-to-brain communication level, miR-132 regulates cholinergic anti-inflammatory signaling by potentiating the cholinergic anti-inflammatory pathway (Box 2)

[34]. By targeting acetylcholinesterase (AChE), miR-132 lowers acetylcholine (ACh) hydrolysis. Both gain- and loss-of-function experiments validated the miR-132/AChE interaction; miR-132 overexpression by lentivirus infection reduces AChE activity, whereas mutagenesis of the miR-132 binding site in AChE mRNA elevates it. Correspondingly, in vivo antisense suppression of miR-132 levels increases AChE activity in the intestine, spleen and serum. Furthermore, mice expressing an AChE transgene devoid ofD00-.5669269.5813534.7841T715467-514322cm()Tj/F51Tf9.4646009.464668.536267-514322cm(he)Tj/F21Tf.566900-.566916.262

Biomedical tools that are being developed for miR modulations include, but are not limited to, chemically protected AntagoMir oligonucleotides for suppressing targeted miRs [34], miR mimics for potentiating their action [54] and lentiviral tools for long-term manipulations [43,55]. Further experiments must be undertaken to establish the efficacy of these potential minimally intrusive manipulations for neuroinflammatory conditions. Promising examples involve AD-related downregulation of the miR-29ab-1 cluster [56] and of miR-107 in a double transgenic mouse AD model [57], all associated with β-amyloid precursor protein-converting enzyme (BACE1), which presumably contributes to AD development, and the predicted regulation of BACE1 by miR-298 and miR-328 [58].

Concluding remarks

The field of miR research has undergone a wide-ranging expansion in the last decade. However, the multiple modes of function of these small RNAs have hampered the progress in understanding the full scope of their effects. This is particularly the case for miRs involved in neuroimmune maladies, which we have named NeurimmiRs. Both peripheral and central immune insults have already been shown to upregulate various NeurimmiRs, either in neurons, surrounding cells (e.g. glia, microglia and infiltrating leukocytes) or in peripheral leukocytes. We propose that miRs are involved in regulating neuroimmune functions both due to central control and by peripheral immune cells sending back messages; such messages alert the central command, leading to a complex set of bidirectional feedback consequences. Owing to their physical properties and multiple roles in the nervous and immune systems, NeurimmiRs might initiate such communication cascades both in health and disease and their manipulation can be therapeutically useful. To progress toward this goal, several open questions remain to be addressed (Box 3). By contributing to the monitoring by the brain of cognition and inflammation alike, as well as to the detrimental effects of systemic and chronic changes in immune functions in neurodegenerative diseases, these miRs might be prime candidates for therapeutic intervention for neuroimmune impairments.

Acknowledgments

The authors are grateful to Eyal Soreq, the Bezalel Art Academy, Jerusalem for creative art work. This study was supported by the Legacy Heritage Biomedical Science Partnership Program of the Israel Science Foundation (Grant No. 1799/10), the German Research Foundation (DFG) and the ROSETREES Foundation (to H.S.).

References

1 Flynt, A.S. and Lai, E.C. (2008) Biological principles of microRNA-mediated regulation: shared themes amid diversity. *Nat. Rev. Genet.* 9, 831–842
2 Tracey, K.J. (2010) Understanding immunity requires more than immunology. *Nat. Immunol.* 11, 561–564
3 Glass, C.K. *et al.* (2010) Mechanisms underlying inflammation in neurodegeneration. *Cell* 140, 918–934
4 Boulanger, L.M. (2009) Immune proteins in brain development and synaptic plasticity. *Neuron* 64, 93–109
5 O'Connell, R.M. *et al.* (2010) Physiological and pathological roles for microRNAs in the immune system. *Nat. Rev. Immunol.* 10, 111–122
6 Li, X. and Jin, P. (2010) Roles of small regulatory RNAs in determining neuronal identity. *Nat. Rev. Neurosci.* 11, 329–338
7 Eacker, S.M. *et al.* (2009) Understanding microRNAs in neurodegeneration. *Nat. Rev. Neurosci.* 10, 837–841
8 Aronica, E. *et al.* (2010) Expression pattern of miR-146a, an inflammation-associated microRNA, in experimental and human temporal lobe epilepsy. *Eur. J. Neurosci.* 31, 1100–1107
9 Lukiw, W.J. *et al.* (2008) An NF-kappaB-sensitive micro RNA-146a-mediated inflammatory circuit in Alzheimer disease and in stressed human brain cells. *J. Biol. Chem.* 283, 31315–31322
10 Tufekci, K.U. *et al.* (2011) MicroRNAs and multiple sclerosis. *Autoimmune Dis.* DOI: 10.4061/2011/807426
11 Shlosberg, D. *et al.* (2010) Blood–brain barrier breakdown as a therapeutic target in traumatic brain injury. *Nat. Rev. Neurol.* 6, 393–403
12 Cheng, L.C. *et al.* (2009) miR-124 regulates adult neurogenesis in the subventricular zone stem cell niche. *Nat. Neurosci.* 12, 399–408
13 Visvanathan, J. *et al.* (2007) The microRNA miR-124 antagonizes the anti-neural REST/SCP1 pathway during embryonic CNS development. *Genes Dev.* 21, 744–749
14 Rajasethupathy, P. *et al.* (2009) Characterization of small RNAs in aplysia reveals a role for miR-124 in constraining synaptic plasticity through CREB. *Neuron* 63, 803–817
15 Chandrasekar, V. and Dreyer, J.L. (2009) microRNAs miR-124, let-7d and miR-181a regulate cocaine-induced plasticity. *Mol. Cell Neurosci.* 42, 350–362
16 Ponomarev, E.D. *et al.* (2011) MicroRNA-124 promotes microglia quiescence and suppresses EAE by deactivating macrophages via the C/EBP-[alpha]-PU.1 pathway. *Nat. Med.* 17, 64–70
17 Vreugdenhil, E. *et al.* (2009) MicroRNA 18 and 124a down-regulate the glucocorticoid receptor: implications for glucocorticoid responsiveness in the brain. *Endocrinology* 150, 2220–2228
18 Packer, A.N. *et al.* (2008) The bifunctional microRNA miR-9/miR-9* regulates REST and CoREST and is downregulated in Huntington's disease. *J. Neurosci.* 28, 14341–14346
19 Otaegi, G. *et al.* (2011) MicroRNA miR-9 modifies motor neuron columns by a tuning regulation of FoxP1 levels in developing spinal cords. *J. Neurosci.* 31, 809–818
20 Haramati, S. *et al.* (2010) miRNA malfunction causes spinal motor neuron disease. *Proc. Natl. Acad. Sci. U.S.A.* 107, 13111–13116
21 Pietrzykowski, A.Z. *et al.* (2008) Posttranscriptional regulation of BK channel splice variant stability by miR-9 underlies neuroadaptation to alcohol. *Neuron* 59, 274–287
22 Bazzoni, F. *et al.* (2009) Induction and regulatory function of miR-9 in human monocytes and neutrophils exposed to proinflammatory signals. *Proc. Natl. Acad. Sci. U.S.A.* 106, 5282–5287
23 Edbauer, D. *et al.* (2010) Regulation of synaptic structure and function by FMRP-associated microRNAs miR-125b and miR-132. *Neuron* 65, 373–384
24 Ooi, A.G. *et al.* (2010) MicroRNA-125b expands hematopoietic stem cells and enriches for the lymphoid-balanced and lymphoid-biased subsets. *Proc. Natl. Acad. Sci. U.S.A.* 107, 21505–21510
25 O'Connell, R.M. *et al.* (2010) MicroRNA-155 promotes autoimmune inflammation by enhancing inflammatory T cell development. *Immunity* 33, 607–619
26 Kuhn, D.E. *et al.* (2010) Chromosome 21-derived microRNAs provide an etiological basis for aberrant protein expression in human Down syndrome brains. *J. Biol. Chem.* 285, 1529–1543
27 Du, C. *et al.* (2009) MicroRNA miR-326 regulates TH-17 differentiation and is associated with the pathogenesis of multiple sclerosis. *Nat. Immunol.* 10, 1252–1259
28 Junker, A. *et al.* (2009) MicroRNA profiling of multiple sclerosis lesions identifies modulators of the regulatory protein CD47. *Brain* 132, 3342–3352
29 Rom, S. *et al.* (2010) CCL8/MCP-2 is a target for mir-146a in HIV-1-infected human microglial cells. *FASEB J.* 24, 2292–2300
30 Urdinguio, R.G. *et al.* (2010) Disrupted microRNA expression caused by Mecp2 loss in a mouse model of Rett syndrome. *Epigenetics* 5, 656–663
31 Szulwach, K.E. *et al.* (2010) Cross talk between microRNA and epigenetic regulation in adult neurogenesis. *J. Cell Biol.* 189, 127–141
32 Simon, D.J. *et al.* (2008) The microRNA miR-1 regulates a MEF-2-dependent retrograde signal at neuromuscular junctions. *Cell* 133, 903–915
33 Krol, J. *et al.* (2010) The widespread regulation of microRNA biogenesis, function and decay. *Nat. Rev. Genet.* 11, 597–610

34 Shaked, I. *et al.* (2009) MicroRNA-132 potentiates cholinergic anti-inflammatory signaling by targeting acetylcholinesterase. *Immunity* 31, 965–973
35 Gehrke, S. *et al.* (2010) Pathogenic LRRK2 negatively regulates microRNA-mediated translational repression. *Nature* 466, 637–641
36 Asikainen, S. *et al.* (2010) Global microRNA expression profiling of *Caenorhabditis elegans* Parkinson's disease models. *J. Mol. Neurosci.* 41, 210–218
37 Williams, A.H. *et al.* (2009) MicroRNA-206 delays ALS progression and promotes regeneration of neuromuscular synapses in mice. *Science* 326, 1549–1554
38 Wayman, G.A. *et al.* (2008) An activity-regulated microRNA controls dendritic plasticity by down-regulating p250GAP. *Proc. Natl. Acad. Sci. U.S.A.* 105, 9093–9098
39 Magill, S.T. *et al.* (2010) microRNA-132 regulates dendritic growth and arborization of newborn neurons in the adult hippocampus. *Proc. Natl. Acad. Sci. U.S.A.* 107, 20382–20387
40 Meerson, A. *et al.* (2010) Changes in brain MicroRNAs contribute to cholinergic stress reactions. *J. Mol. Neurosci.* 40, 47–55
41 Cheng, H.Y. *et al.* (2007) microRNA modulation of circadian-clock period and entrainment. *Neuron* 54, 813–829
42 Pauley, K.M. *et al.* (2008) Upregulated miR-146a expression in peripheral blood mononuclear cells from rheumatoid arthritis patients. *Arthritis Res. Ther.* 10, R101
43 Lagos, D. *et al.* (2010) miR-132 regulates antiviral innate immunity through suppression of the p300 transcriptional co-activator. *Nat. Cell Biol.* 12, 513–519
44 Badner, J.A. and Gershon, E.S. (2002) Meta-analysis of whole-genome linkage scans of bipolar disorder and schizophrenia. *Mol. Psychiatry* 7, 405–411
45 Stark, K.L. *et al.* (2008) Altered brain microRNA biogenesis contributes to phenotypic deficits in a 22q11-deletion mouse model. *Nat. Genet.* 40, 751–760
46 Guimaraes-Sternberg, C. *et al.* (2006) MicroRNA modulation of megakaryoblast fate involves cholinergic signaling. *Leuk. Res.* 30, 583–595
47 Benoit, M.E. and Tenner, A.J. (2011) Complement protein C1q-mediated neuroprotection is correlated with regulation of neuronal gene and microRNA expression. *J. Neurosci.* 31, 3459–3469
48 Cogswell, J.P. *et al.* (2008) Identification of miRNA changes in Alzheimer's disease brain and CSF yields putative biomarkers and insights into disease pathways. *J. Alzheimers Dis.* 14, 27–41
49 Ponomarev, E.D. *et al.* (2011) MicroRNA-124 promotes microglia quiescence and suppresses EAE by deactivating macrophages via the C/EBP-alpha-PU.1 pathway. *Nat. Med.* 17, 64–70
50 Pocock, J.M. and Kettenmann, H. (2007) Neurotransmitter receptors on microglia. *Trends Neurosci.* 30, 527–535
51 Kim, A.H. *et al.* (2010) MicroRNA expression profiling in the prefrontal cortex of individuals affected with schizophrenia and bipolar disorders. *Schizophr. Res.* 124, 183–191
52 Im, H-I. *et al.* (2010) MeCP2 controls BDNF expression and cocaine intake through homeostatic interactions with microRNA-212. *Nat. Neurosci.* 13, 1120–1127
53 Lusardi, T.A. *et al.* (2010) Ischemic preconditioning regulates expression of microRNAs and a predicted target, MeCP2, in mouse cortex. *J. Cereb. Blood Flow Metab.* 30, 744–756
54 Younger, S.T. and Corey, D.R. (2011) Transcriptional gene silencing in mammalian cells by miRNA mimics that target gene promoters. *Nucl. Acids Res.* DOI: 10.1093/nar/gkr155
55 Shin, C. *et al.* (2010) Expanding the microRNA targeting code: functional sites with centered pairing. *Mol. Cell* 38, 789–802
56 Hebert, S.S. *et al.* (2008) Loss of microRNA cluster miR-29a/b-1 in sporadic Alzheimer's disease correlates with increased BACE1/beta-secretase expression. *Proc. Natl. Acad. Sci. U.S.A.* 105, 6415–6420
57 Wang, W.X. *et al.* (2008) The expression of microRNA miR-107 decreases early in Alzheimer's disease and may accelerate disease progression through regulation of beta-site amyloid precursor protein-cleaving enzyme 1. *J. Neurosci.* 28, 1213–1223
58 Boissonneault, V. *et al.* (2009) MicroRNA-298 and microRNA-328 regulate expression of mouse beta-amyloid precursor protein-converting enzyme 1. *J. Biol. Chem.* 284, 1971–1981
59 O'Neill, L.A. (2009) Boosting the brain's ability to block inflammation via microRNA-132. *Immunity* 31, 854–855
60 Stefani, G. and Slack, F.J. (2008) Small non-coding RNAs in animal development. *Nat. Rev. Mol. Cell Biol.* 9, 219–230
61 Ransohoff, R.M. and Cardona, A.E. (2010) The myeloid cells of the central nervous system parenchyma. *Nature* 468, 253–262