Novel Protective Effects of Histone Deacetylase Inhibition on Stroke and White Matter Ischemic Injury

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Abstract Understanding how epigenetics influences the process and progress of a stroke could yield new targets and therapeutics for use in the clinic. Experimental evidence suggests that inhibitors of zinc-dependent histone deacetylases can protect neurons, axons, and associated glia from the devastating effects of oxygen and glucose deprivation. While the specific enzymes involved have yet to be clearly identified, there are hints from somewhat selective chemical inhibitors and also from the use of specific small hairpin RNAs to transiently knockdown protein expression. Neuroprotective mechanisms implicated thus far include the upregulation of extracellular glutamate clearance, inhibition of p53-mediated cell death, and maintenance of mitochondrial integrity. The histone deacetylases have distinct cellular and subcellular localizations, and discrete substrates. As a number of chemical inhibitors are already in clinical use for the treatment of cancer, repurposing for the stroke clinic should be expedited.

Keywords Cerebral ischemia · Zn-dependent histone deacetylase · infarct · mitochondria · p53 · aging

Introduction

Stroke, resulting from cerebral ischemia or hemorrhage, is a major public health concern. Worldwide, 15 million people suffer a stroke each year and, of these, nearly 6 million die and another 5 million are left permanently disabled. After

dementia, stroke is the leading cause of disability, which includes loss of vision and/or speech, paralysis, and confusion. Globally, stroke is the second leading cause of death above the age of 60 years, and the fifth leading cause of death in people aged 15–59 years. Fortunately, the relative rate of stroke death in developed countries appears to have fallen by more than a third in the first decade of 2000, and the actual number of stroke deaths has declined by one quarter [1]. However, predictions in the next 2 decades are for a tripling in stroke mortality in developing countries, based on an aging population and increased risk associated with hypertension, obesity, and diabetes.

Finding therapeutics to improve outcome, in particular from ischemic stroke, has proved to be challenging. Treatment relies on thrombolysis, aspirin, and management in a stroke unit. Precisely targeted neuroprotection strategies have failed, despite there being numerous “leads” from preclinical studies. The ideal stroke therapeutic would preserve both gray and white matter (WM), and also encourage the recovery of function. In this article, we discuss how prolongation of brain acetylation via histone deacetylase (HDAC) inhibition reduces stroke injury, preserves neurons from an apoptotic fate, and sustains WM function in the face of experimental ischemic insult. In addition, HDAC inhibitors drive “neuronal” differentiation of multipotent neural progenitor cells [2, 3]. As 2 HDAC inhibitors are already approved by the Food and Drug Administration (Vorinostat and depsipeptide, both for cutaneous T-cell lymphoma) and others are well advanced in clinical trial [4], repurposing for the stroke clinic could be expedited.

Our focus here is on the zinc (Zn)-dependent HDACs, which illustrate but one epigenetic mechanism [5]. The nicotinamide adenine dinucleotide-dependent HDACs (sirtuins) are dealt with in other articles (see Langley and Sauve, and Perez-Pinzon in this issue). Furthermore, the potential roles for long, non-coding RNAs in ischemia are not covered [6–9]. Nor is the burgeoning stroke literature concerning microRNAs [10–13] and the effects of methylation [14].

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Expression of Zn-dependent HDACs in Mouse Brain

The pattern of expression of HDAC 1, 2, and 3 in mouse brain is cell-, region-, and age-specific. HDAC 1 is mainly expressed in the nuclei of astrocytes and axons. HDAC 2 is expressed abundantly in astrocytes, both in nuclear and cytoplasmic compartments, extending to end-feet, while HDAC 3 shows a punctate pattern mostly in astrocyte distal processes; interestingly, a period of ischemia increases expression of these HDACs in glial cell nuclei [15].

Regional changes in HDAC expression pattern in WM may contribute to the predominant injury mechanisms and determine the functional outcome. For instance, HDAC 1 expression in corpus callosum axons contrasts with expression in astrocyte nuclei and end-feet (Fig. 1) in subcortical white matter (SCWM). Similarly, there is no prominent expression of HDAC 2 in corpus callosum, while SCWM astrocytes express HDAC 2 extensively. The expression of HDAC 3 shows the most variation in that it is expressed in glial nuclei and in axons in corpus callosum, while distinctly punctate in distal astrocyte process in SCWM [15].

WM becomes intrinsically more vulnerable to ischemia as a function of age [16]. Among several age-related changes, the class I HDAC expression pattern also changes in older animals. Aging reorganizes the extent of cellular HDAC expression in a region- and cell-specific manner compared with young optic nerves (Fig. 2). Aging axons express all three HDACs, but most prominently HDAC 2, outlining them throughout their entire course of length as opposed to exclusively labeling astrocytes in young optic nerve. Moreover, HDAC 2 becomes evident in glial cell nuclei that exhibit a “beads on a string” pattern, which is characteristic of oligodendrocytes. The age-related change in HDAC 1 expression is similarly noteworthy, such that astrocyte nuclei and proximal processes become immunoreactive for HDAC 1 in addition to axons. Although it has the lowest intensity, HDAC 3 expression preserves a punctate pattern while extending to axons and glial nuclei, rather than astrocyte processes [16].

Overall, class II isoforms HDAC 4 and HDAC 6 show less intense labeling. The expression of HDAC 4 is primarily located in neuronal cytoplasm and in axons in the parietal cortex, while the expression of HDAC 6, though very low, has been shown to be mostly in neuronal cytoplasm [15].

HDAC Inhibition Reduces Ischemic Injury

We systematically reviewed relevant published studies up to 2010 [17]. Our analysis revealed that, following cerebral ischemia, administration of HDAC inhibitors (such as suberoylanilide hydroxamic acid (SAHA), ITF2357, sodium butyrate (SB), valproic acid (VPA)) had a beneficial effect on both infarct volume and functional (behavioral) outcome.

Corpus Callosum

Subcortical White Matter

Fig. 1 Expression pattern of histone deacetylases (HDACs) in white matter. **(a)** HDAC 1 and HDAC 3 are expressed in neurofilament (NF)-200+ axons (arrow heads), while HDAC 2 is absent from corpus callosum. **(b)** HDAC 1 is in the nuclei and, to some extent, in the proximal portions of the main processes and end-feet (arrow heads) of glial fibrillary acidic protein (GFAP) (+) astrocytes outlining capillaries. HDAC 2 is abundant in astrocyte end-feet (arrows). HDAC 3 colocalizes with nuclei (*) and distal processes of GFAP (+) astrocytes, but is absent from end-feet (blue arrows). Modified from [15] and reproduced with permission

Administration of HDAC inhibitors was effective, whether given pre- or post-injury. Relevant studies since then have revealed that VPA attenuates stroke-induced blood–brain barrier disruption [18], trichostatin A (TSA) is effective as a stroke neuroprotectant [19], and hints at the specific contributions of HDACs 4 and 5 to stroke injury [20].

Human brain comprises gray matter and WM in equal volumes, which means that injury sustained after a stroke involves both. In addition, stroke risk increases considerably with age. In addition to vascular factors, aging results in global changes, predominantly in WM, such that the tissue becomes intrinsically more vulnerable. Experimental stroke studies commonly use rodents, which have little WM by

Fig. 2 HDAC expression in optic nerve changes with age. Histone deacetylase (HDAC) 1 is prominent in axons (arrows) and some glial nuclei. HDAC 2 is most prominent in aging mouse optic nerve (MONs) and outlines axons throughout their length (arrows). Asterisk indicates glial nuclei. HDAC 3 is punctate in axons (arrows) and some glial nuclei (*, blue). Reproduced from [29]

volume compared with gray matter. Consequently, the injury in a mouse after an ischemic attack is mainly due to neuronal loss. Therefore, it is plausible that the efficacy of neuroprotective drugs identified in experimental stroke models using young healthy male rodents is a poor predictor of clinical outcome, and this is why experimental “leads” have failed to successfully translate in human trials [21–24]. An ideal stroke therapeutic must preserve neurons, together with their axons and glial cells, to gain optimal functional recovery. Interventions to promote plasticity and repair mechanisms in the post-acute phase must also target both portions of the brain.

Optic nerve, a pure WM tract, is composed of fully myelinated axons and glial cells, thus preserving the three-dimensional architecture. The optic nerve offers the capacity to quantitatively study the structure–function relationship of WM injury mechanisms. In addition, optic nerve is intrinsically sensitive to the process of aging [16, 25–29], and shows progressive molecular and functional changes as a function of age [16, 28, 29]. HDAC inhibitors preserve function in both young and aging axons [28, 29]. Because aging modifies cell and region-specific expression of Class I HDACs [15, 28], these cellular adjustments may contribute to increased vulnerability of aging WM to ischemia.

Consistent with structural observations, pan- and class I HDAC inhibitor (SAHA and MS-275, respectively) administration before or after a period of oxygen glucose deprivation (OGD) promotes functional recovery of axon function (Fig. 3a, b) and preserves WM structure (Figs. 4 and 5). Despite age-related variations in the expression pattern of class I HDACs, pan- and class I-specific HDAC inhibitors promote axon function and preserve axonal mitochondria

Fig. 3 Histone deacetylase inhibitors promote functional recovery of optic nerve. (a) Pre-treatment with SAHA improves axon function in a dose-dependent manner. (b) Pre- or post-treatment with MS-275 promotes recovery of axons. CAP XXX compound action potential, OGD oxygen glucose deprivation. Reproduced from [27]

Fig. 4 Expression pattern of histone deacetylases (HDACs) in mouse optic nerve is altered by ischemia. **(a)** HDAC 1, 2, and 3 are distinctly expressed in glial cells and axons. Arrows point to astrocyte nuclei, arrow heads show glial fibrillary acidic protein (GFAP) (+) astrocyte processes or NF-200 (+) axons, and asterisks indicate nuclei of oligodendrocytes. Scale bar=10 μ m. Reproduced from [27]. **(b)** HDAC expression is upregulated in glial cell nuclei (arrowheads) and astrocyte processes (arrows) after ischemia and SAHA attenuated the effect of ischemia. Blue arrow points to astrocyte end-feet outlining a capillary. OGD oxygen glucose deprivation. Reproduced from [15] with permission

[15, 28], providing proof of principle that HDAC inhibition has non-transcriptional actions in axons and the distant processes of glial cells irrespective of age. Modifications in HDAC expression in WM may imply an age-related adaptive mechanism to maintain proper function. However, during an ischemic episode these adaptive changes may act against the tissue, and may contribute to expedite and aggravate injury process. Whether other WM tracts across age

Fig. 5 Pre-treatment of optic nerve with histone deacetylase inhibitors prevents the loss of glutamate transporter, GLT-1, expression associated with ischemia. Scale bar=1 μ m. GFAP glial fibrillary acidic protein, OGD oxygen glucose deprivation, SAHA XXX. Reproduced from [27]

groups benefit from HDAC inhibition after an ischemic attack remains to be explored.

Because an episode of ischemia modifies HDAC expression in optic nerve [15], it is reasonable to propose that the region-specific HDAC expression patterns in different WM tracts may contribute to regional vulnerability and determine the mechanisms of injury. Consequently, region-specific HDAC inhibition and development of specific HDAC inhibitors may potentiate protective interventions after ischemia with fewer side effects in WM.

Mechanisms of Protection

Potential explanations for the beneficial effects of HDAC inhibitors in the brain are multi-fold. *Ex vivo* optic nerve and *in vitro* neuronal studies suggest that HDAC inhibitors influence a diverse array of targets to maintain neuronal function and survival in response to injury.

Hints from the Optic Nerve

Ischemic WM injury is a result of converging ionic, excitotoxic, and oxidative stress pathways. Glutamate release due

to the reversal of sodium (Na^+)-dependent transport dictates the irrecoverable nature of the injury [29, 30]. The dominant glutamate transporter, GLT1, is located principally on astrocytes and plays a crucial role in maintaining glutamate homeostasis by removing it from the extracellular space [31]. An episode of OGD depletes the expression of this transporter in optic nerve [30], despite the activation of astrocytes. Administration of HDAC inhibitors robustly upregulates the expression of this pump by ~2-fold above control levels, while attenuating astrocyte activation (Fig. 5) [28]. Interestingly, a 2-fold increase in GLT1 expression and protein levels is correlated with early and robust release of glutamate in aging WM [16], while a similar outcome after HDAC inhibition results in delayed and less glutamate release in optic nerve [28].

The explanation for this perplexing issue lies in the principles of GLT1 function (Fig. 6). The transporter takes up glutamate from the extracellular environment, together with Na^+ ion, and stores glutamate inside astrocytes. During energy deprivation, when intracellular Na^+ accumulation reaches a critical level (ionic pathway), GLT1 reverses and releases glutamate to the extracellular space, initiating excitotoxicity.

The spatiotemporal course of glutamate release from WM reflects the sequential nature of these converging pathways. Glutamate levels are well maintained in the first 25–30 min after the onset of OGD, but after 30 min a steady accumulation of glutamate begins [29, 30]. Therefore, the number of transporters reflects the potential to move glutamate, but it is the direction of transport that determines whether glutamate is stored or released. This inevitably focuses attention on the Na^+ /potassium adenosine triphosphatase pump, which consumes ~50 % of adenosine triphosphate (ATP) in the brain and is therefore highly dependent on the availability of ATP to expel Na^+ . The HDAC inhibitors preserve mitochondria and ATP levels in axons in the face of OGD (Fig. 7); axon function persists during OGD followed by a prominent sustained recovery [28].

Consistent with this, HDAC inhibition prominently delays and attenuates glutamate release (Fig. 6), suggesting that preserved ATP levels keep the GLT1 pump in the forward direction. In support of the proposal that HDAC inhibition must have at least 2 distinct sites of action during the sequential course of ischemic injury in WM, application of HDAC inhibitors after the end of OGD promotes axon function recovery

Fig. 6 Histone deacetylase (HDAC) inhibitors preserve ischemic white matter by conserving mitochondria and reducing excitotoxicity. The principle sodium (Na^+)-dependent glutamate transporter, GLT-1, typically takes up glutamate with the co-transport of Na^+ . With the increased intracellular Na^+ following adenosine triphosphate (ATP) depletion, the transporter reverses and releases glutamate. Therefore, the number of transporters determines the capacity of the system, but it is ATP that determines direction of the transporter (to remove or release glutamate). Consistent with this, glutamate after a stable baseline (red arrows), accumulates during oxygen glucose deprivation (OGD) (black). Note that the glutamate levels return to baseline after ischemia. HDAC inhibitors upregulate GLT-1 and preserve ATP therefore attenuating glutamate release (red). Modified from [29]

Fig. 7 Pre- or post-treatment with MS-275 preserves cyan fluorescent protein (CFP) (+) axonal mitochondria. *OGD* oxygen glucose deprivation, *CAP XXX*, *ATP* adenosine triphosphate. Reproduced from the supplement of [27]

and preserves mitochondria. Pre-OGD, HDAC inhibition acts on the excitotoxic pathway, interrupting subsequent mitochondrial dysfunction and ATP depletion. Post-OGD, HDAC inhibition acts on the oxidative pathway and perhaps even on the downstream components of excitotoxicity [28].

Transcriptional Effects

Inhibitors of HDACs appear to have a propensity for modulating expression of important neurotrophins. Brain-derived neurotrophic factor (BDNF) supports the survival of select differentiated neurons [32, 33] and encourages the growth and differentiation of new neurons and synapses. Addition of VPA to cultured rat cortical neurons induces BDNF messenger RNA [34] and induces vascular endothelial growth factor, angiogenesis, and neurogenesis *in vivo* [35, 36]. In a screen of small molecules structurally related to known HDAC inhibitors a compound referred to as crebinostat, for its ability to concomitantly activate cyclic adenosine monophosphate response element binding-mediated transcription, was identified

[37]. Crebinostat expressed potent inhibition of the deacetylase activity of recombinant class I HDACs 1, 2, 3, and class IIb HDAC6. Addition of crebinostat to mouse embryonic day 17 forebrain neurons increased acetylation of histones H3 and H4.

Addition of SAHA enhanced dopaminergic neuron survival in mixed neuron–glial cultures [38]. Conditioned media derived from astrocyte-enriched cultures treated with SAHA significantly increased the survival of dopaminergic neurons, indicating that inhibitors can target astrocytes to elicit the secretion of soluble neurotrophic factors. Subsequent analysis demonstrated that SAHA increased the expression and release of glial cell-derived neurotrophic factor. Such studies demonstrate that HDAC inhibitors may have multiple targets [38]. Astrocytes were previously shown to be a target in the mouse optic nerve (above), in the upregulation of a transporter reducing extracellular glutamate accumulation [28].

HDAC inhibitors appear to modulate neuronal survival not only by altering gene transcription in general, but by

modifying the activity of other transcription factors. The nuclear erythroid 2-related factor 2 (Nrf2), may represent one critical example of a survival-promoting transcription factor. It appears that Nrf2 signaling promotes multiple pathways of cellular protection by coordinating the transcription of genes that confer protection against oxidative stress and inflammation [39]. Activation of the Nrf2 pathway has been shown to increase the resistance of neurons to oxidative stress and metabolic insults in culture, and to ischemic stroke *in vivo* [40]. More recently, a broad-based HDAC inhibitor (TSA) was shown to increase neuron viability after OGD [19]. *In vitro* analysis demonstrated that the HDAC inhibitor activated Nrf2, in part, by reducing expression of the suppressor Keap1, promoting Keap1/Nrf2 dissociation, and enhancing Nrf2 nuclear translocation [19]. Consistent with these *in vitro* findings, the addition of TSA to Nrf2-deficient mice did not confer protection from ischemia [19].

HDAC inhibitors may also promote neuronal survival by blocking the action of transcription factors that regulate apoptosis. The tumor suppressor, p53, has been implicated as a key regulator of apoptosis in acute and chronic neurologic insults [41]. In contrast to non-neuronal cells, the apoptotic activity of p53 in postnatal cortical neurons does not rely on direct action at the cytosol/mitochondria, but is exclusively mediated through transcription-dependent functions [42]. Interestingly, despite promoting nuclear p53 accumulation, inhibitors of class I/II HDACs protected cultured postnatal cortical neurons from p53-dependent cell death. They suppressed p53-dependent p53 upregulated modulator of apoptosis (PUMA) expression, a critical signaling intermediate linking p53 to Bax activation, thus preventing post-mitochondrial events, including cleavage of caspase-9 and caspase-3. Moreover, inhibitors also prevented caspase-3 cleavage in postnatal cortical neurons treated with staurosporine, 3-nitropropionic acid, and a B-cell lymphoma-2 inhibitor, all of which require the presence of Bax but not p53 to promote apoptosis. Although these 3 toxic agents displayed a requirement for Bax, they did not promote PUMA induction. These results demonstrate that HDAC inhibitors block Bax-dependent cell death by two distinct mechanisms to prevent neuronal apoptosis and, for the first time, define a molecular target [42].

It was shown that pulsed exposure of cultured embryonic cortical neurons to HDAC inhibitors blocked cell death induced by oxidative stress [43], and that protection was associated with transcriptional activation of the cell cycle inhibitor, p21. Overexpression of p21 in neurons mimicked the protective effect of HDAC inhibitors against oxidative stress-induced toxicity, including death induced by glutathione depletion or peroxide addition. However, despite p21 induction being sufficient to promote neuronal survival in embryonic neurons subjected to oxidative stress, it was not necessary, as HDAC inhibitors conferred protection on neurons from p21-deficient mice [43].

Mitochondrial Integrity

Abnormalities in mitochondrial transport and function are very deleterious to neuronal function and survival. Inhibition of HDAC 6 results in higher levels of acetylated tubulin, thereby enhancing the binding of the motor protein kinesin-1 to tubulin, which promotes intracellular transport along microtubules. Addition of an HDAC 6-specific inhibitor to cultured hippocampal neurons significantly enhanced the movement of mitochondria [44]. Interestingly, addition of the broad-based HDAC inhibitor TSA also enhanced the number of mitochondria that moved, as well as their average velocity. Changes in mitochondrial morphology were not detected. These findings are consistent with an earlier report [45], which demonstrated that TSA increased the vesicular transport of BDNF by inhibiting HDAC 6 and elevating the acetylation of tubulin [ref].

A role for HDAC 1 in mitochondrial transport has also been demonstrated. While HDAC 1 is normally a nuclear enzyme involved in transcriptional repression, cytosolic HDAC 1 was detected in damaged axons and cultured neurons exposed to glutamate and tumor necrosis factor- α (TNF- α) [46]. Knockdown of HDAC 1, but not HDACs 2, 3, 4, 6, or 8, abolished neuritic damage induced by glutamate or TNF- α . Subsequently, it was shown that HDAC 1 binds to members of the kinesin family of motor proteins, obstructing the interaction between these proteins and transported cargo such as mitochondria. Knockdown of HDAC 1, inhibition of its transport out of the nucleus, or addition of the class I inhibitor, MS-275, significantly improved mitochondrial transport in neurons damaged by glutamate or TNF- α . In the same study, addition of an HDAC 6-specific inhibitor failed to protect neurons or improve mitochondrial transport in response to damage.

While *in vitro* studies clearly demonstrate that HDAC isoforms can negatively affect mitochondrial transport, their influence on mitochondrial biology is broader than this. Morphological changes in mitochondria, consistent with increased fission (fragmentation) and loss of ATP, are induced in the mouse optic nerve during ischemic injury (see above). The addition of pan- and class I-specific inhibitors, before or after OGD, promotes functional recovery of axons and preserves WM cellular architecture, including preservation of mitochondria and maintenance of ATP (Fig. 7) [27, 28].

Oxidative stress has recently been shown to cause changes in the expression of proteins that regulate fission and fusion, including dynamin-related protein-1 (Drp1) and parkin. The addition of the DNA damaging agent camptothecin significantly reduced the levels of both prior to the activation of cell death [47]. Addition of SAHA, previously shown to protect cultured postnatal cortical neurons from camptothecin-induced cell death [48], can prevent the DNA damage-induced decline in Drp1 and parkin expression (unpublished

observations). This might relate to the neuroprotective action of HDAC inhibitors against DNA damage-induced cell death, as the elevated expression of parkin or Drp1 was able to confer significant protection against camptothecin-induced cell death and prevent the changes in mitochondrial morphology [47].

Chaperone Function

Abnormalities in folding, and the elimination of improperly folded and damaged proteins, are associated with neuronal dysfunction and loss of viability. Heat-shock protein 70 (HSP70) is a molecular chaperone, and is neuroprotective and anti-inflammatory. Using rat cortical neurons in culture, VPA was found to significantly upregulate the expression of HSP70 [49]. This finding was subsequently validated using multiple pan- and class I-specific HDAC inhibitors. Importantly, VPA-mediated neuroprotection against glutamate-induced cell death was lost if HSP70 induction was blocked. In a subsequent study, structural derivatives of VPA were found to be more potent inducers of HSP70 and more effective at promoting neuroprotection against glutamate toxicity [50].

Identifying the HDACs Involved

A host of *in vitro* studies has shown that not all HDACs are created equal when it comes to their involvement in stress-induced cell death, and HDAC 4 presents an excellent example. As seen with HDAC 1, the inappropriate translocation of HDAC 4 from the cytosol to the nucleus may initiate neuronal cell death. Shifting cerebellar granule neurons from high to low potassium or glutamate exposure induced the rapid translocation of HDAC4 from the cytosol to the nucleus [51]. Suppressing HDAC 4 expression and activity reduced neuronal cell death. Hydrogen peroxide (H₂O₂) treatment was also shown to induce the inappropriate translocation of HDAC 4 into the nucleus of embryonic cortical neurons in culture [52]. HDAC 4 will bind and inactivate the transcriptional activity of the peroxisome proliferator-activated receptor γ (PPAR γ), which serves an essential protective function in neurons. Knockdown of HDAC 4 abolished the oxidative stress-induced repression of PPAR γ activity in neurons and conferred protection against H₂O₂-induced cell death. Moreover, PPAR γ overexpression was sufficient to protect cortical neurons from HDAC4 overexpression or H₂O₂-induced cell death providing some insight into the mechanism by which nuclear HDAC4 may alter neuronal sensitivity to stress.

“Neuronogenesis” after Ischemic Injury

Following an injury to the brain, such as a stroke, the rate of production of new neurons is amplified [53]. Furthermore, transplantation of exogenous stem/progenitor cells into a

damaged brain is touted as having potential for future stroke therapy. Whether new neurons, from either endogenous or exogenous sources, survive long enough to contribute to recovery of function remains unclear. However, it is very clear that epigenetic re-programming of cells occurs at different stages of life, and that HDAC inhibitors can drive both endogenous and isolated embryonic and adult neural progenitor cells to differentiate into neurons [54–57].

Class I HDAC knockout mice are embryonic lethal. While deletion of either HDAC 1 or 2 evokes no overt phenotype, deletion of both causes defects in multiple aspects of neuronal development [58]. Humphrey et al. [59] expressed dominant-negative HDAC mutants in embryonic cortical neural stem cell lines to reveal that HDAC 1–3 activities inhibit differentiation to oligodendrocytes and astrocytes, and HDAC 1 activity is required for neuronal differentiation [60]. This might suggest that the adult cellular context is quite distinct. It is clear that HDAC 2 deficiency in mice [61], and the nitrosylation and subsequent displacement of HDAC 2 [62], correlate with the effects of chronic HDAC inhibitor treatment, which include an increase in synapse number and also memory facilitation.

Summary

In summary, accumulating evidence predicts long-term benefits of (specific) HDAC inhibition following acute central nervous system injury. Distinct effects of HDAC inhibitors may be attributed to their specific pattern of distribution among neurons and glial cells, and their ability to employ both transcriptional and non-transcriptional actions in axons and glial cell processes. Current drugs broadly inhibit class I and class II HDACs, and are generally not specific with regard to the proteins they affect. Identifying those specific HDACs whose inhibition results in neuroprotection and neurorestoration will point to more desirable therapeutic targets.

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