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Antidepressant action: to the nucleus and beyond

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After decades of effort, the field of depression research is far from understanding how antidepressant drugs mediate their clinical effects. The time lag of 2–6 weeks of therapy that is necessary to obtain antidepressant efficacy indicates a requirement for long-term regulation of molecules activated by drug treatment. The focus of antidepressant research has thus expanded from examining acute monoamine-mediated mechanisms to include long-term transcriptional regulators such as cAMP response element-binding protein (CREB) and trophic factors such as brain-derived nerve growth factor and insulin-like growth factor. In addition, the recent discovery of antidepressant-induced neurogenesis provides another avenue by which antidepressants might exert their effects. Current efforts are aimed at understanding how CREB and trophic factor signaling pathways are coupled to neurogenic effects and how alterations in behavioral, molecular and cellular endpoints are related to the alleviation of the symptoms of depression.

What is the mechanism of action of antidepressant drugs?

Depression is a clinically and biologically heterogeneous disease. It is one of the most prevalent and costly psychiatric disorders worldwide, with 10–30% of women and 7–15% of men likely to suffer from depression in their life-time [1]. Despite the prevalence and societal cost of depression of an estimated US$50 billion [2], currently used antidepressants do not improve symptoms in all patients. This is, at least in part, the consequence of our limited understanding of the mechanisms of antidepressant action and the pathophysiology of depression. Treatment for depression requires administration of antidepressant drugs for 2–6 weeks before clinical efficacy is observed. This time lag suggests that long-term adaptations in neurotransmitter systems and/or their downstream targets might be necessary for therapeutic effects. However, a novel hypothesis is emerging that is changing the way we think about both the pathology of depression and the mechanisms that underlie antidepressant drug action. This hypothesis postulates that antidepressants activate not only second messenger systems leading to the activation of transcription factors such as cAMP response element-binding protein (CREB) but also activate neurotrophic pathways and increase hippocampal neurogenesis.

Multiple classes of antidepressant drugs, including monoamine oxidase inhibitors (MAOIs), selective serotonin reuptake inhibitors (SSRIs) and selective noradrenaline reuptake inhibitors (SNRIs), increase both cell proliferation and neurogenesis in the dentate gyrus of the adult hippocampus [3,4]. These cellular effects occur only after chronic, and not acute, dosing, which corresponds with the therapeutic time-course for clinical efficacy of antidepressants. The challenge of pharmacological studies and animal models is to elucidate the mechanism of antidepressant action beyond neurotransmitters to identify specific intracellular second messenger pathways, determine how they impact on neurogenesis and evaluate the behavioral consequences of these cellular modifications.

Antidepressant drugs: beyond neurotransmitters to activation of second messenger pathways and transcriptional regulators

The monoamine hypothesis of depression postulates that a functional deficiency of 5-hydroxytryptamine [5-HT (serotonin)] or noradrenaline in the brain is key to the pathology and/or behavioral manifestations associated with depression [5,6]. In support of this theory, the majority of antidepressant drugs used clinically produce acute increases in the levels of 5-HT and noradrenaline. This in turn causes the activation of seven-transmembrane domain receptors that are coupled to heterotrimeric G proteins. Through G-protein activation of adenylyl cyclase, cAMP production is increased, enabling the activation of cAMP-dependent protein kinase (PKA) and phosphorylation of target proteins. Regulation of G proteins, at the level of enhanced coupling of G_s to adenylyl cyclase and increased adenylyl cyclase activity, occurs following antidepressant treatment [7,8]. In addition, PKA activity is increased following chronic treatment of rats with either a tricyclic antidepressant (imipramine), an MAOI (tranylcypromine) or electroconvulsive shock (ECS) [9].

Additional evidence implicating the cAMP–PKA cascade in the mechanism of action of antidepressants comes from studies employing rolipram, a compound that inhibits the high-affinity cAMP-selective

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phosphodiesterase type 4 (PDE4). Rolipram activates the cAMP–PKA cascade [10] and chronic but not acute treatment with rolipram produces antidepressant effects in multiple behavioral tests in rats and mice [11,12]. In addition, certain isoforms of PDE4 (PDE4A and 4B) are upregulated after chronic antidepressant treatment [13], again suggesting that increased intracellular levels of cAMP are one of the consequences of antidepressant treatment, and that long-term regulation of this pathway is necessary to observe clinical efficacy.

Although most antidepressant drugs increase intracellular levels of cAMP through activation of adrenoceptors or 5-HT receptors, it is important to note that not all subtypes of these receptors are coupled to the adenylyl cyclase–cAMP–PKA pathway. For example, activation of phospholipase C (PLC) by α_1-adrenoceptors can lead to mobilization of internal Ca^{2+} stores and subsequent activation of Ca^{2+}–calmodulin (CaM)-dependent kinases. Indeed, several antidepressants, including fluoxetine, desipramine and reboxetine, markedly increase the enzymatic activity of Ca^{2+}–CaM kinase II and IV in the prefrontal cortex of rats [14,15].

Activation of protein kinases in the cell thus enables phosphorylation of downstream effectors. One of the best-characterized targets for phosphorylation by a variety of kinases is the transcription factor CREB [16] (Figure 1). PKA and CaM kinase catalyze the transfer of phosphate from ATP to specific serine residues on this protein substrate. Activation of PKA leads to phosphorylation of a serine residue (S133) in the CREB protein that enables recruitment of co-activator proteins to initiate gene transcription [17]. CREB is also a substrate for Ca^{2+}–CaM kinase II and IV, which activate or inhibit CREB transactivation depending on the serine residues that are phosphorylated [18,19]. In addition, mitogen, signaling through the Ras–mitogen-activated protein (MAP) kinase kinase (MEK)–extracellular signal-regulated protein kinase (ERK) pathway, can phosphorylate CREB via RSK2, a member of the ribosomal S6 kinase family [20], and at least one study has reported changes in downstream components of the MEK–ERK–RSK2-mediated signaling cascade following chronic antidepressant treatment [15].

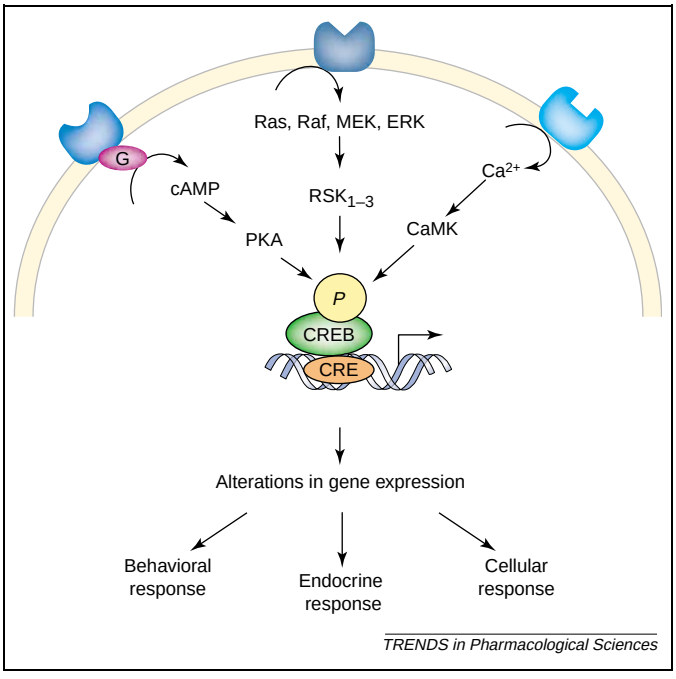

Figure 1. Regulation of cAMP response element-binding protein (CREB) phosphorylation by antidepressant drugs. Most clinically effective antidepressants alter noradrenaline or 5-HT neurotransmitter levels by a variety of mechanisms. Cell-surface receptors can respond to these neurotransmitters by altering intracellular second messengers, such as cAMP and Ca^{2+}, in addition to several kinases, such as cAMP-dependent protein kinase (PKA), Ca^{2+}–CaM-dependent kinases (CaMK), mitogen-activated protein kinase kinase (MEK), extracellular signal-regulated protein kinase (ERK) and several forms of ribosomal S6 kinase (RSK_{1-3}). Kinases phosphorylate protein substrates such as the transcription factor CREB. CREB binds to a cAMP responsive element (CRE) in DNA to regulate gene expression. These CREB-target genes might ultimately modulate behavior, endocrine or cellular changes associated with chronic antidepressant drug treatment.

The ability of antidepressants to activate protein kinases in the cell and the potential for these kinases to phosphorylate CREB suggest that antidepressants can activate CREB as part of their mechanism of action. Indeed, chronic administration of fluoxetine to rats increases phosphorylated CREB (P-CREB) levels in several brain regions, including the amygdala, cortex, dentate gyrus and hypothalamus [21]. Interestingly, desipramine increases the levels of P-CREB only in the dentate gyrus [21]. In a separate study, chronic treatment with the SSRI fluoxetine increased P-CREB levels in the prefrontal and frontal cortex to a greater extent than the selective noradrenaline reuptake inhibitors desipramine and reboxetine [15]. Moreover, activation of the Ca^{2+}–CaM kinase IV and MAP kinase cascades contributed more to this increase in P-CREB levels than did activation of the cAMP–PKA pathway. The selectivity of antidepressant drugs at reuptake sites throughout the brain does not vary, however, depending on which postsynaptic receptors and associated signal transduction pathways are activated in various brain regions, the subsequent effects on CREB activity will be different.

Genetic models of CREB activity

Because antidepressant drugs can activate CREB, recent studies have examined whether manipulation of CREB activity and/or levels of CREB can recapitulate some effects of antidepressants. To date, the function of CREB has been investigated using animal models or viral-vector-mediated gene overexpression (Table 1). Given that the majority of studies have reported increased CREB and/or CREB phosphorylation after chronic antidepressant treatment, the hypothesis of many of these studies was that alterations in CREB would have effects on baseline measures in antidepressant tests. Unfortunately, to date, a clear picture has not emerged as a result of these *in vivo* studies because the outcomes vary depending on the spatial and temporal expression of CREB. For example, acute overexpression of CREB in the dentate gyrus in rats induced antidepressant-like responses in the learned helplessness paradigm and in the forced swim test (FST) [22]. Learned helplessness provides a model for the psychological aspect of depression because prior exposure to an inescapable shock in animals interferes with their ability to learn in a new situation where escape is possible, whereas the FST is a standard paradigm used to evaluate antidepressant drug efficacy. Overexpression of CREB in the basolateral amygdala using a similar viral gene

Table 1. Animal models for behavioral analysis of CREB function

Approach[a]	Effect	Brain region	Phenotype	Refs
Viral expression HSV-CREBα	Gain of function	Hippocampus, dentate gyrus	Antidepressant	[22]
Viral expression HSV-CREBα	Gain of function	Basolateral amygdala	Antidepressant, pro-depressant[b]	[23]
Viral expression HSV-CREBα	Gain of function	Nucleus accumbens	Pro-depressant	[24]
Viral expression HSV-mCREB	Loss of function	Nucleus accumbens	Antidepressant	[24,25]
Transgenic NSE-tTA/TetOp CREBα	Gain of function	Forebrain[b]	Pro-depressant	[25]
Transgenic NSE-tTA/TetOp mCREB	Loss of function	Forebrain[c]	Antidepressant	[25]
Gene ablation	Loss of function	Global	Antidepressant	[28]

[a]Abbreviations: HSV, herpes simplex virus; mCREB, a dominant mutant form of CREB; NSE, neuron-specific enolase; tTA, tetracycline transactivator; TetOp, tetracycline operon.
[b]Temporal expression of CREB determines the behavioral phenotype; expression of CREB before training is associated with a pro-depressant phenotype, whereas expression after training is associated with an antidepressant phenotype.
[c]Expression included parts of the striatum (including dorsal striatum and the nucleus accumbens) and certain aspects of cerebral cortex and subfields of the hippocampus.

transfer approach produced an antidepressant-like effect when expression was increased after training in the learned helplessness model of depression [23]. However, when CREB expression was elevated in the nucleus accumbens, a pro-depressive response was observed [24]. Similarly, overexpression of CREB in the basolateral amygdala produced a pro-depressant effect but only when expression was induced before training in learned helplessness [23]. Lastly, overexpression of CREB in a transgenic mouse line resulted in depressive-like behaviors. However, because CREB overexpression was not localized to specific brain structures in these mice, it is difficult to identify the neuroanatomical substrate of this effect [25]. Together, these data highlight the regional and temporal importance of CREB activation in mediating behavioral responses associated with antidepressant drug treatment.

Several studies have demonstrated that viral expression of a dominant mutant form of CREB, mCREB, produces an antidepressant-like effect when infused into the nucleus accumbens of mice [24], and transgenic expression of mCREB in mice also produces an antidepressant-like effect [25]. These results are similar to those obtained in a CREB-deficient mouse model. This CREB-deficient mouse was generated by targeted deletion of two major isoforms of the gene encoding CREB by homologous recombination that resulted in a global reduction of CREB by ~90%, compared with wild-type mice [26,27]. These mice demonstrate an antidepressant-like phenotype in three different behavior tests: the FST, tail suspension test (TST) and learned helplessness [28]. In addition, the antidepressant efficacy of desipramine and fluoxetine is maintained in CREB-deficient mice at the behavioral and endocrine level [28] despite the significant reduction of CREB protein levels throughout the brains of these animals. These data suggest that CREB might not be required for acute antidepressant efficacy as it is assessed in current behavioral paradigms. Few behavioral tests exist to evaluate chronic antidepressant efficacy and the role of CREB in mediating long-term effects of antidepressant drugs has not yet been discerned.

Antidepressant drugs, neurotrophins and growth factors

Brain-derived nerve growth factor

Plasticity in the nervous system is subserved by a variety of neurotrophins and growth factors. One well-characterized neurotrophic factor involved in activity-dependent neuronal plasticity, survival and differentiation of peripheral and central neurons is brain-derived nerve growth factor (BDNF) [29–31]. Several studies suggest that BDNF is a target of antidepressant action. Robust increases in the levels of BDNF mRNA in cortical and hippocampal regions have been reported following chronic antidepressant drug administration in rats [32,33]; however, other studies indicate that this is not common to all antidepressant drugs. For example, although tranylcypromine (a MAOI) and ECS increase BDNF mRNA levels [34–36], the more-selective antidepressants such as desipramine and fluoxetine have variable effects [32,34,35,37]. Alterations in BDNF protein levels have been reported only in a few studies [36,38] with tranylcypromine and ECS increasing protein levels but fluoxetine and desipramine having no effect [38]. BDNF has also been shown to be regulated by exposure to stress [39], and antidepressant treatment can block this downregulation [32,40]. Furthermore, the duration of drug treatment and interval following drug administration can have an impact on the overall levels of BDNF [34]. Thus, differences in stress states of animals in various studies, in addition to the duration of treatment and when BDNF levels are analyzed following treatment, might contribute to these conflicting reports.

BDNF binds to the trkB receptor in the brain. Upon activation by ligand-dependent autophosphorylation, this tyrosine kinase initiates a variety of intracellular signaling cascades including the MEK–ERK pathway and downstream activation of RSK2, which can phosphorylate CREB. In addition, the promoter region of exon 3 in the gene encoding BDNF contains CRE elements, identifying it as a potential CREB-target gene [41]. This information, combined with the temporal and spatial correlation of upregulation of BDNF and CREB activation following antidepressant treatment, suggest that enhanced CREB expression might lead to an upregulation of BDNF; however, no studies to date have determined whether this is a direct mechanism. A link between CREB and BDNF is strongly suggested by the finding that antidepressant-mediated upregulation of BDNF is blocked in CREB-deficient mice [28]. Thus, although CREB does not appear to mediate acute behavioral or endocrine effects of antidepressant drugs, endpoints that rely on chronic drug

administration, such as changes in gene expression, are reduced when CREB function is impaired.

BNDF is not only a putative target of antidepressant action but BDNF itself produces antidepressant-like effects and might thus be one of the molecular mediators of antidepressant drugs. For example, central administration of BDNF into the ventricles, hippocampus or midbrain of rats [42] has antidepressant-like effects on multiple models of depression [42,43]. One recent study has reported that after BDNF infusion into the ventral tegmental area and nucleus accumbens, depressive-like behaviors are observed [44]. However, the majority of studies indicates that BDNF infusion and, analogously, trkB activation produces a positive effect in the brain. In addition to its direct antidepressant effects, BDNF infusion also induces neurogenesis [45], which can directly or indirectly contribute to its antidepressant action in the long term. Further investigation is necessary to determine if a causal relationship exists between increased CREB activation and BDNF expression on the one hand, and hippocampal neurogenesis and antidepressant behavioral effects on the other hand.

Genetic models of BDNF action

Mice that lack BDNF display severe neuronal deficits and early postnatal death [31]. However, studies examining mice that lack only one allele of the gene encoding BDNF ($BDNF^{+/-}$) have identified alterations in learning [46] and synaptic plasticity [47,48]. Some lines of $BDNF^{+/-}$ mice display a reduction in BDNF protein levels but without any accompanying changes in baseline behavior in models such as the FST [49]. Other lines of $BDNF^{+/-}$ mice display altered synaptic responses, but a specific behavioral phenotype associated with these alterations has not been identified [48]. These data reveal that a partial loss of BDNF is not sufficient to affect baseline behavior but this interpretation is compromised by the fact that these animals had a loss of BDNF from birth and might have evolved compensatory responses as adults. However, both $trkB^{+/-}$ and $BDNF^{+/-}$ mice have been reported to be resistant to antidepressants in the FST [50].

The use of an inducible knockout system was employed recently to delete BDNF specifically in the forebrain. Although these mutant mice did not demonstrate a depressive-like phenotype that might have been predicted with the loss of BDNF, they did exhibit an attenuated response to antidepressant administration in the FST [51]. By extension, mice that lack the BDNF receptor trkB in a forebrain-specific deletion demonstrate antidepressant behaviors in the FST; however, specific responses to antidepressant drugs were not tested in these mice [52]. Taken together, these studies indicate that BDNF is intricately involved in depression and antidepressant action, even though its mechanisms of action are far from clear.

Insulin like growth factor

Recent studies in rats have identified another neurotrophic factor, insulin-like growth factor (IGF-1), in antidepressant action and neurogenesis. Both systemic and central administration of IGF-1 increase cell proliferation in the adult hippocampus, and IGF-1 has been shown to selectively increase maturation of neurons [53,54]. Central administration of IGF-1 produces antidepressant-like effects in the forced swim tests in rats, which indicates that IGF-1 and IGF-1-induced signal transduction pathways might be another mechanism by which antidepressants exert their behavioral effects [55]. IGF-1 stimulates the phosphoinositide 3-kinase–PKB pathway, and has been shown to phosphorylate CREB in addition to the pro-apoptotic effector proteins glycogen synthase kinase 3β (GSK-3β) and the winged helix transcription factor Foxo1 [56].

The downstream signaling pathways of IGF-1, BDNF and even 5-HT demonstrate a high degree of overlap [57]. The functional effect of these pathways might be a combination of increasing cell proliferation and neurogenic pathways, all with the net effect of increasing synaptic strength and synaptic plasticity. Currently, there is little information available with respect to the behavioral effects of antidepressants in IGF-1 genetic models because IGF-1 mutant mice have a limited lifespan [58] and no conditional deletions of the gene encoding IGF-1 are available.

Antidepressant-induced neurogenesis

Reduced hippocampal cell volume has been observed in depressed humans in both magnetic resonance imaging (MRI) and post-mortem studies, compared with normal individuals [59,60]. Furthermore, antidepressant treatment has been shown to reverse or prevent this decrease in hippocampal volume [61]. Multiple classes of antidepressant drugs increase both cell proliferation and neurogenesis in the dentate gyrus of the adult hippocampus (Table 2), and this requires a chronic, and not an acute, time-course of administration. These findings have led to the hypothesis that antidepressant drugs might exert some of their therapeutic benefits by increasing hippocampal neurogenesis [62]. However, a major drawback in the majority of these studies is that a causal link between the administration of antidepressants, cell proliferation or neurogenesis and antidepressant efficacy has not been demonstrated.

A relationship between CREB and antidepressant-induced neurogenesis is supported by the finding that the majority of antidepressants that increase neurogenesis also activate a cAMP-related second messenger pathway and increase CREB phosphorylation. Furthermore, specific activation of the cAMP pathway has been shown to be neurogenic. The PDE4 inhibitor rolipram has recently been shown to activate CREB and increase hippocampal cell proliferation and neurogenesis in a chronic but not an acute time-course [63]. Rolipram also increases cell survival of newly formed cells and increases the number of branch points and length of dendrites in newly formed cells [63].

The role of P-CREB in neurogenesis is currently being studied by many laboratories. Newly formed hippocampal cells labeled with 5-bromo-2′deoxyuridine (BrdU) start to express P-CREB one to two weeks after their formation, with reduced expression of P-CREB at 4 weeks [64,65]. Although the function of CREB at these time points is

Table 2. Signaling pathways and antidepressant efficacy of compounds that increase hippocampal cell proliferation or neurogenesis[a]

Compound or treatment[b]	Signaling pathway or neurotransmitter system	Effect on proliferation or neurogenesis	Efficacy in animal models of depression	Clinical efficacy	Refs
Fluoxetine (SSRI)	5-HT	Increases, also increases cell survival	Yes	Yes	[3]
Tranylcypromine (MAOI)	5-HT	Increases	Yes	Yes	[3]
Imipramine(tricyclic)	5-HT and NA	Increases	Yes	Yes	[68]
Desipramine (tricyclic)	5-HT and NA	Increases	Yes	Yes	[3]
Reboxetine (SNRI)	NA	Increases	Yes	Yes	[3]
Venlafaxine(SNRI)	NA	Increases	Yes	Yes	[72]
Rolipram (PDE4 inhibitor)	CAMP–CREB	Increases, also increases cell survival	Yes	Unknown	[63]
Tianeptine	5-HT	Increases	Yes	Unknown	[4]
Tachykinin NK_1 receptor antagonist	NK_1 receptor	Increases	Yes	Unknown	[73]
Vasopressin V_3 receptor	Vasopressin	Increases	Yes	Unknown	[74]
ECT	cAMP–CREB (and others)	Increases	Yes	Yes	[3]
IGF-1	Ras–MAPK	Increases, also increases differentiation into neurons	Yes	Unknown	[55,75]
BNDF	cAMP–CREB	Increases	Yes	Unknown	[45]
Lithium	ERK	Increases	No	No	[76]
Valproate	ERK	Increases	No	No	[76]
5-HT_{1A} receptor agonist: 8-OH-DPAT	$G_{i/o}$–PKA, ERK	Increases	Yes	Unknown	[62,70]
5-HT_{1A} receptor antagonist: WAY100635[c]	$G_{i/o}$–PKA, ERK	Decreases or no change	No	Unknown	[77]
5-HT_{1B} receptor agonist: sumatriptan (GR43175)	$G_{i/o}$–PKA, ERK	No change but reverses PCPA-induced decrease	No	Unknown	[78]
5-HT_{1B} receptor antagonist: GR127935[c]	$G_{i/o}$–PKA, ERK	No change	No	No	[78]
5-$HT_{2A/C}$ receptor agonist: DOI	G_q–PKC	Increase	No	No	[78]
5-$HT_{2A/C}$ receptor antagonist: ketanserin	G_q–PKC	Decreases	No	No	[78]
5-HT_{2C} receptor agonist: RO600175[c]	G_q–PKC	Small change	Yes	Unknown	[78]

[a]Abbreviations: BDNF, brain-derived nerve growth factor; CREB, cAMP response element-binding protein; DOI, 1-(2,5-dimethoxy-4 iodophenyl) 2-aminopropane HCl; ECT, electroconvulsive therapy; ERK, extracellular signal-regulated kinase; IGF-1, insulin growth factor; MAOI, monoamine oxidase inhibitor; MAPK, mitogen-activated protein kinase; NA, noradrenaline; 8-OH-DPAT, (+/−)-8-hydroxy-2-(di-*n*-propylamino)tetralin; PCPA, para-chlorophenylalanine; PDE, phosphodiesterase; PKA, protein kinase A; SNRI, selective noradrenaline reuptake inhibitor; SSRI, selective serotonin reuptake inhibitor.
[b]The type of antidepressant is given in parentheses.
[c]See Chemical names.

currently unknown, it might be that it is acting in a pro-survival program. In support of this notion, Hastings and colleagues have shown that the number of newly formed cells marked with BrdU is greatly reduced between 2 and 4 weeks (compared with initial levels). The reduced expression of P-CREB in these newly formed cells at these same time-points might be a mechanism by which cells prevent apoptotic mechanisms and ensure their survival into mature neurons [66].

The associations between BDNF, neurogenesis and behavior have not as yet yielded a predictive effect. Baseline neurogenesis is reduced in $BDNF^{+/-}$ mice, and this is associated with a significant reduction in the volume of the dentate gyrus [67]. However, baseline changes in behavior have not been observed in these mice, suggesting a dissociation between behavioral and cellular endpoints of antidepressant effects in this particular genetic model. Recently, the effect of chronic antidepressant treatment was examined in $BDNF^{+/-}$ mice. Although chronic imipramine treatment increased cell proliferation immediately in both wild-type and $BDNF^{+/-}$ mice, three weeks later the amount of newly formed cells was significantly reduced in $BDNF^{+/-}$ mice, compared with wild-type mice [68]. These data indicate that BDNF is required for the long-term survival of newly formed neurons in the hippocampus. Future studies employing conditional knockout mice might reveal more about the interaction of CREB, BDNF and neurogenesis on behavior. In addition, the development of behavioral paradigms that respond to chronic antidepressant drug

Chemical names

GR127935: 2′-methyl-4′-(5-methyl-[1,2,4]oxadiazol-3-yl)-biphenyl-4-carboxylic acid
RO600175: 6-chloro-5-methyl-N-[6-(2-methylpyridin-3-yloxy)pyridin-3-yloxy]indoline-1-carboxamide
WAY100635: N-{2-[4-(2-methoxy-phenyl)-1-piperazinyl]ethyl}-N-(2-pyrindyl)cyclohexanecarboxamide trihydrochloride

treatment might be more sensitive to alterations in these long-term gene manipulations.

Although the majority of antidepressants activate the cAMP pathway and are neurogenic, there are some important exceptions. The atypical antidepressant tianeptine has not been shown to signal through cAMP, although it produces antidepressant-like behavioral effects and increases hippocampal cell proliferation and neurogenesis in multiple species [4]. Importantly, tianeptine not only increases cell proliferation in normal animals but also prevents stress-induced decreases in proliferation and neurogenesis.

In addition, there are compounds that activate similar second messenger pathways but are not clinically effective antidepressants. Lithium and valproate, for example, activate many of the same pathways as clinically effective antidepressants via inhibition of GSK3-β and activation of the MEK–ERK pathway. Lithium can also reverse the deleterious effects of stress on hippocampal plasticity [69]. Although CREB and ERK activation might be responsible for some of these effects, it does not follow that administration of a drug that activates ERK is an antidepressant. The differences between lithium, tianeptine and other antidepressants still need to be elucidated.

The final proof of concept for the role of neurogenesis in depression and antidepressant action would be to determine the effect of ablation of specific cells and/or cell types on behavior. A recent study in mice showed that X-ray irradiation, which inhibits cell proliferation, also prevented the behavioral effects of chronic administration of antidepressants [70]. The authors' conclusion was that the lack of cell proliferation was responsible for the loss of behavioral effects. However, it is also possible that irradiation affected multiple signal transduction mechanisms in addition to the direct effect on proliferating cells. It has been reported that low-dose irradiation produced not only perturbations in neurogenesis but ERK1,2, protein kinase B and CREB activation in the hippocampus and frontal cortex of mice, with repetitive exposure having a much more pronounced effect then acute exposure [71]. It is possible that the effect of the irradiation on the trophic and signal transduction pathways or factors might have contributed to the inability of antidepressants to produce the expected behavioral effect in the irradiated adult mice.

Conclusions and future directions

Although the field of antidepressant pharmacology has progressed rapidly during the past 10 years, there are many unanswered questions about the function and mechanism by which antidepressants exert their therapeutic effects. Recent research has moved beyond neurotransmitters to understanding the role of second messengers and their targets such as phosphorylated CREB and BDNF. Advanced genetic techniques have enabled the development of various rodent models, which permit evaluation of antidepressant effects at behavioral, molecular and cellular levels. Antidepressants might alter some or all of these effects through distinct pathways, many of which are related to CREB, BDNF and neurogenesis. In addition, antidepressant-induced neurogenesis might be one step in a larger pathway of signal transduction signaling that might contribute to the therapeutic effects of antidepressants.

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