Epigenetic dysregulation in schizophrenia: Molecular and clinical aspects of histone deacetylase inhibitors

ARTICLE *in* EUROPEAN ARCHIVES OF PSYCHIATRY AND CLINICAL NEUROSCIENCE · FEBRUARY 2013

Impact Factor: 3.53 · DOI: 10.1007/s00406-013-0395-2 · Source: PubMed

Eur Arch Psychiatry Clin Neurosci (2013) 263:273–284
DOI 10.1007/s00406-013-0395-2

INVITED REVIEW

Epigenetic dysregulation in schizophrenia: molecular and clinical aspects of histone deacetylase inhibitors

Alkomiet Hasan · Amanda Mitchell · Anja Schneider · Tobias Halene · Schahram Akbarian

Received: 15 December 2012 / Accepted: 18 January 2013 / Published online: 5 February 2013

Abstract Notwithstanding the considerable advances in the treatment options for schizophrenia, the cognitive symptoms in particular are not receptive to antipsychotic treatment and considered one of the main predictors for poor social and functional outcome of the disease. Recent findings in preclinical model systems indicate that epigenetic modulation might emerge as a promising target for the treatment of cognitive disorders. The aim of this review is to introduce some of the principles of chromatin biology to the reader and to discuss a possible role in the neurobiology and pathophysiology of schizophrenia. We will discuss potential epigenetic targets for drug therapy, including histone deacetylase inhibitors (HDACi). In a second part, conceptual and practical challenges associated with clinical trials of chromatin-modifying drugs in psychiatric patient populations are discussed, including safety profiles, the potential for adverse effects and general issues revolving around pharmacokinetics and pharmacodynamics. Additional investigations are required in order to fully evaluate the potential of HDACi and similar "epigenetic therapies" as novel treatment options for schizophrenia and other psychotic disease.

Keywords Schizophrenia · Epigenetics · Histone deacetylase inhibitor · Chromatin biology · Cognitive deficits

A. Hasan (✉)
Deparment of Psychiatry and Psychotherapy, Ludwig-Maximilians-University, Nussbaumstr. 7, 80336 Munich, Germany
e-mail: ahasan@gwdg.de; Alkomiet.Hasan@med.uni-muenchen.de

A. Mitchell · T. Halene · S. Akbarian
Divsion of Psychiatric Epigenomics, Mount Sinai School of Medicine, New York, NY, USA

A. Schneider
German Center for Neurodegenerative Diseases (DZNE), Göttingen, Germany

A. Schneider
Department of Psychiatry and Psychotherapy, University Medical Center, Göttingen, Germany

Introduction

Despite more than a century of research, the treatment of schizophrenia is still one of the greatest challenges in clinical psychiatry. Antipsychotics have revolutionized the treatment of schizophrenia, but until today, the majority of schizophrenia patients still suffer from a poor outcome and incomplete response to treatment [7, 130]. The recommendations from recently published treatment guidelines [20, 56] clearly emphasize that positive symptoms and symptoms of disorganization can adequately be treated in the majority of cases with currently approved antipsychotics. Unfortunately, the remaining negative and cognitive symptoms are responsible for much of schizophrenia's debilitating effects and do not respond to pharmacological treatment [56, 69, 102]. Therefore, there is still a desperate search for new treatment options in schizophrenia beyond the established targets of the available antipsychotics.

One major problem of drug development for schizophrenia and other psychotic disorders (e.g. bipolar mania, major depression with psychotic features, schizoaffective disorder) is that the underlying neurobiology and the pathobiology mechanisms remain poorly understood. Each one of the extensively investigated parameters in schizophrenia patients that has been shown to be deficient (e.g. volume and cell loss, structural dysconnectivity, impaired

plasticity, neurochemical alterations, impaired functional activation, altered sensory gating, and many others [27, 107, 117]) only represent one small piece of a big and confusing jigsaw puzzle because none of it stands for a unifying neuropathology and none of it can be used to diagnose schizophrenia. Therefore, there is a need for preclinical work to investigate, on an empirical basis, novel and hitherto unexplored therapeutic avenues. Epigenetic regulation might be one such promising target to understand the pathophysiology of schizophrenia and to develop treatment alternatives. Preclinical studies in animal models point to several promising, pharmacological [105], and non-pharmacological [84] treatment modalities that should be further explored in future research.

This review will discuss the potential of epigenetic drug targets for the treatment of schizophrenia. After introducing the basic concepts of chromatin regulation and epigenetics, we summarize the current knowledge as it pertains to epigenetic alterations in the brain of patients diagnosed with schizophrenia. We then discuss recent findings in animal models, highlight neurobehavioral effects of chromatin-modifying drugs, including HDACi. As some of these findings could be highly relevant for the treatment of schizophrenia, we predict that "epigenetic therapies" will remain a promising area of basic and translational neuroscience research. That said, multiple issues (e.g. target selectivity, adverse effects, and identification of eligible patients) need to be clarified before clinical trials with epigenetic drug targets could be considered for the treatment of schizophrenia.

A primer on post-translational histone modifications, including acetylation

The elementary unit of chromatin is the nucleosome—146 base pairs of DNA wrapped around an octamer of core histones H2A/H2B/H3/H4, connected by linker DNA and linker histones. DNA and histone modifications and variant histones provide the major building blocks for the "epi (greek for *over, above*)-genome" which defines the functional architecture of the genome, including its organization into many tens of thousands of transcriptional units, clusters of condensed chromatin, and other features that are differentially regulated in different cell types and developmental stages of the organism [88, 115]. An in-depth description of all epigenetic markings would be far beyond this review chapter, but multiple recent excellent reviews on this topic provide a good starting point for the reader interested to learn more about this [35, 76, 139].

Epigenetics is defined as a long-lasting change in phenotype and gene expression that is not due to changes in DNA sequence [28]. Changes in gene expression can be mediated by either DNA or histone modifications which modify gene transcription or by non-coding RNAs which interfere on the translational level. DNA modifications include cytosine methylation resulting in decreased transcriptional activity [28]. Interestingly, based on studies in human sperm, up to 4 % of genome is believed to maintain some degree of cytosine methylation and nucleosomal organization (including retainment of some of the histone modifications), potentially passing on heritable epigenetic information to offspring [54, 114].

The epigenetic regulation of chromatin by virtue of chemical histone modifications is complex, with more than 100 amino residue-specific post-translational modifications (PTMs) [126], including lysine mono-, di-, and trimethylation, acetylation, and crotonylation, polyADP-ribosylation, and small protein modification (ubiquitin, small ubiquitin-like modifier), as well as arginine (R) methylation and citrullination, serine (S) phosphorylation, and tyrosine (T) hydroxylation[79, 126, 129]. Different combinations of these site- and residue-specific PTMs define regulatory sequences such as promoters and enhancer, gene bodies, condensed heterochromatin, and so forth [16, 139]. Many active promoters, for example, are defined by sharp enrichment for a subset of histone methylation markings, including H3 trimethylated at lysine 4, and high levels of overall histone lysine acetylation [139]. In general, histone acetylation is associated with a more flexible and "open" chromatin state, thereby facilitating gene expression. One example of this is that it enables enhancer and other regulatory sequences separated from a gene target site by thousands of kilobases, or even megabases, to engage with distant promoters in chromosomal loop formations [90], and promoting general nucleosomal mobility and disassembly [121], a prerequisite for a multitude of functions include DNA repair and gene expression.

Histone acetylation is regulated by the opposing effects of histone acetyltransferases (HATs) and deacetylases (HDACs). Among the highly diverse group of HATs are well-known transcriptional regulators with a key role for neuronal plasticity, including the circadian pacemaker gene *CLOCK* (Circadian Locomotor Output Cycles Kaput) encoding a basic helix-loop-helix (PAS) transcription factor [29] and the Ca^{2+}/cAMP response element-binding protein (CREB)-binding protein CBP/CREBBP [5, 77, 133, 136].

In humans, the activity of HATs is counterbalanced by at least 18 different HDACs, which are commonly divided into 4 classes, based on their equivalents in yeast [30]. Class I includes HDAC1/2/3/8, class IIa encompasses HDAC 4/5/7/9, class IIb HDAC 6/10, and HDAC11 is the sole representative of class IV [30]. All these HDACs are defined by a zinc ion site in the catalytic-binding pocket which also explains why many classical HDAC inhibitors (HDACi) [30] are unspecific and act on multiple HDACs. The class III HDACs include sirtuins 1-7 are structurally

different and require nicotinamide dinucleotide (NAD+) as a co-factor [30]. Furthermore, it is likely that all HDACs target various nuclear and cytoplasmic non-histone proteins for deacetylation [30]. One example would be SIRT1, a class III HDAC. SIRT1 deacetylates the transcription factor NHLH2 which results in upregulated expression of monoamine oxidase A (MAO-A), and could explain lower brain serotonin levels and altered psychomotor behavior in SIRT1 mutant mice [91].

Histone acetylation in schizophrenia and related disease

Schizophrenia is a complex psychiatric disorder. As mentioned above, both neuropathology and genetic underpinnings remain poorly defined while post-mortem studies indicate that many of the affected cases exhibit widespread dysregulation in gene expression in the cerebral cortex and other brain regions. For example, altered expression of oligodendrocyte proteins and messenger RNAs (mRNAs) has been shown for prefrontal and temporal cortex of schizophrenia subjects [8, 52, 73, 94, 113, 131]. Furthermore, ligand-gated ion receptors, reuptake transporters, and metabolic enzymes for inhibitory or excitatory neurotransmission pathways show altered expression in cerebral cortex in subjects with schizophrenia [3, 13, 15, 22, 32, 33, 51, 58, 60, 98, 135]. Whether these transcriptional changes are directly related to the underlying etiology(ies) or events further downstream in the pathophysiology of disease is unclear.

Supporting the hypothesis that histone acetylation is affected in schizophrenia, a microarray analysis of a post-mortem brain collection of 19 subjects with schizophrenia compared with 25 controls revealed significantly increased expression of the class I histone deacetylase, in prefrontal cortex (on average 30–50 %) [118]. The authors hypothesize that similar changes may affect a subset of patients diagnosed with bipolar disorder [118]. Increased HDAC1 expression has also been reported for the neuronal layers of the hippocampus and medial temporal lobe in an independent cohort of schizophrenia subjects [14]. Therefore, abnormally high levels of HDAC1 transcripts in corticolimbic circuitry appear to be a molecular pathology in a significant number of patients on the mood and psychosis spectrum. Notably, HDAC1 was also upregulated in neurons in response to hypoxia [134], and perinatal hypoxia is discussed as one factor contributing to disease pathogenesis [127]. In addition, acute overexpression of HDAC1 in the adult mouse hippocampus resulted in decreased prepulse inhibition, a phenomenon which is described in schizophrenia patients and animal models [10].

Consistent with these observations, loss of promoter-associated histone acetylation in prefrontal cortex of subjects with schizophrenia has been reported for glutamic acid decarboxylase 1 (GAD1), 5-hydroxytryptamine receptor 2C (HTR2C), and other genes with a key role in the pathophysiology of schizophrenia [128]. Circumstantial evidence for a possible role of HDACs in the pathogenesis of schizophrenia was recently provided by Kurita et al. (2012) who reported that inhibition of HDAC2 might increase the antipsychotic activity of second-generation antipsychotic drugs. Metabotropic glutamate receptors mGlu2 and 3 agonists are studied in clinical trials for potential beneficial effects on psychotic symptoms and are down-regulated in response to increased HDAC2 activity. Furthermore, HSV-mediated overexpression of HDAC2 in a mouse model resulted in a schizophrenia-like behavior, including diminished prepulse inhibition [82]. However, the finding that chronic treatment with clozapine induces HDAC2 would suggest that some of the changes in HDAC levels in post-mortem brain should be interpreted with caution and could be due to drug effects rather than represent underlying disease pathology. Epigenetic dysregulation in schizophrenia brain is likely to go beyond dysregulated HDAC1 and HDAC 2 expression, because changes in methylation of DNA cytosines and of histone lysine and arginine methylation were reported for a select set of gene promoters important for neurotransmission, myelination, metabolism, and various other cellular functions [1, 4, 45, 66, 67, 70, 72, 101, 119].

Given the aforementioned epigenetic changes in post-mortem brain tissue subjects, chromatin-modifying drugs are expected to elicit robust changes in cognition and behavior in animal models related to psychiatric conditions. So far, this hypothesis appears to be correct. For example, virus-mediated expression of HDAC1 (which is upregulated in post-mortem brain tissue of schizophrenia patients) in hippocampal neurons facilitates fear extinction in mice. Fear extinction can be considered as a certain kind of emotional memory which has been shown to be affected in schizophrenia patients [10, 63]. Interestingly, opposite effects were observed when the HDAC activity was inhibited by treatment with MS-275, an HDACi drug with some specificity for HDAC1 [65, 75], or by decreasing HDAC1 expression altogether [10]. However, other HDACi drugs that broadly target multiple class I/II HDACs appear to enhance the consolidation of fear memories when administered into rat amygdala [104] or facilitate extinction when injected into mouse hippocampus [83]. These few examples from the fear and anxiety literature illustrate the emerging complexity of epigenetic mechanisms in psychiatry: results will not only depend on the targeted HDAC subtype but may vary with the brain region where the drug was delivered [80]. Other potential treatment options for schizophrenia, such as phosphodiesterase inhibitors face a similar challenge of both subtype specificity and (brain) region-specific drug delivery [53].

There is also an increasing amount of literature exploring epigenetic effects of valproate treatment in schizophrenia. VPA and related compounds including butyrate compounds and other short-chain fatty acids inhibit multiple members of class I HDACs [41, 50]. However, an augmentative effect of VPA when used in combination with atypical antipsychotic drugs could not be established in clinical trials [56]. This might be due to the fact that it is still unclear whether therapeutic doses of VPA affect brain histone acetylation.

For example, in acute mania, one possible initial therapeutic dosing strategy, that is, often applied in the United States is 750 mg per day in divided doses. This is increased every two to three days as tolerated to a maintenance dosage of 1,000–3,000 mg per day, or 30–60 mg/kg [48] to achieve a therapeutic blood level of 50–125 μg/mL, with blood levels above 94 μg/mL shown to result in the best response [6]. While some have argued that these therapeutic doses are comparable to VPA concentrations used in cell culture systems to induce histone hyperacetylation [111] (for example, in the neural SK-N-SH cell line, H3 acetylation is increased in a concentration-dependent manner over a range of VPA concentrations (0–2 mM) [106]), much higher systemic doses are used to induce histone H3 hyperacetylation in the rodent brain. A 200 mg/kg injection has been shown to increase histone H3 acetylation by 50 % in the mouse hippocampus 2 h later [138]. Similarly, a 300 mg/kg dose (2 mmol/kg) of valproic acid in mice induces both fourfold increases in H3 histone acetylation 2 h after injection and reverses the down-regulation of reelin and GAD67 after L-methionine treatment in the brain after 15 days of treatment [132]. However, translation of drug dosage from mouse to humans based on surface area (Baur's mouse dose) results in an equivalent doses of 545 mg/kg body weight in humans which is much higher than the VPA doses in a clinical setting [12, 112].

There is increasing evidence that antipsychotic drugs elicit robust effects on post-translational histone modifications (including acetylation) in fronto-striatal circuitry: Acute blockade of striatal dopamine D_2 receptors by haloperidol results in a large, global increase in a dual phospho-acetylation mark [89], and exposure to the second-generation antipsychotic clozapine, which is still the mainstay of treatment in patients with an insufficient response to other antipsychotics [20, 56], elicits histone and DNA methylation changes at a subset of GABAergic gene promoters in the frontal cortex[31, 67]. A recent report indicates that second-generation antipsychotics, including clozapine, induce a compensatory upregulation of HDAC2 expression in frontal cortex via a 5-$HT2_A$ serotonin receptor-dependent mechanism. This is detrimental for cognitive function by repressing metabotropic glutamate receptor *Grm2* expression [82]. Interestingly, the same study reported that systemic administration of a broad-acting HDACi, suberoylanilide hydroxamic acid (SAHA, vorinostat/Zolinza®), greatly improved clozapine's therapeutic efficiency, as measured by a decrease in head-twitching response after exposure to a hallucinogenic drug. Moreover, SAHA potentiated the antipsychotic response to novel antipsychotic compounds such as LY379268, a metabotropic glutamate receptor agonist [82]. This preclinical work, if confirmed, could indeed open up promising avenues for radically novel, epigenetics-based pharmacotherapies for schizophrenia and related disease. However, it should be kept in mind that second-generation antipsychotics are not superior to first-generation antipsychotics with regard to reducing psychotic symptoms and delusions [56]. Therefore, molecular mechanisms of antipsychotic drug action in the neural system of schizophrenia patients are certainly not limited to the above-described epigenetic processes [82].

The clinical perspective

As outlined in the first part of this review article, modulations of histone acetylation and other epigenetic regulatory mechanisms are potential targets for therapeutic applications in psychiatric disorders. One potential molecular pathway to modulate histone acetylation is the inhibition of histone deacetylase activities leading to a subsequent change in the chromatin conformation and activation of gene transcription [74, 78]. Various agents from different chemical classes are available for human use and investigated in Phase I to Phase III trials. To date the only currently approved clinical indication for such modern HDACi is cancer therapy. Consequently, clinical research in oncology is attracting the greatest attention in this field, but this important issue is reviewed elsewhere [2, 78, 95]. Recent preclinical findings and hypothetical consideration from molecular biology indicate that HDACi are not only effective drugs to fight cancer, but that these drugs provide potential treatment options for severe psychiatric disorders [34, 46, 71, 74]. The following sections will focus on the potential and promising effect of HDACi for the treatment of schizophrenia. The reader should be aware that apart from studies with the low potency and pan-selective HDACi valproate (see above and below) no HDACi have been studied to treat psychiatric disorders in humans yet. Therefore, our aim is not just to summarize for a potential therapeutic effect, but to point out risks and problem areas of this new research field.

HDACi for the treatment of cognitive symptoms in psychosis

Schizophrenia shows a complex symptomatology, including positive, negative, and cognitive symptoms. These

cognitive impairments are very frequent and can be considered to be core features in schizophrenia patients [56, 59, 69, 100, 102, 108]. Different studies reveal that cognitive symptoms are primary symptoms. They appear to be independent from other symptoms domains, they are already present before positive symptoms, they are detectable in unaffected first-degree relatives, and occur even before the onset of frank psychosis [17, 40, 64, 100, 103, 124]. It is noteworthy that these impairments in cognition are one important predictor for poor social and functional outcome and a "major contributor to disability" in schizophrenia patients [39, 47, 62, 120]. However, despite the great impact of cognitive symptoms and a magnitude of different treatment approaches [56], there is still no established pharmacological treatment. Additionally, for some of the most severely affected patients, it is difficult to participate in sophisticated therapeutic approaches, like cognitive behavioral therapy or cognitive remediation, a fact that highlights the need for alternative treatment options.

Recent research indicates that the modulation of neuronal histone acetylation is involved in plasticity mechanisms and in memory consolidation (see above), a finding that could be broadly relevant for a range of cognitive disorders (both in the absence or presence of chronic neurodegeneration). For example, alterations in histone acetylation may be one of the reasons of memory deficits observed in Alzheimer's disease [10, 18, 71, 110]. Several studies in amyloid precursor protein (APP) mouse models showed that application of the HDACi inhibitor SAHA can reverse cognitive deficits and induce plasticity (sprouting of dendrites, increased number of synapses) indicating that HDACi might be suitable therapeutic options for neurodegenerative disorders [11, 37, 49, 110, 116]. Even more related to the focus of this review, it was recently reported that hippocampal overexpression of HDAC1 modulates fear extinction learning [10] which is also affected in schizophrenia patients [10, 49, 63]. However, in some models, manipulation of HDAC1 does not have a severe impact on memory consolidation [49] and other targets, such as HDAC2, 3, and 6 seem to be more relevant for memory formation. For example, HDAC2 levels are elevated in AD patients [44] and overexpression of HDAC2 in the mouse brain impaired memory formation while deletion of HDAC2 from neurons improved memory formation [49]. Likewise, deletion of either HDAC2, HDAC3, or HDAC6 improved memory formation in AD mouse models [43, 96, 97].

Taken together, there is emerging evidence that HDAC inhibition has a beneficial effect on learning and cognition in neurodegenerative dementia [37, 46]. Perhaps, these therapeutic effects may apply to non-neurodegenerative cognitive disease as well. One could speculate that the treatment of schizophrenia patients with HDACi might have the potential to reverse cognitive symptoms. Currently, the treatment of cognitive symptoms in schizophrenia with first-generation and second-generation antipsychotics has only very limited evidence [56], and the desperate search of a treatment options might be accelerated by HDACi or other new drug groups which target the histone complex.

Valproate: a non-selective HDACi

VPA is a well-established neuroactive drug, which is frequently used as anticonvulsant, mood stabilizer, for the treatment of acute mania and as prophylactic drug in different types of headache. However, a beneficial effect of VPA in improving cognition in schizophrenia patients could not be established [56], and a potential unfavorable effect of VPA on cognition is frequently discussed [61]. As discussed above, VPA, in the conventional therapeutic dose range, is unlikely to elicit a significant effect on brain histone acetylation. However, preclinical research over the last years revealed that valproate partly acts as HDACi and decreases methylated sites at reelin and GAD67 (glutamate acid decarboxylase) promoters leading to a consecutive increase in both proteins [42, 50]. A great deal of work has been done to investigate the effects of valproate as add-on therapy in schizophrenia. The combination of valproate with different antipsychotics yielded inconsistent results with regard to improvement of disease severity, hostility, aggression, motor side effects, and as add-on treatment of patients with a poor treatment response with antipsychotics. As a result, recently published evidence-based guidelines for the pharmacological treatment of schizophrenia do not recommend valproate for the add-on treatment of schizophrenia [20, 56].

Clozapine, other atypical antipsychotics, epigenetics, and GABA

Clozapine is probably the most effective available antipsychotic and is sometimes considered as the only "silver bullet" for the treatment of schizophrenia: It is effective in first-episode and chronically ill schizophrenia patients and it is the best established antipsychotic in treatment-resistant schizophrenia [20, 56]. Despite some severe and potential life-threatening side effects, its use is recommended across guidelines [20, 56, 85]. Remarkably, some reports suggest that there is a dopamine-D2/D3-receptor independent epigenetic regulatory effect of clozapine on GABAergic neurons [31, 50, 67]. A recent review [50] reported related effects for high dosages of olanzapine and quetiapine, but

not for risperidone and haloperidol. The authors proposed that changes in chromatin remodeling and activation of DNA-demethylation within GABAergic gene promoters are related to the dibenzodiazepine-structure (clozapine, olanzapine, or quetiapine) [50].

Some clinical trials implicate that olanzapine might have a slight and marginal superiority over other antipsychotics and it is the first choice in severely ill patients are not able to receive clozapine [56, 92]. Quetiapine received good evidence-based recommendation for the treatment of schizophrenia [20, 56], but in clinical practice, one primary field of application is to treat symptoms of bipolar disorders and other mood disorders disease [23, 125]. The aforementioned preclinical studies further showed that the DNA-demethylating actions of a dibenzodiazepine as one epigenetic regulatory process that could be enhanced by the co-application of valproate, whereas the highest effects were observed for clozapine given in combination with valproate [50]. The underlying molecular mechanisms of these phenomena remain to be elucidated.

The reader should be aware that other antipsychotics, like haloperidol, risperidone, aripiprazole, amisulpride, and many others are not necessarily inferior to olanzapine and quetiapine and that they are still effective in a significant amount of patients. Furthermore, while the add-on treatment with valproate seems not to be clinically effective, the link between epigenetics, GABAergic activity, and the treatment with dibenzodiazepines holds promise for the future. As discussed before, the GABAergic deficits in cortical neurons might be the results of epigenetic modulations triggered by a DNA methyltransferase-1-induced promoter hypermethlation [26, 46, 81]. A cortical GABAergic deficit and the association with deficient plasticity are evident in schizophrenia [14, 57, 87] Also, it was recently demonstrated that clozapine directly interacts with the $GABA_B$ receptor [137]. Therefore, there is hope that epigenetic modulators acting in the GABAergic systems might correct the observed epigenetic alterations in schizophrenia patients. The co-administration of valproate and dibenzodiazpines has been employed in preclinical models but has yet not been successful in clinical practice.

Brain permeability, target selectivity, dose ranges, and treatment duration

Any HDACi, in order to be seriously considered for the treatment of schizophrenia and other neuropsychiatric disease, must penetrate the blood–brain barrier to reach sufficient drug concentrations in the cerebrospinal fluid and the brain tissue. The FDA-approved vorinostat (SAHA) shows some blood brain barrier permeability [109], but to a lesser extent than, for example, MS-275 (entinostat, actual in Phase II clinical trials), a benzamide-based HDACi. The aforementioned MS-275 is an interesting candidate HDACi, for which one study has shown a regional selectivity with specific activity in frontal cortex, the striatum, and the hippocampus [122]. These regions are part of the neuronal circuitries underlying schizophrenia and might be a target for epigenetic modulations. Additionally, MS-275 was shown to be 30–100 times more potent than the well-established HDACi valproate in increasing histone-acetylation [46, 74, 122]. Therefore, MS-275 and other benzamide-based HDACi are currently considered as potentially promising for the treatment of neuropsychiatric disorders [46, 81]. In contrast to vorinostat (SAHA), no preclinical (animal) data for a beneficial use of entinostat to improve cognitive functions are yet available.

The ideal HDACi for the treatment of psychiatric disease should act in a brain region- and HDAC isoforms-specific manner and show a high potency for inhibiting HDACs, to allow a specific disease-related treatment and to reduce side effects. Obviously, such type of drug has not been described yet. To mention just two examples for the shortcomings of currently available HDACi, the HDACi valproate is only a weak HDACi, while the hydroxamate-based HDACi, vorinostat (SAHA), exhibits low isoform activity resulting in potentially severe and non-specific cytotoxicity [74].

Another major limitation that makes it difficult to design clinical trials to test HDACi in psychiatric conditions is that dose ranges, and other guidelines (including duration of treatment) for some of the newer HDACi are only available for cancer treatments. Furthermore, the application of HDACi is currently limited by severe side effects. This type of data has been derived from Phase II and Phase III trials, but a "translation" for the possible treatment of schizophrenia is not easily possible. According to the information of the package insert of romidipsin and vorinostat (SAHA), the mean duration in patients suffering from cutaneous T-cell lymphoma is short (below 6 months or no definite treatment durations has been defined), and it should be mentioned that no studies with a long study duration are available [24, 25]. This limits the application of these drugs in schizophrenia patients, as these patients might need long-term or even lifelong treatment. Even for some of the most frequently prescribed antipsychotics, no good evidence-based data are available to make statement for treatment duration with the highest grades of evidence. There are differences across guidelines, and the recently published WFSBP guideline recommends an antipsychotic treatment duration of 1–2 years in first-episode patients, 2–5 years in patients who have experienced one relapse, and over 5 years (maybe even lifelong) in patients with multiple episodes [55]. Another important question is which dose to choose? The package inserts for vorinostat

suggest a dose of 400 mg per os (po) daily with food and for romidepsin 14 mg/m^2 intravenously over 4 h on days 1, 8, and 15 in a 28-day cycle [21, 99]. This dosage information is not really useful for the application of these drugs in schizophrenia, but allows an appreciation for the dose ranges. However, dose-finding studies are necessary as HDACi in non-cancer patients may have another biological impact and interactions with antipsychotics might need further dose adaption.

Implications for clinical trials in schizophrenia

Apart from valproate, no HDACi has been investigated in experimental or clinical trials in patients suffering from schizophrenia. Notwithstanding the hope for a potential therapeutic effect, one must remember that HDACi are drugs with side effects. To date, only two drugs of this class are clinically approved, primarily for the application in treatment-resistant cutaneous T-cell lymphoma (SAHA and romidepsin). However, sodium phenylbutyrate (Ammonaps ®, triButyrate®) is approved as an orphan drug for the treatment of different urea cycle disorders. Sodium phenylbutyrate is a histone deacetylase inhibitor, but it is likely that its mechanisms of action in, for example, urea cycle disorders do not primarily involve histone acetylation [68]. Therefore, it will not be discussed in this review.

Contrary to DNA sequence mutations, epigenetic regulations are dynamic and therefore potentially reversible, which explains their appeal for drug development. [50]. However, other authors have made clear that a number of issues need to be addressed before any HDACi would be approved for the treatment of neuropsychiatric disorders [46, 74]. Issues ahead include (1) the selectivity and CNS bioavailability of HDACi, (2) identification of biomarkers with predictive value for treatment outcome, (3) choice of monotherapy versus combination therapy (especially with antipsychotics), and most importantly (4) safety and a benign side effect profile [46].

Safety profiles

Until now, no active clinical trial in schizophrenia patients is registered at www.clinicaltrials.gov, and as far as, we are informed no trial with patients suffering from schizophrenia or other psychosis is planned. This is of particular importance as no safety data in this patient group are available, which is an inherent limitation for a possible approval of trials in schizophrenia patients by local ethic committees and other involved authorities. However, many trials are listed for HDACi in cancer research and for that populations safety data are available. In cancer research, various adverse effects have been reported, which are within the range of mild to potential life-threatening (e.g. fatigue, gastrointestinal symptoms (diarrhea or vomiting), infections, anemia, thrombocytopenia, pulmonary embolism, increased bleeding propensity, muscle spasm, and most importantly cardiovascular toxicity, e.g. QTc prolongation) [21, 36, 99]. It is important to note that different HDACi show large differences in their toxicity profile owing to different molecular structures, pharmacokinetics, and pharmacodynamics [19, 74]. It is very likely that each HDACi could affect mitotic and post-mitotic cells (e.g. neurons) differently [46], resulting in broad side effects depending on the HDAC-selectivity, cell type affinity, and many other factors.

Although only limited data are available, carcinogenicity following HDACi treatment cannot be ruled out. For example, vorinostat (SAHA) was mutagenic in in vitro tests (Ames test, Chinese hamster ovary (CHO) cells) and increased the incidence of micro-nucleated erythrocytes (index of cytogenetic damage [123]) when administered to mice (mouse micronucleus assay) [99]. This potential carcinogenicity becomes important in the light of a potential long-term treatment in schizophrenia patients. However, further research is needed to clarify this issue.

For the two FDA-approved HDACi vorinostat (SAHA) and romidepsin, many of the aforementioned adverse effects with mild to moderate grades according to common toxicity criteria have been reported in a substantial amount of the investigated patients [21, 99]. However, even severe to life-threatening adverse effects (but no adverse affects related to death) have been reported in a substantial amount of patients treated with one of both drugs. This and the fact that HDACis have not been investigated in animal models of schizophrenia limits currently the application of modern HDACi in schizophrenia patients. There is a dire need for experimental data derived from animal models and a better understanding of HDACi dysregulation in schizophrenia.

Drug interactions and combination treatment

One important question is whether HDACi should be used as monotherapy in schizophrenia patients or whether they should be added to an ongoing antipsychotic treatment. Grayson et al. [46] stated that a monotherapy with a HDACi would not be suitable to treat disease with such a wide spectrum of symptoms like schizophrenia. Furthermore, in a bipolar disorder model of cultured cerebellar granule cells, lithium, and the HDACi valproate showed synergistic effects [86]. Despite these promising preclinical findings, combination therapy in schizophrenia (e.g. combining antipsychotics, or combining antipsychotics with antidepressants etc.) often is not superior to monotherapy,

but does increase side effects [56]. We cannot foresee the side effect rate of HDACi monotherapy versus combination therapy. However, combination therapy between HDACi and antipsychotics might increase side effects with consecutive risks for the patients. Consider the well-known example QTc prolongation and cardiotoxicity, as recent guidelines state clearly that a combination of antipsychotics with other drugs which prolong QTc time has to be avoided [56]. This is an important issue because some HDACi are known to prolong QTc time [9, 93]. From our point of view, future clinical trials with schizophrenia patients should be undertaken with a HDACi monotherapy to reduce the risk of adverse effects and to focus on clear hypotheses based on molecular findings.

Conclusions

HDACi are very promising agents to alter protein (including histone) acetylation levels in cells. Apart from their present day use in cancer therapy, additional medical and neuropsychiatric conditions, like schizophrenia, are currently actively discussed and explored by basic scientists and clinicians alike. Before regulatory authorities would approve a trial with HDACi, unresolved questions of drug safety and toxicity, target selectivity, treatment duration, and the treatment population need to be addressed. For future clinical trials, schizophrenia patients with severe cognitive deficits might be a suitable target populations as long as the patient is able to give consent. Furthermore, patients might be preselected by quantifying histone acetylation (e.g. lymphocytes)—a strategy that is based on using drug-induced histone acetylation as a biomarker, which might allow therapeutic monitoring [38, 46]. As HDACi are unspecific, more "specific therapies" might be better placed to improve the outcome of severe neuropsychiatric disorders. Bromodomain-acting drugs, which modulate the activity of histone acetyl transferases and the histone lysine acetylation, have a higher specificity and might therefore be a worthwhile drug target with higher specificity and potentially less side effects.

Is the time ripe for clinical trials with HDACi in schizophrenia patients? The answer is, probably, "not yet" but the available preclinical evidence is promising, and pending the availability of HDACi with a favorable safety profile, such trials should then give serious consideration. In the absence of good animal models for schizophrenia, with only limited research in that field and with regard to the severe side effects of the currently available drugs, clinical trials may be premature in this patient population at this time. However, this situation could change quickly, pending further advances in this rapidly evolving field.

The introduction of dopamine-D2-receptor antagonists revolutionized the treatment of schizophrenia in the 1950s, and the consecutive development of new antipsychotics reduced some major side effects (e.g. extrapyramidal side effects) and offered some alternative pharmacological interventions. Nevertheless, available treatment options are still limited, particularly for the cognitive deficits of the disorder. Perhaps HDACi, or some other type of chromatin-modifying drug, will emerge as a valuable treatment strategy for schizophrenia and other chronic psychiatric diseases.

Acknowledgments Work in the authors' laboratory is supported by grants from the National Institutes of Mental Health (USA).

Conflict of interest The authors reports no conflict of interest as it related to the content of this work.

References

1. Abdolmaleky HM, Cheng KH, Faraone SV, Wilcox M, Glatt SJ, Gao F, Smith CL, Shafa R, Aeali B, Carnevale J, Pan H, Papageorgis P, Ponte JF, Sivaraman V, Tsuang MT, Thiagalingam S (2006) Hypomethylation of MB-COMT promoter is a major risk factor for schizophrenia and bipolar disorder. Hum Mol Genet 15:3132–3145
2. Abujamra AL, Dos Santos MP, Roesler R, Schwartsmann G, Brunetto AL (2010) Histone deacetylase inhibitors: a new perspective for the treatment of leukemia. Leuk Res 34:687–695
3. Akbarian S, Huang HS (2006) Molecular and cellular mechanisms of altered GAD1/GAD67 expression in schizophrenia and related disorders. Brain Res Rev 52:293–304
4. Akbarian S, Ruehl MG, Bliven E, Luiz LA, Peranelli AC, Baker SP, Roberts RC, Bunney WE Jr, Conley RC, Jones EG, Tamminga CA, Guo Y (2005) Chromatin alterations associated with down-regulated metabolic gene expression in the prefrontal cortex of subjects with schizophrenia. Arch Gen Psychiatry 62:829–840
5. Alarcon JM, Malleret G, Touzani K, Vronskaya S, Ishii S, Kandel ER, Barco A (2004) Chromatin acetylation, memory, and LTP are impaired in CBP+/− mice: a model for the cognitive deficit in Rubinstein-Taybi syndrome and its amelioration. Neuron 42:947–959
6. Allen MH, Hirschfeld RM, Wozniak PJ, Baker JD, Bowden CL (2006) Linear relationship of valproate serum concentration to response and optimal serum levels for acute mania. Am J Psychiatry 163:272–275
7. An der Heiden W, Häfner H (2011) Course and outcome. In: Weinberger D, Harrison P (eds) Schizophrenia, 3rd edn. Wiley-Blackwell, Oxford
8. Aston C, Jiang L, Sokolov BP (2004) Microarray analysis of postmortem temporal cortex from patients with schizophrenia. J Neurosci Res 77:858–866
9. Bagnes C, Panchuk PN, Recondo G (2010) Antineoplastic chemotherapy induced QTc prolongation. Curr Drug Saf 5:93–96
10. Bahari-Javan S, Maddalena A, Kerimoglu C, Wittnam J, Held T, Bahr M, Burkhardt S, Delalle I, Kugler S, Fischer A, Sananbenesi F (2012) HDAC1 regulates fear extinction in mice. J Neurosci 32:5062–5073

11. Barrett RM, Malvaez M, Kramar E, Matheos DP, Arrizon A, Cabrera SM, Lynch G, Greene RW, Wood MA (2011) Hippocampal focal knockout of CBP affects specific histone modifications, long-term potentiation, and long-term memory. Neuropsychopharmacology 36:1545–1556
12. Baur JA, Pearson KJ, Price NL, Jamieson HA, Lerin C, Kalra A, Prabhu VV, Allard JS, Lopez-Lluch G, Lewis K, Pistell PJ, Poosala S, Becker KG, Boss O, Gwinn D, Wang M, Ramaswamy S, Fishbein KW, Spencer RG, Lakatta EG, Le Couteur D, Shaw RJ, Navas P, Puigserver P, Ingram DK, de Cabo R, Sinclair DA (2006) Resveratrol improves health and survival of mice on a high-calorie diet. Nature 444:337–342
13. Benes FM (2010) Amygdalocortical circuitry in schizophrenia: from circuits to molecules. Neuropsychopharmacology 35:239–257
14. Benes FM, Lim B, Matzilevich D, Walsh JP, Subburaju S, Minns M (2007) Regulation of the GABA cell phenotype in hippocampus of schizophrenics and bipolars. Proc Natl Acad Sci USA 104:10164–10169
15. Beneyto M, Kristiansen LV, Oni-Orisan A, McCullumsmith RE, Meador-Woodruff JH (2007) Abnormal glutamate receptor expression in the medial temporal lobe in schizophrenia and mood disorders. Neuropsychopharmacology 32:1888–1902
16. Berger SL (2007) The complex language of chromatin regulation during transcription. Nature 447:407–412
17. Bora E, Yucel M, Pantelis C (2009) Cognitive functioning in schizophrenia, schizoaffective disorder and affective psychoses: meta-analytic study. Br J Psychiatry 195:475–482
18. Bousiges O, Vasconcelos AP, Neidl R, Cosquer B, Herbeaux K, Panteleeva I, Loeffler JP, Cassel JC, Boutillier AL (2010) Spatial memory consolidation is associated with induction of several lysine-acetyltransferase (histone acetyltransferase) expression levels and H2B/H4 acetylation-dependent transcriptional events in the rat hippocampus. Neuropsychopharmacology 35:2521–2537
19. Bruserud O, Stapnes C, Ersvaer E, Gjertsen BT, Ryningen A (2007) Histone deacetylase inhibitors in cancer treatment: a review of the clinical toxicity and the modulation of gene expression in cancer cell. Curr Pharm Biotechnol 8:388–400
20. Buchanan RW, Kreyenbuhl J, Kelly DL, Noel JM, Boggs DL, Fischer BA, Himelhoch S, Fang B, Peterson E, Aquino PR, Keller W (2010) The 2009 schizophrenia PORT psychopharmacological treatment recommendations and summary statements. Schizophr Bull 36:71–93
21. Celegene (2012) Presribing Information ISTODAX (romidepsin) for injection: Update Manufacturing Information 3/2012
22. Charych EI, Liu F, Moss SJ, Brandon NJ (2009) GABA(A) receptors and their associated proteins: implications in the etiology and treatment of schizophrenia and related disorders. Neuropharmacology 57:481–495
23. Chiesa A, Chierzi F, De Ronchi D, Serretti A (2012) Quetiapine for bipolar depression: a systematic review and meta-analysis. Int Clin Psychopharmacol 27:76–90
24. Co M (2011) Presribing Information ZOLINZA (vorinostat) capsules: Revised 11/2011
25. Corporation C (2012) Presribing information ISTODAX (romidepsin) for injection: update manufacturing information 3/2012
26. Costa E, Chen Y, Dong E, Grayson DR, Kundakovic M, Maloku E, Ruzicka W, Satta R, Veldic M, Zhubi A, Guidotti A (2009) GABAergic promoter hypermethylation as a model to study the neurochemistry of schizophrenia vulnerability. Expert Rev Neurother 9:87–98
27. Crow TJ (2011) The missing genes: what happened to the heritability of psychiatric disorders? Mol Psychiatry 16:362–364
28. Daxinger L, Whitelaw E (2012) Understanding transgenerational epigenetic inheritance via the gametes in mammals. Nat Rev Genet 13:153–162
29. Doi M, Hirayama J, Sassone-Corsi P (2006) Circadian regulator CLOCK is a histone acetyltransferase. Cell 125:497–508
30. Dokmanovic M, Clarke C, Marks PA (2007) Histone deacetylase inhibitors: overview and perspectives. Mol Cancer Res 5:981–989
31. Dong E, Nelson M, Grayson DR, Costa E, Guidotti A (2008) Clozapine and sulpiride but not haloperidol or olanzapine activate brain DNA demethylation. Proc Natl Acad Sci USA 105:13614–13619
32. Dracheva S, Elhakem SL, McGurk SR, Davis KL, Haroutunian V (2004) GAD67 and GAD65 mRNA and protein expression in cerebrocortical regions of elderly patients with schizophrenia. J Neurosci Res 76:581–592
33. Duncan CE, Webster MJ, Rothmond DA, Bahn S, Elashoff M, Shannon Weickert C (2010) Prefrontal GABA(A) receptor alpha-subunit expression in normal postnatal human development and schizophrenia. J Psychiatr Res 44(10):678–681
34. Echaniz-Laguna A, Bousiges O, Loeffler JP, Boutillier AL (2008) Histone deacetylase inhibitors: therapeutic agents and research tools for deciphering motor neuron diseases. Curr Med Chem 15:1263–1273
35. Ederveen TH, Mandemaker IK, Logie C (2011) The human histone H3 complement anno 2011. Biochim Biophys Acta 1809:577–586
36. Elaut G, Rogiers V, Vanhaecke T (2007) The pharmaceutical potential of histone deacetylase inhibitors. Curr Pharm Des 13:2584–2620
37. Fischer A, Sananbenesi F, Mungenast A, Tsai LH (2011) Targeting the correct HDAC(s) to treat cognitive disorders. Trends Pharmacol Sci 31:605–617
38. Gavin DP, Kartan S, Chase K, Grayson DR, Sharma RP (2008) Reduced baseline acetylated histone 3 levels, and a blunted response to HDAC inhibition in lymphocyte cultures from schizophrenia subjects. Schizophr Res 103:330–332
39. Goff DC, Hill M, Barch D (2010) The treatment of cognitive impairment in schizophrenia. Pharmacol Biochem Behav 99:245–253
40. Gold JM (2004) Cognitive deficits as treatment targets in schizophrenia. Schizophr Res 72:21–28
41. Gottlicher M (2004) Valproic acid: an old drug newly discovered as inhibitor of histone deacetylases. Ann Hematol 83(Suppl 1): S91–S92
42. Gottlicher M, Minucci S, Zhu P, Kramer OH, Schimpf A, Giavara S, Sleeman JP, Lo Coco F, Nervi C, Pelicci PG, Heinzel T (2001) Valproic acid defines a novel class of HDAC inhibitors inducing differentiation of transformed cells. EMBO J 20:6969–6978
43. Govindarajan N, Rao P, Burkhardt S, Sananbenesi F, Schlüter OM, Bradke F, Lu J, Fischer A (2013) Reducing HDAC6 ameliorates cognitive deficits in a mouse model for Alzheimer's disease. EMBO Mol Med 5(1):52–63. doi:10.1002/emmm.201201923
44. Graff J, Rei D, Guan JS, Wang WY, Seo J, Hennig KM, Nieland TJ, Fass DM, Kao PF, Kahn M, Su SC, Samiei A, Joseph N, Haggarty SJ, Delalle I, Tsai LH (2012) An epigenetic blockade of cognitive functions in the neurodegenerating brain. Nature 483:222–226
45. Grayson DR, Jia X, Chen Y, Sharma RP, Mitchell CP, Guidotti A, Costa E (2005) Reelin promoter hypermethylation in schizophrenia. Proc Natl Acad Sci USA 102:9341–9346
46. Grayson DR, Kundakovic M, Sharma RP (2010) Is there a future for histone deacetylase inhibitors in the pharmacotherapy of psychiatric disorders? Mol Pharmacol 77:126–135

47. Green MF (1996) What are the functional consequences of neurocognitive deficits in schizophrenia? Am J Psychiatry 153:321–330
48. Griswold KS, Pessar LF (2000) Management of bipolar disorder. Am Fam Physician 62:1343–1358
49. Guan JS, Haggarty SJ, Giacometti E, Dannenberg JH, Joseph N, Gao J, Nieland TJ, Zhou Y, Wang X, Mazitschek R, Bradner JE, DePinho RA, Jaenisch R, Tsai LH (2009) HDAC2 negatively regulates memory formation and synaptic plasticity. Nature 459:55–60
50. Guidotti A, Auta J, Chen Y, Davis JM, Dong E, Gavin DP, Grayson DR, Matrisciano F, Pinna G, Satta R, Sharma RP, Tremolizzo L, Tueting P (2011) Epigenetic GABAergic targets in schizophrenia and bipolar disorder. Neuropharmacology 60:1007–1016
51. Guidotti A, Auta J, Davis JM, Dong E, Grayson DR, Veldic M, Zhang X, Costa E (2005) GABAergic dysfunction in schizophrenia: new treatment strategies on the horizon. Psychopharmacology 180:191–205
52. Hakak Y, Walker JR, Li C, Wong WH, Davis KL, Buxbaum JD, Haroutunian V, Fienberg AA (2001) Genome-wide expression analysis reveals dysregulation of myelination-related genes in chronic schizophrenia. Proc Natl Acad Sci USA 98:4746–4751
53. Halene TB, Siegel SJ (2007) PDE inhibitors in psychiatry—future options for dementia, depression and schizophrenia? Drug Discov Today 12:870–878
54. Hammoud SS, Nix DA, Zhang H, Purwar J, Carrell DT, Cairns BR (2009) Distinctive chromatin in human sperm packages genes for embryo development. Nature 460:473–478
55. Hasan A, Falkai P, Wobrock T, Lieberman J, Glenthoj B, Gattaz WF, Thibault P, Möller HJ (2013) World Federation of Societies of Biological Psychiatry (WFSBP): Guidelines for biological treatment of schizophrenia, Part 2: update 2012 on the long-term treatment of schizophrenia and management of antipsychotic-induced side effects. World J Biol Psychiatry 14(1):2–44. doi: 10.3109/15622975.2012.739708
56. Hasan A, Falkai P, Wobrock T, Lieberman J, Glenthoj B, Gattaz WF, Thibaut F, Moller HJ (2012) World Federation of Societies of Biological Psychiatry (WFSBP) Guidelines for Biological Treatment of Schizophrenia, part 1: update 2012 on the acute treatment of schizophrenia and the management of treatment resistance. World J Biol Psychiatry 13:318–378
57. Hasan A, Nitsche MA, Rein B, Schneider-Axmann T, Guse B, Gruber O, Falkai P, Wobrock T (2011) Dysfunctional long-term potentiation-like plasticity in schizophrenia revealed by transcranial direct current stimulation. Behav Brain Res 224:15–22
58. Hashimoto T, Bazmi HH, Mirnics K, Wu Q, Sampson AR, Lewis DA (2008) Conserved regional patterns of GABA-related transcript expression in the neocortex of subjects with schizophrenia. Am J Psychiatry 165:479–489
59. Heinrichs RW, Zakzanis KK (1998) Neurocognitive deficit in schizophrenia: a quantitative review of the evidence. Neuropsychology 12:426–445
60. Hemby SE, Ginsberg SD, Brunk B, Arnold SE, Trojanowski JQ, Eberwine JH (2002) Gene expression profile for schizophrenia: discrete neuron transcription patterns in the entorhinal cortex. Arch Gen Psychiatry 59:631–640
61. Hirsch E, Schmitz B, Carreno M (2003) Epilepsy, antiepileptic drugs (AEDs) and cognition. Acta Neurol Scand Suppl 180:23–32
62. Hoe M, Nakagami E, Green MF, Brekke JS (2012) The causal relationships between neurocognition, social cognition and functional outcome over time in schizophrenia: a latent difference score approach. Psychol Med 42:1–13
63. Holt DJ, Lebron-Milad K, Milad MR, Rauch SL, Pitman RK, Orr SP, Cassidy BS, Walsh JP, Goff DC (2009) Extinction memory is impaired in schizophrenia. Biol Psychiatry 65:455–463
64. Horan WP, Kern RS, Harvey PO, Green MF (2011) Neurocognition, social cogniton, and functional outcome in schizophrenia. In: Gaebel W (ed) Schizophenia—current science and clinical practice. Wiley, Chichester
65. Hu E, Dul E, Sung CM, Chen Z, Kirkpatrick R, Zhang GF, Johanson K, Liu R, Lago A, Hofmann G, Macarron R, De Los Frailes M, Perez P, Krawiec J, Winkler J, Jaye M (2003) Identification of novel isoform-selective inhibitors within class I histone deacetylases. J Pharmacol Exp Ther 307:720–728
66. Huang HS, Akbarian S (2007) GAD1 mRNA expression and DNA methylation in prefrontal cortex of subjects with schizophrenia. PLoS One 2:e809
67. Huang HS, Matevossian A, Whittle C, Kim SY, Schumacher A, Baker SP, Akbarian S (2007) Prefrontal dysfunction in schizophrenia involves mixed-lineage leukemia 1-regulated histone methylation at GABAergic gene promoters. J Neurosci 27:11254–11262
68. Iannitti T, Palmieri B (2011) Clinical and experimental applications of sodium phenylbutyrate. Drugs R D 11:227–249
69. Ibrahim HM, Tamminga CA (2011) Schizophrenia: treatment targets beyond monoamine systems. Annu Rev Pharmacol Toxicol 51:189–209
70. Iwamoto K, Bundo M, Yamada K, Takao H, Iwayama-Shigeno Y, Yoshikawa T, Kato T (2005) DNA methylation status of SOX10 correlates with its downregulation and oligodendrocyte dysfunction in schizophrenia. J Neurosci 25:5376–5381
71. Kalin JH, Butler KV, Kozikowski AP (2009) Creating zinc monkey wrenches in the treatment of epigenetic disorders. Curr Opin Chem Biol 13:263–271
72. Kano S, Colantuoni C, Han F, Zhou Z, Yuan Q, Wilson A, Takayanagi Y, Lee Y, Rapoport J, Eaton W, Cascella N, Ji H, Goldman D, Sawa A (2012) Genome-wide profiling of multiple histone methylations in olfactory cells: further implications for cellular susceptibility to oxidative stress in schizophrenia. Mol Psychiatry. doi:10.1038/mp.2012.120
73. Katsel P, Davis KL, Haroutunian V (2005) Variations in myelin and oligodendrocyte-related gene expression across multiple brain regions in schizophrenia: a gene ontology study. Schizophr Res 79:157–173
74. Kazantsev AG, Thompson LM (2008) Therapeutic application of histone deacetylase inhibitors for central nervous system disorders. Nat Rev Drug Discov 7:854–868
75. Khan N, Jeffers M, Kumar S, Hackett C, Boldog F, Khramtsov N, Qian X, Mills E, Berghs SC, Carey N, Finn PW, Collins LS, Tumber A, Ritchie JW, Jensen PB, Lichenstein HS, Sehested M (2008) Determination of the class and isoform selectivity of small-molecule histone deacetylase inhibitors. Biochem J 409:581–589
76. Kinney SM, Chin HG, Vaisvila R, Bitinaite J, Zheng Y, Esteve PO, Feng S, Stroud H, Jacobsen SE, Pradhan S (2011) Tissue-specific distribution and dynamic changes of 5-hydroxymethylcytosine in mammalian genomes. J Biol Chem 286:24685–24693
77. Korzus E, Rosenfeld MG, Mayford M (2004) CBP histone acetyltransferase activity is a critical component of memory consolidation. Neuron 42:961–972
78. Kouraklis G, Theocharis S (2006) Histone deacetylase inhibitors: a novel target of anticancer therapy (review). Oncol Rep 15:489–494
79. Kouzarides T (2007) Chromatin modifications and their function. Cell 128:693–705
80. Krishnan V, Nestler EJ (2010) Linking molecules to mood: new insight into the biology of depression. Am J Psychiatry 167:1305–1320

81. Kundakovic M, Chen Y, Guidotti A, Grayson DR (2009) The reelin and GAD67 promoters are activated by epigenetic drugs that facilitate the disruption of local repressor complexes. Mol Pharmacol 75:342–354
82. Kurita M, Holloway T, Garcia-Bea A, Kozlenkov A, Friedman AK, Moreno JL, Heshmati M, Golden SA, Kennedy PJ, Takahashi N, Dietz DM, Mocci G, Gabilondo AM, Hanks J, Umali A, Callado LF, Gallitano AL, Neve RL, Shen L, Buxbaum JD, Han MH, Nestler EJ, Meana JJ, Russo SJ, Gonzalez-Maeso J (2012) HDAC2 regulates atypical antipsychotic responses through the modulation of mGlu2 promoter activity. Nat Neurosci 15:1245–1254
83. Lattal KM, Barrett RM, Wood MA (2007) Systemic or intrahippocampal delivery of histone deacetylase inhibitors facilitates fear extinction. Behav Neurosci 121:1125–1131
84. Lee H, Dvorak D, Kao HY, Duffy AM, Scharfman HE, Fenton AA (2012) Early cognitive experience prevents adult deficits in a neurodevelopmental schizophrenia model. Neuron 75:714–724
85. Lehman AF, Lieberman JA, Dixon LB, McGlashan TH, Miller AL, Perkins DO, Kreyenbuhl J (2004) Practice guideline for the treatment of patients with schizophrenia, second edition. Am J Psychiatry 161:1–56
86. Leng Y, Liang MH, Ren M, Marinova Z, Leeds P, Chuang DM (2008) Synergistic neuroprotective effects of lithium and valproic acid or other histone deacetylase inhibitors in neurons: roles of glycogen synthase kinase-3 inhibition. J Neurosci 28:2576–2588
87. Lewis DA, Hashimoto T, Volk DW (2005) Cortical inhibitory neurons and schizophrenia. Nat Rev Neurosci 6:312–324
88. Li G, Reinberg D (2011) Chromatin higher-order structures and gene regulation. Curr Opin Genet Dev 21:175–186
89. Li J, Guo Y, Schroeder FA, Youngs RM, Schmidt TW, Ferris C, Konradi C, Akbarian S (2004) Dopamine D2-like antagonists induce chromatin remodeling in striatal neurons through cyclic AMP-protein kinase A and NMDA receptor signaling. J Neurochem 90:1117–1131
90. Li Q, Barkess G, Qian H (2006) Chromatin looping and the probability of transcription. Trends Genet 22:197–202
91. Libert S, Pointer K, Bell EL, Das A, Cohen DE, Asara JM, Kapur K, Bergmann S, Preisig M, Otowa T, Kendler KS, Chen X, Hettema JM, van den Oord EJ, Rubio JP, Guarente L (2011) SIRT1 activates MAO-A in the brain to mediate anxiety and exploratory drive. Cell 147:1459–1472
92. Lieberman JA, Stroup TS, McEvoy JP, Swartz MS, Rosenheck RA, Perkins DO, Keefe RS, Davis SM, Davis CE, Lebowitz BD, Severe J, Hsiao JK (2005) Effectiveness of antipsychotic drugs in patients with chronic schizophrenia. N Engl J Med 353:1209–1223
93. Lynch DR Jr, Washam JB, Newby LK (2012) QT interval prolongation and torsades de pointes in a patient undergoing treatment with vorinostat: a case report and review of the literature. Cardiol J 19:434–438
94. Martins-de-Souza D, Gattaz WF, Schmitt A, Rewerts C, Marangoni S, Novello JC, Maccarrone G, Turck CW, Dias-Neto E (2009) Alterations in oligodendrocyte proteins, calcium homeostasis and new potential markers in schizophrenia anterior temporal lobe are revealed by shotgun proteome analysis. J Neural Transm 116:275–289
95. Masetti R, Serravalle S, Biagi C, Pession A (2011) The role of HDACs inhibitors in childhood and adolescence acute leukemias. J Biomed Biotechnol 2011:148046
96. McQuown SC, Barrett RM, Matheos DP, Post RJ, Rogge GA, Alenghat T, Mullican SE, Jones S, Rusche JR, Lazar MA, Wood MA (2011) HDAC3 is a critical negative regulator of long-term memory formation. J Neurosci 31:764–774
97. McQuown SC, Wood MA (2011) HDAC3 and the molecular brake pad hypothesis. Neurobiol Learn Mem 96:27–34
98. Meador-Woodruff JH, Healy DJ (2000) Glutamate receptor expression in schizophrenic brain. Brain Res Brain Res Rev 31:288–294
99. Merck (2011) Presribing information ZOLINZA (vorinostat) capsules: revised 11/2011
100. Mesholam-Gately RI, Giuliano AJ, Goff KP, Faraone SV, Seidman LJ (2009) Neurocognition in first-episode schizophrenia: a meta-analytic review. Neuropsychology 23:315–336
101. Mill J, Tang T, Kaminsky Z, Khare T, Yazdanpanah S, Bouchard L, Jia P, Assadzadeh A, Flanagan J, Schumacher A, Wang SC, Petronis A (2008) Epigenomic profiling reveals DNA-methylation changes associated with major psychosis. Am J Hum Genet 82:696–711
102. Miyamoto S, Miyake N, Jarskog LF, Fleischhacker WW, Lieberman JA (2012) Pharmacological treatment of schizophrenia: a critical review of the pharmacology and clinical effects of current and future therapeutic agents. Mol Psychiatry 17:1206–1227
103. Mohamed S, Paulsen JS, O'Leary D, Arndt S, Andreasen N (1999) Generalized cognitive deficits in schizophrenia: a study of first-episode patients. Arch Gen Psychiatry 56:749–754
104. Monsey MS, Ota KT, Akingbade IF, Hong ES, Schafe GE (2011) Epigenetic alterations are critical for fear memory consolidation and synaptic plasticity in the lateral amygdala. PLoS One 6:e19958
105. Mouri A, Nagai T, Ibi D, Yamada K (2012) Animal models of schizophrenia for molecular and pharmacological intervention and potential candidate molecules. Neurobiol Dis. doi:10.1016/j.nbd.2012.10.025
106. Narayan PJ, Dragunow M (2010) High content analysis of histone acetylation in human cells and tissues. J Neurosci Methods 193:54–61
107. Olabi B, Ellison-Wright I, McIntosh AM, Wood SJ, Bullmore E, Lawrie SM (2011) Are there progressive brain changes in schizophrenia? A meta-analysis of structural magnetic resonance imaging studies. Biol Psychiatry 70:88–96
108. Palmer BW, Heaton RK, Paulsen JS, Kuck J, Braff D, Harris MJ, Zisook S, Jeste DV (1997) Is it possible to be schizophrenic yet neuropsychologically normal? Neuropsychology 11:437–446
109. Palmieri D, Lockman PR, Thomas FC, Hua E, Herring J, Hargrave E, Johnson M, Flores N, Qian Y, Vega-Valle E, Taskar KS, Rudraraju V, Mittapalli RK, Gaasch JA, Bohn KA, Thorsheim HR, Liewehr DJ, Davis S, Reilly JF, Walker R, Bronder JL, Feigenbaum L, Steinberg SM, Camphausen K, Meltzer PS, Richon VM, Smith QR, Steeg PS (2009) Vorinostat inhibits brain metastatic colonization in a model of triple-negative breast cancer and induces DNA double-strand breaks. Clin Cancer Res 15:6148–6157
110. Peleg S, Sananbenesi F, Zovoilis A, Burkhardt S, Bahari-Javan S, Agis-Balboa RC, Cota P, Wittnam JL, Gogol-Doering A, Opitz L, Salinas-Riester G, Dettenhofer M, Kang H, Farinelli L, Chen W, Fischer A (2010) Altered histone acetylation is associated with age-dependent memory impairment in mice. Science 328:753–756
111. Phiel CJ, Zhang F, Huang EY, Guenther MG, Lazar MA, Klein PS (2001) Histone deacetylase is a direct target of valproic acid, a potent anticonvulsant, mood stabilizer, and teratogen. J Biol Chem 276:36734–36741
112. Reagan-Shaw S, Nihal M, Ahmad N (2008) Dose translation from animal to human studies revisited. FASEB J 22:659–661
113. Regenold WT, Phatak P, Marano CM, Gearhart L, Viens CH, Hisley KC (2007) Myelin staining of deep white matter in the

dorsolateral prefrontal cortex in schizophrenia, bipolar disorder, and unipolar major depression. Psychiatry Res 151:179–188
114. Rehan VK, Liu J, Naeem E, Tian J, Sakurai R, Kwong K, Akbari O, Torday JS (2012) Perinatal nicotine exposure induces asthma in second generation offspring. BMC Med 10:129
115. Rodriguez-Paredes M, Esteller M (2011) Cancer epigenetics reaches mainstream oncology. Nat Med 17:330–339
116. Sananbenesi F, Fischer A (2009) The epigenetic bottleneck of neurodegenerative and psychiatric diseases. Biol Chem 390:1145–1153
117. Schmitt A, Hasan A, Gruber O, Falkai P (2011) Schizophrenia as a disorder of disconnectivity. Eur Arch Psychiatry Clin Neurosci 261(Suppl 2):S150–S154
118. Sharma RP, Grayson DR, Gavin DP (2008) Histone deactylase 1 expression is increased in the prefrontal cortex of schizophrenia subjects: analysis of the National Brain Databank microarray collection. Schizophr Res 98:111–117
119. Shulha HP, Cheung I, Whittle C, Wang J, Virgil D, Lin CL, Guo Y, Lessard A, Akbarian S, Weng Z (2012) Epigenetic signatures of autism: trimethylated H3K4 landscapes in prefrontal neurons. Arch Gen Psychiatry 69:314–324
120. Silverstein SM, Schenkel LS, Valone C, Nuernberger SW (1998) Cognitive deficits and psychiatric rehabilitation outcomes in schizophrenia. Psychiatr Q 69:169–191
121. Simon M, North JA, Shimko JC, Forties RA, Ferdinand MB, Manohar M, Zhang M, Fishel R, Ottesen JJ, Poirier MG (2011) Histone fold modifications control nucleosome unwrapping and disassembly. Proc Natl Acad Sci USA 108:12711–12716
122. Simonini MV, Camargo LM, Dong E, Maloku E, Veldic M, Costa E, Guidotti A (2006) The benzamide MS-275 is a potent, long-lasting brain region-selective inhibitor of histone deacetylases. Proc Natl Acad Sci USA 103:1587–1592
123. Smith DF, MacGregor JT, Hiatt RA, Hooper NK, Wehr CM, Peters B, Goldman LR, Yuan LA, Smith PA, Becker CE (1990) Micronucleated erythrocytes as an index of cytogenetic damage in humans: demographic and dietary factors associated with micronucleated erythrocytes in splenectomized subjects. Cancer Res 50:5049–5054
124. Snitz BE, Macdonald AW 3rd, Carter CS (2006) Cognitive deficits in unaffected first-degree relatives of schizophrenia patients: a meta-analytic review of putative endophenotypes. Schizophr Bull 32:179–194
125. Tamayo JM, Zarate CA Jr, Vieta E, Vazquez G, Tohen M (2010) Level of response and safety of pharmacological monotherapy in the treatment of acute bipolar I disorder phases: a systematic review and meta-analysis. Int J Neuropsychopharmacol 13:813–832
126. Tan M, Luo H, Lee S, Jin F, Yang JS, Montellier E, Buchou T, Cheng Z, Rousseaux S, Rajagopal N, Lu Z, Ye Z, Zhu Q, Wysocka J, Ye Y, Khochbin S, Ren B, Zhao Y (2011) Identification of 67 histone marks and histone lysine crotonylation as a new type of histone modification. Cell 146:1016–1028
127. Tandon R, Keshavan MS, Nasrallah HA (2008) Schizophrenia, "just the facts" what we know in 2008. 2. Epidemiology and etiology. Schizophr Res 102:1–18
128. Tang B, Dean B, Thomas EA (2011) Disease- and age-related changes in histone acetylation at gene promoters in psychiatric disorders. Transl Psychiatry 1:e64
129. Taverna SD, Li H, Ruthenburg AJ, Allis CD, Patel DJ (2007) How chromatin-binding modules interpret histone modifications: lessons from professional pocket pickers. Nat Struct Mol Biol 14:1025–1040
130. Thornicroft G, Tansella M, Becker T, Knapp M, Leese M, Schene A, Vazquez-Barquero JL (2004) The personal impact of schizophrenia in Europe. Schizophr Res 69:125–132
131. Tkachev D, Mimmack ML, Ryan MM, Wayland M, Freeman T, Jones PB, Starkey M, Webster MJ, Yolken RH, Bahn S (2003) Oligodendrocyte dysfunction in schizophrenia and bipolar disorder. Lancet 362:798–805
132. Tremolizzo L, Carboni G, Ruzicka WB, Mitchell CP, Sugaya I, Tueting P, Sharma R, Grayson DR, Costa E, Guidotti A (2002) An epigenetic mouse model for molecular and behavioral neuropathologies related to schizophrenia vulnerability. Proc Natl Acad Sci USA 99:17095–17100
133. Vecsey CG, Hawk JD, Lattal KM, Stein JM, Fabian SA, Attner MA, Cabrera SM, McDonough CB, Brindle PK, Abel T, Wood MA (2007) Histone deacetylase inhibitors enhance memory and synaptic plasticity via CREB:CBP-dependent transcriptional activation. J Neurosci 27:6128–6140
134. Wang Z, Yang D, Zhang X, Li T, Li J, Tang Y, Le W (2011) Hypoxia-induced down-regulation of neprilysin by histone modification in mouse primary cortical and hippocampal neurons. PLoS One 6:e19229
135. Woo TU, Kim AM, Viscidi E (2008) Disease-specific alterations in glutamatergic neurotransmission on inhibitory interneurons in the prefrontal cortex in schizophrenia. Brain Res 1218:267–277
136. Wood MA, Attner MA, Oliveira AM, Brindle PK, Abel T (2006) A transcription factor-binding domain of the coactivator CBP is essential for long-term memory and the expression of specific target genes. Learn Mem 13:609–617
137. Wu Y, Blichowski M, Daskalakis ZJ, Wu Z, Liu CC, Cortez MA, Snead OC 3rd (2011) Evidence that clozapine directly interacts on the GABAB receptor. NeuroReport 22:637–641
138. Yildirim E, Zhang Z, Uz T, Chen CQ, Manev R, Manev H (2003) Valproate administration to mice increases histone acetylation and 5-lipoxygenase content in the hippocampus. Neurosci Lett 345:141–143
139. Zhou VW, Goren A, Bernstein BE (2011) Charting histone modifications and the functional organization of mammalian genomes. Nat Rev Genet 12:7–18