

## *Epigenetic alterations in depression and antidepressant treatment*

Andreas Menke, MD; Elisabeth B. Binder, MD, PhD



*Epigenetic modifications control chromatin structure and function, and thus mediate changes in gene expression, ultimately influencing protein levels. Recent research indicates that environmental events can induce epigenetic changes and, by this, contribute to long-term changes in neural circuits and endocrine systems associated with altered risk for stress-related psychiatric disorders such as major depression. In this review, we describe recent approaches investigating epigenetic modifications associated with altered risk for major depression or response to antidepressant drugs, both on the candidate gene levels as well as the genome-wide level. In this review we focus on DNA methylation, as this is the most investigated epigenetic change in depression research.*

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*Dialogues Clin Neurosci.* 2014;16:395–404.

**Keywords:** acetylation; antidepressant; epigenetics; epigenome-wide association study; EWAS; histone modification; major depression; methylation

**Author affiliations:** Department of Translational Psychiatry, Max Planck Institute of Psychiatry, Munich, Germany (Andreas Menke, Elisabeth Binder); Department of Psychiatry and Behavioral Sciences, Emory University School of Medicine, Atlanta, Georgia, USA (Elisabeth Binder)

### Introduction

**M**ajor depression accounts for more disability than most other diseases worldwide. It is associated with significant morbidity, mortality, increased medical comorbidities, and disability.<sup>1</sup> Additionally, the efficacy of currently available antidepressant treatment strategies is suboptimal,<sup>2</sup> and strategies to improve treatment are hampered by our still-limited understanding of the pathophysiology of major depression. A fact that is well established is that genetic factors together with environmental factors occurring across the lifespan likely underlie the vulnerability for depression.<sup>3</sup> A meta-analysis derived from five studies including more than 21 000 subjects revealed a moderate genetic contribution of 37% (95% confidence interval [CI], 31% to 42%) for unipolar depression.<sup>4</sup> Common environmental influences had very small effects, while individual environmental factors showed a significant contribution of 63% (95% CI, 58% to 67%). When considering severity of illness, relapse, and early onset of disease, the estimated heritability can increase to up to 70% in specific studies.<sup>5</sup> So far, however, no specific genetic variants have been identified to robustly contribute to

**Address for correspondence:** Andreas Menke, MD, Max Planck Institute of Psychiatry, Kraepelinstr. 10, D-80804 Munich, Germany (e-mail: menke@mpipsykl.mpg.de)

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major depression. A mega-analysis of several genome-wide association studies (GWAS), which analyzed 1.2 million autosomal and X chromosome single-nucleotide polymorphisms (SNPs) in a discovery phase with 9240 depressed patients and 9519 healthy controls, and 554 SNPs in a replication phase with 6783 depressed patients and 50695 healthy controls, found no SNP achieving genome-wide significance.<sup>6</sup> This discrepancy between the consistently observed substantial heritability of major depression and the failure to successfully replicate detected genes has been attributed to several factors.<sup>7,8</sup> One hypothesis is balancing selection,<sup>7</sup> a balance between genetic advantage, for example creative intelligence, and genetic disadvantage, for example psychotic symptoms, is assumed, which would preserve the gene variant throughout generations.<sup>9</sup> An alternative hypothesis, ancestral neutrality, assumes that genetic variants were neutral throughout most of human evolution and have only recently become harmful.<sup>10,11</sup> The hypothesis of Keller and Miller, the polygenic mutation selection balance, proposes that mutations with harmful effects are eliminated very fast, thus they are very rare and, if present, recent.<sup>7</sup> However, mutations with small effects are eliminated slowly; thus they are more common, older, and remain longer in a population. In addition, genetic factors contributing to risk for major depression may not be selected against at all, as this disorder is not associated with an obvious reduction in fertility.<sup>12</sup> The identification of genetic effects with small relative risks requires very large samples, which would suggest that even larger samples will be required to reliably detect risk genes.<sup>7</sup> Furthermore, none of the GWAS so far have incorporated environmental risk factors, and it is possible that these genetic risk factors are only unmasked conditional to a specific environmental exposure. Genome-wide gene x environment studies may address this issue, but are fraught with a number of problems, such as the need for even larger samples for sufficient power, the heterogeneity of environmental exposures in type, timing, and severity, and the lack of reliable measures or documentation of such exposures, especially in cross-sectional studies.

Genetic variation and environmental exposure have both been shown to impinge on epigenetic factors,<sup>13</sup> and gene-environment interactions may be mediated by such changes. Investigating epigenetic factors in depression may thus allow an integrated view of how genetic and environmental factors alter risk. Epigenetics refers

to the changes in chromatin structure that underlie changes in gene expression and that are not associated with alterations in DNA sequence. Recent findings of changed epigenetic marks in depression are the focus of the remainder of this article. For a review of the importance of epigenetic factors coming from animal models, we refer the reader to several excellent articles; for example references 13 and 14.

## DNA methylation and demethylation

DNA methylation involves direct chemical modifications to the DNA and has been discovered in mammals at the same time as DNA was identified as the carrier of genetic information.<sup>15,16</sup> Methylation of the DNA requires the addition of a methyl group at the 5' position of cytosines in CpG dinucleotides.<sup>17</sup> However, nonCpG cytosines such as CpA, CpC, or CpT dinucleotides may also be modified, but to a lesser extent. Methylation of a single CpG site within a single DNA strand is binary. Tissue samples used for research, however, usually contain a large number of DNA strands. Therefore, the DNA methylation at a single CpG site in a specific sample becomes a quantitative trait, with a certain percentage of DNA strands methylated.<sup>18</sup> Following cytosine methylation, access of transcription factors to regulatory elements is reduced. If the binding site is for enhancers, DNA methylation is associated with transcriptional repression, if the binding site is for repressors, DNA methylation will have the opposite effect on transcription.<sup>19,20</sup> DNA methylation is catalyzed by DNA methyltransferases (DNMTs), a family of enzymes including DNMT1, DNMT2, DNMT3a, and DNMT3b.<sup>21</sup> DNMT1 preserves the methylation patterns during DNA replication, DNMT3a and DNMT3b perform de novo methylation of double-stranded DNA.<sup>17,22</sup> In contrast to methylation, demethylation by an active and not passive process has been discussed controversially in the past. It was known that passive demethylation could occur when maintenance methylation does not take place before cell division. However, passive demethylation during the cell cycle would not explain the remarkably fast and flexible reconfiguration of DNA methylation in mammalian cells upon stimulation. Several mechanisms have been proposed for active demethylation.<sup>23</sup> These include: (i) active demethylation through proteins removing methylcytosines by DNA repair mechanisms. This includes thymidine DNA glycosylase (TDG), methyl-CpG-binding domain protein

4 (MBD4), and the AID/APOBEC (activation-induced deaminase/apolipoprotein B mRNA editing enzyme, catalytic polypeptide-like) family; (ii) Active demethylation through proteins that interact with methylcytosine by DNA modifications. Oxidation of methylcytosine leads to the formation of hydroxymethylcytosine, an epigenetic mark that is most common in neuronal tissues and embryonic stem cells. Hydroxymethylcytosine is considered an intermediate step in DNA demethylation. Oxidation of methylcytosine to hydroxymethylcytosine is mediated by the ten-eleven translocation proteins (ten-eleven translocation [TET] proteins: TET1, TET2, and TET3).<sup>23</sup> Active demethylation through proteins that indirectly interact with methylcytosine binding of transcription factors has been shown to be involved in the active demethylation of DNA.<sup>24</sup>

### Epigenetic effects of environmental exposure and disease status

Exposure to stressful or traumatic life events, especially early in life, is one of the strongest risk factors for the development of several psychiatric disorders, including depression. Epigenetic modifications are candidate mechanisms for environmental stress exposure-induced effects on gene expression and long-term effects on neuronal functioning, and thus disease risk.<sup>13</sup> Several studies have shown that early adverse environment leaves long lasting epigenetic marks via changes in both DNA methylation and histone modifications.<sup>11,25-29</sup> Interestingly, these marks can in some instances be observed in DNA from peripheral tissue (blood or epithelial cells), as well as in brain or neuronal cells.<sup>30-33</sup> So while most of these epigenetic changes may be tissue specific, there is evidence for overlap.<sup>30-33</sup> For example, early life trauma-associated changes in DNA methylation in the promoter region of the gene encoding the glucocorticoid receptor (GR), *NR3C1* are seen at overlapping CpG sites in brain tissue and peripheral blood cells.<sup>32,34-36</sup> This region is the promoter region of the alternate exon 1F (humans) or 1<sub>7</sub> (rodents) and associated with a binding site of the nerve growth factor (NGF)1A transcription factor.<sup>37,38</sup> Binding of this transcription factor has been shown to be essential to both induce the observed changes in DNA methylation and mediate the gene expression changes associated with the changes in DNA methylation.<sup>37,38</sup> Our group has shown that the same specific CpGs located in a gluco-

corticoid response element of the GR-regulating gene *FKBP5* were demethylated in peripheral blood cells of individuals exposed to child abuse, as well as in a neuronal progenitor cell line after exposure to GR agonists.<sup>31</sup> These changes appeared sensitive to the timing of the GR activation. At least in the neuronal progenitor cell line, DNA methylation effects of treatment during late proliferation and early differentiation could be seen even after 3 weeks without GR stimulation. However, when the cell line was treated after differentiation, no effect on methylation on these sites was observed. This fits well with the fact that the gene x environment with *FKBP5* is only observed with trauma experienced during childhood, but not later in life.<sup>39</sup> Using a rhesus monkey model of early adversity, Provencal et al demonstrated that long-lasting changes in DNA methylation were seen both in T cells and in DNA from the frontal cortex.<sup>30</sup> However, there was little direct overlap of the differentially methylated CpG.

In studies investigating case/control effects on DNA methylation, it is thus of paramount importance to also control for environmental exposure, especially early adverse environment. Extrapolations of changes observed in one tissue to others should be made only cautiously. It is also not clear to date whether environmentally sensitive marks are identical with methylation differences observed in case/control studies. For the environmentally sensitive sites in the *NR3C1* locus for example, consistent evidence has been found in brain and peripheral blood for exposure to child abuse<sup>32,33,35</sup> or other early adverse experience<sup>40</sup>; effects related to disease status are less consistent. For example, Alt et al did not find GR expression differences due to epigenetic programming of GR promoters in postmortem brain tissue of depressed patients not documented to be exposed to child abuse.<sup>41</sup> In the studies investigating postmortem hippocampal samples of suicide completers that have reported increased methylation of *NR3C1* associated with childhood abuse, not all suicide completers had suffered from major depression.<sup>32,34,35</sup>

Other studies have focused on the monoaminergic transporter genes, most often the gene encoding the serotonin transporter gene *SLC6A4*. Changes in methylation in the promoter of this gene have been reported with both environmental factors as well as depression in peripheral tissue; however, the direction of the effects is not always congruent. A study enrolling shiftworking nurses found that environmental stress is concurrent

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with decreased methylation of the *SLC6A4* promoter, and after adjustment for stress and symptom severity, environmental stress as well as job related exhaustion/burnout significantly contributed to lower *SLC6A4* methylation levels.<sup>42</sup> In a sample of depressed patients, however, higher methylation status of the promoter of gene encoding serotonin transporter (*SLC6A4*) was associated with childhood adversities and worse clinical presentation, but not with treatment response.<sup>43</sup> A similar direction was found in a study with monozygotic twins, which revealed that *SLC6A4* promoter methylation levels were positively correlated with depressive symptoms.<sup>44</sup> A study on poststroke depression (PSD) observed a higher *SLC6A4* promoter methylation status in patients suffering from PSD already 2 weeks after stroke, and this was also associated with worsening of depressive symptoms over 1 year.<sup>45</sup> However, in this study different CpG sites were examined, as in the study by Zhao et al.<sup>44</sup> To resolve issues of directionality, it is important to consider the specific CpGs affected. While increased methylation has traditionally been associated with decreased gene expression, this seems to be restricted to certain promoter elements. In regulatory regions, the directionality depends on whether enhancers or repressors bind to the affected sequence. Since DNA methylation decreases binding of transcriptional regulators<sup>19,46,47</sup> this can lead to both increased and decreased gene expression.<sup>46-48</sup>

The third gene that has been repeatedly investigated for epigenetic changes in depression is the gene encoding brain-derived neurotrophic factor, *BDNF*. *BDNF* belongs to the neurotrophins, which are involved in proliferation, migration, differentiation, and survival of neurons in the human central nervous system,<sup>49</sup> and has been consistently associated with depression and treatment response to antidepressants.<sup>50</sup> Several studies observed a reduced *BDNF* mRNA expression in peripheral blood of acutely depressed patients, which was reversed by successful antidepressant treatment.<sup>51</sup> A study enrolling 108 depressed patients observed a higher *BDNF* promoter methylation status associated with previous suicidal attempt history and suicidal ideation during treatment.<sup>52</sup> This direction is similar to the one observed in postmortem brain samples from suicide subjects, where an increase of DNA methylation at CpG sites in *BDNF* promoter compared with nonsuicide control subjects were found.<sup>53</sup> However, another study investigating peripheral blood DNA meth-

ylation profiles within CpG islands at the promoter of the *BDNF* gene found a reduction in methylation in depressed patients compared with healthy controls.<sup>54</sup>

Interestingly, in patients suffering from borderline personality disorder a higher methylation status of CpG sites within the *BDNF* gene was associated with a higher number of childhood traumatic experiences, and the methylation could be decreased by successful intensive dialectical behavior therapy.<sup>55</sup>

Overall, these studies suggest that genes carrying epigenetic changes associated with environmental risk factors for major depression are often also affected with disease, although not always at the exact same CpG sites or in the same direction. At least for some specific changes, similar effects are observed in peripheral tissue and the brain. Environmental and genetic risk factors may thus impinge on epigenetic regulation genes within systems that have been shown to be dysregulated in major depression, such as the stress hormone system, the monoamine system, and the neurotrophic system. These long-term epigenetic effects could be the underlying cause of the observed dysregulations of these systems, as epigenetic changes translate in mRNA and protein expression changes, and dysfunctions of circuits dependent on these genes. Animal studies have shown that stress-related epigenetic changes may be reversible, although this has, so far, not been shown in humans.<sup>38,56</sup>

## Epigenetics and antidepressants

In addition to disease risk, such epigenetic mechanisms may also moderate or even mediate therapeutic effects of antidepressant drugs. Several facets of this relationship have been investigated.

### Currently used antidepressants drugs may exert direct epigenetic effects

The tricyclic antidepressants amitriptyline and imipramine, but also the SSRI paroxetine, were shown to reduce promoter DNA methylation in rat primary astrocytes, possibly by reducing DNA methyltransferase 1 activity.<sup>57,58</sup> The mood stabilizer and anticonvulsant valproate may induce a global reduction in DNA methylation levels, which was observed in human embryonal kidney cells and in rat hepatic cells.<sup>59,60</sup> Additionally, the antipsychotics sulpiride and clozapine could activate DNA demethylation in the frontal cortex and striatum.<sup>61</sup>

### **Effective antidepressant treatment may be associated with epigenetic changes in genes conferring risk for depression**

It has been repeatedly suggested that effective treatment with antidepressants increases peripheral BDNF levels,<sup>62</sup> and that a nonincrease of BDNF plasma levels within the first week of treatment could predict treatment resistance with high sensitivity.<sup>63</sup> Hence, the methylation levels of 13 CpG sites within the BDNF exon IV promoter in a sample of depressed patients treated with several antidepressants in a naturalistic setting were investigated.<sup>64</sup> A lower methylation was observed in the final responders, without a significant interaction of gender or class of antidepressant. This was paralleled by a decrease of BDNF plasma levels during the first week of treatment.<sup>64</sup> Additionally, Lopez et al investigated the impact of 8 weeks of citalopram treatment on BDNF expression in initially treatment-naïve depressed patients.<sup>65</sup> Treatment responders displayed increased BDNF mRNA levels with a significant correlation between change in depression severity and change in BDNF expression, and also a significant decrease in histone H3 lysine 27 trimethylation (H3K27me3) levels (a marker for silencing genes) at BDNF exon IV promoter, which was negatively correlated with change in depression severity. Also, BDNF expression and H3K27me3 levels were significantly negatively correlated.<sup>65</sup> In a cohort of combat veterans suffering from post-traumatic stress disorder (PTSD), a higher methylation of the *NR3C1* exon 1F promoter assessed before prolonged exposure psychotherapy was associated with successful treatment outcome,<sup>66</sup> in line with the findings of increased sensitivity of the GR being associated with PTSD.<sup>67</sup> DNA methylation of one of the main targets of antidepressant drug action, the serotonin transporter, was shown to predict treatment outcome. Domschke et al observed a higher methylation status of the *SLC6A4* gene in depressed patients who showed a better response following 6 weeks' treatment with escitalopram.<sup>68</sup>

### **Epigenetic factors interacting with genetic predictors of treatment response**

A study linking a single-nucleotide polymorphism (SNP) (rs1126757) in the interleukin-11 (*IL11*) gene to treatment response prediction following treatment

with the selective serotonin reuptake inhibitor escitalopram<sup>69</sup> gave rise to a DNA methylation analysis across the only CpG island located in the *IL11* gene. Within the Genome-Based Therapeutic Drugs for Depression (GENDEP) project where depressed patients are treated with either escitalopram or nortriptyline,<sup>70</sup> the Sequenom EpiTYPER platform was used to assess DNA methylation of *IL11* in peripheral blood. In fact, CpG methylation of this gene predicted overall antidepressant response as well as differential response to the two antidepressants. Methylation levels of the CpG sites 21 to 22 predicted overall response; namely lower methylation levels were observed in patients with better response. A differential response to antidepressants could be predicted by methylation levels of the CpG sites 19 to 20; higher methylation levels were associated with better response in subjects taking escitalopram, but were associated with worse response in those taking nortriptyline. Finally, a SNP, rs1126757 by CpG site interaction predicting antidepressant response was observed,<sup>71</sup> suggesting that the SNP effect may be moderated by additional epigenetic factors.

### **Using drugs interfering with epigenetic mechanisms as a potential antidepressant**

Dietary levels of methyl donor components such as homocysteine and folic acid may impact DNA methylation levels. Increased DNA methylation and altered methylation-dependent phenotypes have been observed in offspring of mothers taking dietary supplements of folic acid during pregnancy.<sup>72</sup> Furthermore, individuals with folate deficiency are more likely to develop depression<sup>73</sup> and less likely to respond to antidepressant drugs.<sup>74</sup> Interestingly, treatment with folate has been shown to reduce depressive symptoms.<sup>75,76</sup> Reduced folate levels were also associated with decreased leukocyte DNA methylation and increased plasma homocysteine levels in women.<sup>77,78</sup> Homocysteine is metabolized to S-adenosyl-methionine, a methyl donor, which might influence DNA methylation. Several studies observed that homocysteine impacts global gene promoter DNA methylation, and that administration of homocysteine induces a transient demethylation of the promoter DNA of Herp (homocysteine-induced endoplasmic reticulum protein, an endoplasmic reticulum-resident membrane protein) with an enhancement of Herp gene expression and a subsequent global increase in DNA methylation in

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human embryonic kidney cells and human neuroblastoma.<sup>79</sup> This mechanism may be important for DNA methylation of the BDNF locus. In fact, BDNF levels in the hippocampus were reduced in an animal model following homocysteine treatment, which was paralleled by impaired memory consolidation.<sup>80</sup> In addition, a reduction of BDNF levels by homocysteine was prevented by treatment with folic acid,<sup>80</sup> which increases DNA methylation.<sup>81</sup> Indeed, elevated homocysteine levels have been associated with major depression.<sup>82,83</sup> These data further support the fact that clinical response to antidepressant drugs, as well as risk for depression, are associated with dynamic changes in DNA methylation, so that factors promoting or interfering with this system also moderate risk or treatment response.

## Epigenome-wide association studies

Epigenome-wide association studies (EWAS), aimed at detecting DNA methylation differences associated with disease predisposition or progression, are underway for many complex disease phenotypes.<sup>84</sup> In contrast to candidate approaches, EWAS hold promise for the detection of new regulatory mechanisms and candidate pathways relevant for disease.<sup>85</sup> Epigenomic profiling is now being assessed to improve clinical diagnosis and subtyping of disease.<sup>86</sup> So far, several epigenetic alterations have been identified to allow personalized medicine, especially in cancer. Epigenetic profiling has been used in disease diagnosis, disease prognosis, therapy response, and tumor type identification from cancers of unknown primary origin.<sup>86</sup> A promising example was the identification of the *O*<sup>6</sup>-methylguanine-DNA methyltransferase (MGMT), which protects normal cells against mutations by removing alkyl groups.<sup>87</sup> A hypermethylated MGMT leads to a blocked gene expression, resulting in an accumulation of alkylated guanines and subsequent DNA damage. A hypermethylated and thus inactive MGMT in glioblastomas has been found to be a predictor of successful response to DNA-alkylating drugs as carmustine and temozolomide, because an active MGMT is able to repair altered bases introduced by the therapy.<sup>87</sup> However, in psychiatry such achievements of clinically applied personalized medicine are still lacking. Complexity of psychiatric phenotypes, as well as a number of technical issues, especially lack of access to the target tissue, the brain, make it more difficult to separate noise from signal than in cancer tissue.<sup>85</sup>

## Technical aspects

New powerful technologies have emerged that enable a hypothesis-free approach to investigate DNA methylation at the genome-wide level. The first methods relied on affinity-based methods such as methylated DNA binding domain sequencing (MBD-seq) and methylation DNA immunoprecipitation sequencing (MeDIP-seq), in which methylated regions are enriched by binding to either methyl binding proteins or antibodies against methylated DNA.<sup>88</sup> This strategy is most useful to identify large differences in methylation. Its disadvantage is the lack of single CpG resolution. The restriction enzyme-based comprehensive high-throughput arrays for relative methylation (CHARM) platform employs the methylation-dependent specificity of the methylation-sensitive restriction enzyme McrBC with subsequent analysis on tiling arrays<sup>89</sup>; however, this technique also does not give single CpG resolution. For this, bisulfite-conversion needs to be used, where unmethylated cytosines are converted into thymidines while methylated CpGs are protected, leading to a T/C difference that can be detected using several methods. One group are array-based techniques such as the human Methylation 450 Beadchip Assay provided by Illumina. This assay enables a high resolution genome-wide DNA methylation profiling of human samples, covering 99% of all RefSeq genes and approximately 450 000 CpGs overall.<sup>90</sup> The other method is next-generation sequencing for whole methylome representation. This approach is very costly, so that methods reducing the sequence are often used. One is reduced representation bisulfite sequencing (RRBS) where pretreatment with the methylation-insensitive restriction enzyme (MspI) reduces the number of noninformative sequences by DNA digestion. Such sequence reduction strategies are warranted, as over 70% of the methylome are invariant to tissue, environmental, and developmental factors, and thus likely of little relevance for disease risk.<sup>91</sup>

## EWAS in major depression

One of the first genome-wide DNA methylation scans in major depression investigated postmortem frontal cortex tissue of 39 depressed patients and 26 controls using the CHARM platform.<sup>92</sup> With a threshold of >10% DNA methylation differences 224 candidate regions

were identified, with an enrichment for neuronal growth and developmental genes. The greatest DNA methylation differences were observed in proline rich membrane anchor 1 (PRIMA1), together with a concomitant decrease in gene expression.<sup>92</sup> Another EWAS study enrolling 33 subjects with lifetime history of depression and 67 nondepressed controls using the HumanMethylation27 (HM27) DNA Analysis Illumina BeadChip revealed uniquely unmethylated gene sets derived from whole-blood DNA samples distinguishing between those with vs without lifetime depression.<sup>93</sup> Interestingly, IL-6 and C-reactive protein (CRP) plasma levels were increased in those persons with lifetime depression, and among those with depression only, IL-6 methylation showed an inverse correlation with plasma IL-6 and CRP.<sup>93</sup> However, these findings have to be interpreted very cautiously because of the small sample size.

Labonte et al observed 362 differentially methylated promoters in hippocampal samples of individuals with a history of abuse compared with controls using a custom-designed 400K promoter tiling array by Agilent.<sup>34</sup> Within these regions genes involved in cellular and neuronal plasticity were among the most significantly differentially methylated.<sup>34</sup> The same group of investigators found 366 promoters differentially methylated in suicide completers compared with control subjects, with 273 hypermethylated and 93 hypomethylated.<sup>94</sup> Promoter methylation differences were inversely correlated with gene expression differences, with higher methylation associated with lower gene expression as expected.<sup>94</sup> However, other relationships can be observed, depending on the location of the methylation sites and methylation in enhancer or repression regions can have both positive as well as negative correlations with gene expression.<sup>48,95</sup>

To uncover brain cell type-dependent DNA methylation patterns, Guintivano et al combined fluorescence activated cell sorting (FACS) of neural nuclei with Illumina HM450 DNA methylation profiling in postmortem frontal cortex of depressed patients and matched controls.<sup>96</sup> An overlap of depression-associated DNA methylation changes and DNA methylation signature from the different cell types suggested that the depression-associated changes were enriched among neuronal cells.<sup>96</sup> This approach also highlights the importance to control for differences in cell types in the analysed tissue to decrease confounding due to different tissue proportions. Methods to generate tissue proportion from

DNA methylation data using bioinformatic strategies have been developed for peripheral blood and its cell types<sup>97</sup> as well as for brain, where the ratio of neurons vs glia can be estimated.<sup>96</sup> EWAS need to consider such tissue effects and either control for them or preselect certain cell types using cell sorting.

While the first EWAS for major depression have yielded some interesting findings, both in peripheral blood as well as brain samples, they remain largely unreplicated. In general, genome-wide methylation studies in psychiatry have so far likely been underpowered with the small effect sizes observed and expected for more subtle influences on gene regulation than the ones observed in cancer. Similar approaches, such as the ones used in GWAS studies, with the formation of large consortia and meta-analysis will be important, but are methodologically more challenging for DNA methylation than for genotype analysis.

To our knowledge no epigenome-wide study on response to treatment with antidepressants has yet been published. As epigenetic measures represent both genetic and environmental influences, epigenetic profiling may provide a better understanding of antidepressant treatment effect and could be a more robust biomarker than either transcriptome or GWAS analyses by themselves.<sup>98</sup> Studies on epigenetic alterations in cancer research were able to identify new drugs eliciting epigenetic effects, such as DNA methylation inhibition. For example, the DNMT inhibitors 5-azacytidine (Vidaza) and 5-aza-2-deoxycytidine (Dacogen) were both approved by the US Food and Drug Administration for treatment of myelodysplastic syndrome.<sup>99</sup> Systemic administration of these DNMT inhibitors induced a dose-dependent antidepressant-like effect in an animal model for depressive behavior,<sup>100</sup> but clinical studies on major depression are still lacking.

## Conclusions

Epigenetics may provide new insight into pathophysiology of major depression and may yield novel biomarkers for diagnosis and treatment response. Reflecting environmental factors as well as genetic contribution to the development of major depression, epigenetic mechanisms may help in dissecting the complex phenotype of major depression. While the impact of early life stress on epigenetic alterations has been clearly demonstrated,<sup>13,101</sup> it is not yet clear how and if they translate to risk for

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major depression for all of the investigated candidates. Interestingly, some of these same candidates also show epigenetic changes with antidepressant treatment, and antidepressant drugs may directly interfere with specific epigenetic mechanisms. Targeting selective epigenetic mechanisms is being considered as a possible new avenue for the development of novel antidepressant drugs.

In addition, some studies using epigenome-wide approaches have been published, and several methylation differences have been identified in postmortem brain tissue and peripheral blood cells between depressed patients and healthy controls. Albeit promising, these results need to be confirmed by independent replication and in larger samples. □

## **Alteraciones epigenéticas en la depresión y el tratamiento antidepresivo**

*Las modificaciones epigenéticas controlan la estructura y la función de la cromatina, y de ese modo median los cambios en la expresión génica, influyendo finalmente en los niveles de proteína. La investigación reciente señala que los acontecimientos ambientales pueden inducir cambios epigenéticos y a través de éstos contribuir a cambios a largo plazo en los circuitos neurales y los sistemas endocrinos asociados con un riesgo modificado para trastornos psiquiátricos relacionados con el estrés como la depresión mayor. En esta revisión se describen los enfoques recientes que investigan las modificaciones epigenéticas asociadas con el riesgo modificado para la depresión mayor o la respuesta a fármacos antidepresivos, tanto a nivel de los genes candidato como del genoma completo. Este artículo se concentra en la metilación del ADN, ya que este es el cambio epigenético más estudiado en la investigación en depresión.*

## **Modifications épigénétiques au cours de la dépression et d'un traitement par antidépresseurs**

*Les modifications épigénétiques, en contrôlant la structure et la fonction de la chromatine, interviennent dans les changements de l'expression génique, et influent finalement sur les taux des protéines. D'après des recherches récentes, des événements liés à l'environnement peuvent provoquer des modifications épigénétiques et, de ce fait, induire des changements à long terme dans des circuits neuronaux et les systèmes endocriniens qui sont associés à un risque modifié de troubles psychiatriques liés au stress comme une dépression caractérisée (majeure). Nous décrivons dans cet article les études récentes portant sur les modifications épigénétiques associées à un risque modifié de dépression majeure ou de réponse à un traitement antidépresseur, à la fois au niveau des gènes candidats et à celui de l'ensemble du génome. Nous nous intéressons particulièrement à la méthylation de l'ADN, modification épigénétique la plus étudiée dans la recherche sur la dépression.*

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