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Research update

Emerging mechanisms and treatments for depression beyond SSRIs and SNRIs

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ABSTRACT

The monoamine hypothesis has been the prevailing hypothesis of depression over the last several decades. It states that depression is associated with reduced monoamine function. Hence efforts to increase monoamine transmission by inhibiting serotonin (5-HT) and norepinephrine (NE) transporters has been a central theme in depression research since the 1960s. The selective 5-HT reuptake inhibitors (SSRIs) and 5-HT and NE reuptake inhibitors (SNRIs) that have emerged from this line of research are currently first line treatment options for major depressive disorder (MDD). One of the recent trends in antidepressant research has been to refine monoaminergic mechanisms by targeting monoaminergic receptors and additional transporters (e.g. with multimodal drugs and triple re-uptake inhibitors) or by adding atypical antipsychotics to SSRI or SNRI treatment. In addition, several other hypotheses of depression have been brought forward in pre-clinical and clinical research based on biological hallmarks of the disease and efficacy of pharmacological interventions. A central strategy has been to target glutamate receptors (for example, with intravenous infusions of the N-methyl-D-aspartate (NMDA) receptor antagonist ketamine). Other strategies have been based on modulation of cholinergic and γ -aminobutyric acid (GABA)ergic transmission, neuronal plasticity, stress/hypothalamic pituitary adrenal (HPA)-axis, the reward system and neuroinflammation. Here we review the pharmacological profiles of compounds that derived from these strategies and have been recently tested in clinical trials with published results. In addition, we discuss putative treatments for depression that are being investigated at the preclinical level and outline future directions for antidepressant research.

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1. Introduction

The selective serotonin (5-HT) reuptake inhibitor (SSRI) and 5-HT and norepinephrine (NE) reuptake inhibitor (SNRI) antidepressants that were launched during the 1980s and 1990s are among the most successful drug treatments for psychiatric disorders and still remain the first line treatments for major depressive disorder (MDD). Monoamine oxidase (MAO) inhibitors have also been used for the treatment of MDD, but to a lesser extent. SSRIs and SNRIs are often presented as two major classes of antidepressants, with each class comprised of individual drugs that are highly similar. However, thorough pharmacological characterization has shown that drugs from these two classes have different levels of selectivity for their primary pharmacological target(s), i.e.

NE and/or 5-HT transporters (NET and SERT, respectively). Furthermore, they have notable affinity for secondary receptor or transporter targets at clinically relevant doses. For example, among the SSRIs paroxetine shows affinity for the NET and the muscarinic cholinergic receptor, sertraline for the dopamine (DA) transporter (DAT), fluoxetine for the 5-HT_{2C} receptor, citalopram for the sigma₁ and histamine (HA) H₁ receptors, and escitalopram for the sigma₁ receptor [1–3]. Similarly, SNRIs show differences in potency for SERT vs. NET with venlafaxine having a preference for SERT over NET, and duloxetine being a more balanced drug with respect to 5-HT and NE reuptake inhibitory potencies [4]. These distinct pharmacological properties may lead to different clinical efficacy and/or tolerability profiles [4,5]. However, possibly due to a lack of validated biomarkers that can reliably predict a patient's response to a given antidepressant, it appears that only ~50% of patients diagnosed with MDD go into clinical remission using treatments from these two drug classes regardless of the drug chosen [6]. For four consecutive treatment steps, the overall

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cumulative remission rate is only ~56% [6]. This leaves a large group of patients who respond inadequately or not at all to extensive therapeutic intervention. Moreover, the therapeutic response to SSRIs/SNRIs is often delayed, requiring several weeks of treatment. Many of the current antidepressants also have drug-related adverse effects, such as nausea and sexual dysfunction [7]. Thus, there is a large unmet need for additional treatment options.

While the etiology and pathology underlying MDD still by large remain unknown, many hypotheses of depression have been brought forward in antidepressant research. Several of these investigations have led to drug discovery efforts and in some cases advanced into drug development programs (Fig. 1, Tables 1 and 2). Interestingly, the rationale of these hypotheses often originated from pharmacological manipulations performed in depressed patients and was not based on an understanding of the pathophysiology of disease.

Over the last 40–50 years the prevailing hypothesis of depression has been the monoamine hypothesis which included the catecholamine [8] and 5-HT [9] hypotheses. It originated from mechanistic studies of the serendipitously discovered tricyclic antidepressants (TCA) and MAO inhibitors. SSRIs and SNRIs that came out of this line of research were conceived to mainly improve tolerability of TCAs. Since the discovery of SSRIs and SNRIs, one strategy in antidepressant research has been to refine and expand on the monoaminergic mechanisms by either targeting monoaminergic receptors or additional transporters in one molecule or by augmenting the effects of SSRIs or SNRIs by adjunctive treatment with another drug. The main objectives have been to improve efficacy and/or reduce the time to onset the therapeutic effect of SSRIs and SNRIs. We review drug target profiles that came out of these efforts and have been tested in the clinic in Section 2 and Table 2. Additional target profiles that have only been investigated in preclinical models are summarized in Table 1 and Section 5.

Other research efforts have focused on hypotheses of depression that go beyond the monoamines (Fig. 1, Table 1).

During the last decade there has been an increasing interest in targeting glutamate neurotransmission. The interest in glutamate targets precipitated from the spectacular clinical finding that intravenous infusion of the N-methyl-D-aspartate (NMDA) receptor antagonist ketamine can produce an immediate antidepressant effect in patients with treatment-resistant depression (TRD) [10]. Several other glutamate targets have been defined, some of which have been tested in the clinic and are discussed in Section 3.

The neuroplasticity hypothesis of depression links to the glutamate hypothesis. It is based on the observation that enhanced neuronal plasticity (e.g. neurogenesis, dendritic branching, synaptogenesis) appears to be a shared mechanism for antidepressants and that depression risk factors, such as stress, reduce neuroplasticity in the hippocampus and prefrontal cortex (PFC) (Fig. 1, [11]). There have been extensive preclinical efforts to identify neuronal plasticity related drug targets (Table 1 and Section 5), but the hypothesis has so far not been validated clinically.

Additional hypotheses of depression have been based on modulation of cholinergic transmission, stress/hypothalamic pituitary adrenal (HPA)-axis, reward system, neuroinflammation and γ -butyric acid (GABA) transmission (Fig. 1, Table 1). Compounds that were generated to test these theories (e.g. corticotropin releasing factor (CRF) antagonists, neurokinin (NK) antagonists, kappa-opioid antagonists, Tumor Necrosis Factor (TNF) α antibody) are discussed in Section 4. Compounds with only preclinical mechanistic data are outlined in Table 1 and some of them are discussed in Section 5. Finally, possible future directions for antidepressant research are discussed at the end of this review in Section 6.

2. Refining and expanding monoaminergic mechanisms beyond SSRIs and SNRIs

Whereas a general increase in extracellular monoamine levels via inhibition of monoamine transporters may trigger a dampening

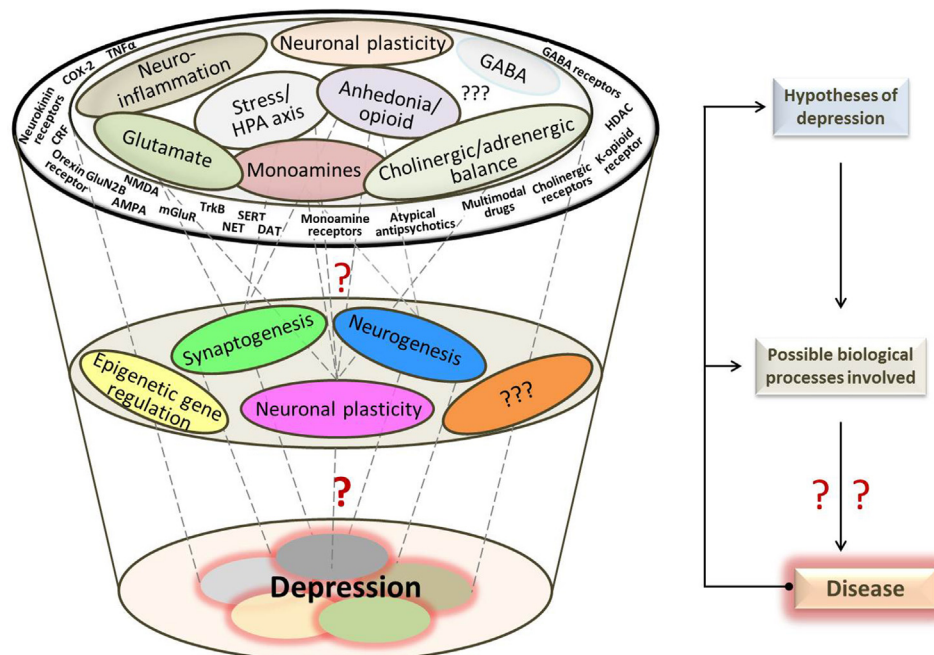


Fig. 1. Summary of current hypotheses of depression. Proposed hypotheses of depression and their associated drug targets are shown in the top oval. For a full list of drug targets see Table 1. Several biological processes involved in the etiology of depression are listed in the middle oval. The bottom oval represents human depression with different colors depicting heterogeneity in the etiology, diagnosis, and clinical manifestation of the disease. Examples of relationships between different hypotheses of depression, the biological processes that they are proposed to impact and the human disease are shown with dotted lines.

Table 1

Overview of hypotheses of depression and the associated drug target profiles. For the preclinical column, only target profiles with available tools compounds are listed. The table does not include established antidepressants, such as TCAs, MAO inhibitors, SSRIs and SNRIs that have been on the market for many years.

Hypothesis	Biological evidence in humans	Drug target profile	
		Clinical validation	Preclinical validation only
Monoamine			
The pathophysiology of depression involves low levels of 5-HT, NE, and/or DA levels in the CNS [9,207]	Antidepressant drugs elevate CNS levels of one or more monoamines Tryptophan depletion can provoke relapse of depression SSRIs, SNRIs, TCAs, MAOIs have clinical efficacy No direct evidence for a causal relation between depression and monoaminergic dysregulation	Triple uptake inh Atypical antipsychotics (add on) Multimodal	5-HT _{1A} ago (postsynaptic selective) 5-HT _{2B} ant 5-HT _{2C} ant 5-HT ₃ ant 5-HT ₇ ant α_{2A} adrenoceptor ago DA D ₂ ago DA D ₃ ago VMAT2 inh TAAR1 ago
Glutamate			
Depression involves dysfunctional glutamate signaling in the brain which leads to impaired neuroplasticity [11,120]	Clinical efficacy of ketamine Changes in plasma and CSF levels of glutamate and brain glutamate and glutamine levels in MDD patients, but results are inconsistent Reduced expression of excitatory amino acid transporters in brain tissue from MDD patients suggestive of impaired glutamate clearance	NMDA open channel blockers GluN2B ant Glycine ago mGluR2 NAM mGluR5 NAM	AMPAkines
Neuronal plasticity			
Depression is ascribed to impaired plasticity of neural circuits and connections, and involves reduced hippocampal neurogenesis, dendritic de-branching and loss of dendritic spines and synapses [187]	Most antidepressant treatments have shown to increase neuronal plasticity in preclinical studies Reduced hippocampal volume in MDD patients Reduced neuronal and glial density in post mortem brain tissue from MDD patients Reduced serum levels of BDNF No direct evidence for a causal relation to MDD	Neurogenesis stimulator (NSI-189)	TrkB ago
Cholinergic/adrenergic balance			
Depression is associated with hyperactivation of the cholinergic system and as a consequence decreased activity in the noradrenergic system [163,164,167]	Cholinesterase inhibition exacerbates depression Muscarinic antagonists (TCAs, scopolamine) have antidepressant activity No direct evidence for a casual relation to MDD	Muscarinic ant $\alpha 4\beta 2$ nicotinic ant	Muscarinic M ₃ ant $\alpha 7$ nicotinic ago
Anhedonia/Opioid			
Anhedonia, a core symptom of depression, is ascribed to a disrupted reward circuitry where the brain opioid system plays a key regulatory role. Stress induces the release of dynorphin, which activates kappa opioid receptors and down regulates DA levels [169]	Kappa opioid agonist induces depression	Kappa opioid ant Nociceptin ant	
Stress/HPA-axis			
Depression is precipitated through a dysregulation of the hypothalamic pituitary-axis [146,148,208]	Stress can precipitate depressive episodes in a subset of patients Some depressed patients have hyperactive adrenal glands, exhibit elevated responses to stress and have elevated levels of CRF in cerebrospinal fluid	CRF ant GR ant NK1 ant NK2 ant Vasopressin ant Orexin ant	CB ₁ ago FAAH inh
Neuroinflammation			
Pro-inflammatory and kynurenine pathways are activated in depression and contribute to the pathophysiology of disease [209]	Immunosuppressant drugs can induce depression High incidence of depression in patients with inflammatory disorders Elevated levels of inflammatory markers in plasma and post mortem brains from MDD patients	TNF α antibody COX-2 inh	KMO ant P2x7 ant
GABA			
Depression is associated with reduced GABA neurotransmission in cortical circuits [210]	Reduced CNS level of GABA in some MDD patients		GABA _A modulators (neurosteroids) GABA _B ant

Abbreviations: Ant: antagonist; Inh: inhibitor; Ago: agonist; NAM: negative allosteric modulator.

Table 2
Overview of recently approved antidepressants and drugs in clinical development for the treatment of MDD. *For approved drugs, doses are shown according to their drug label; for experimental drugs, doses were taken from clinical trials according to <https://clinicaltrials.gov>.

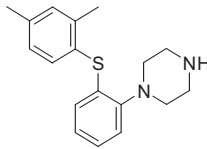
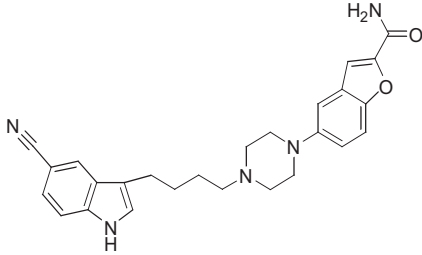
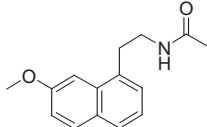
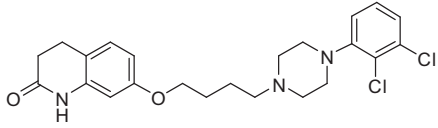
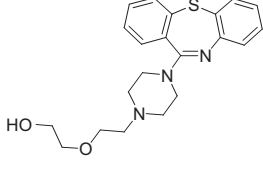
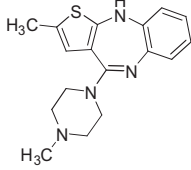
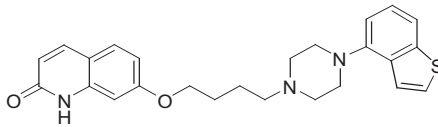
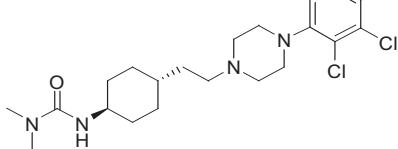
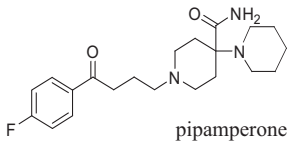
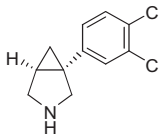
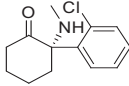
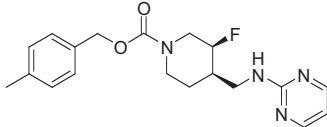
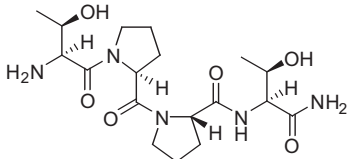
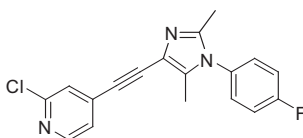
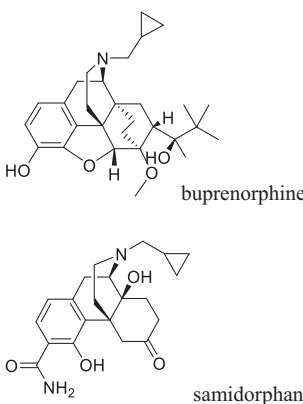
Drug (dose, mg/day, po)*	Putative MoA in MDD	Treatment regimen	Status	Chemical structure
Vortioxetine (Lu AA21004) (5–20)	5-HT ₃ , 5-HT ₇ and 5-HT _{1D} ant, 5-HT _{1B} partial ago, 5-HT _{1A} ago, SERT inh	Monotherapy	Launched in 2014	
Vilazodone (40)	5-HT _{1A} partial ago, SERT inh	Monotherapy	Launched in 2011	
Agomelatine (25–50)	MT ₁ and MT ₂ ago, 5-HT _{2C} ant	Monotherapy	Launched in 2009	
Aripiprazole (2–15)	D ₂ and 5-HT _{1A} partial ago, 5-HT ₂ ant	Add-on	Launched in 2007	
Quetiapine (150–300)	D ₂ and 5-HT ₂ ant	Add-on	Launched in 2009	
Symbyax [®] (olanzapine and fluoxetine) (6–12 olanzapine, 25–50 fluoxetine)	D ₂ and 5-HT ₂ ant	Fixed dose combination of olanzapine and fluoxetine	Launched in 2009	 olanzapine
Brexipiprazole (OPC-34712) (1–3)	D ₂ and 5-HT _{1A} partial ago, 5-HT ₂ , alpha1B and alpha2C ant	Add-on	NDA submitted in 2014	
Cariprazine (RGH-188) (2–4.5)	D ₂ , D ₃ and 5-HT _{1A} partial ago	Add-on	Phase 3	
PNB-01 (pipamperone 15, citalopram 20–40)	D ₂ , D ₄ and 5-HT ₂ ant	Combination of pipamperone and citalopram	Phase 3 ongoing but not recruiting	 pipamperone

Table 2 (Continued)

Drug (dose, mg/day, po) ^a	Putative MoA in MDD	Treatment regiment	Status	Chemical structure
Amitifadine (DOV-21947/EB-1010) (50–100)	DAT, SERT, NET inh	Monotherapy	Phase 3	
Esketamine ((S)-enantiomer of ketamine) (0.20–0.40 mg/kg, i.v.; 14–84 mg intra nasal)	Non-competitive open-channel blocker of NMDA receptors	Monotherapy	Phase 2	
CERC-301 (MK-0657) (4–12)	NMDA GluN2B ant	Add-on or monotherapy	Phase 2	
GLYX-13 (rapastinel) (5, 10 mg/kg, i.v.)	NMDA glycine site partial ago	Monotherapy	Phase 2	
NRX-1074 (1.5, 10 mg, i.v.)	NMDA glycine site partial ago	Monotherapy	Phase 2	Structure not published
Basimglurant (RG-7090) (0.5, 1.5) [145]	mGluR5 NAM	Monotherapy	Phase 2	
LY-2940094 (40)	Nociceptin ant	Monotherapy	Phase 2	Structure not published
ALKS-5461 (1:1 ratio for best efficacy; establishment of clinical dose response is ongoing) [171]	Kappa ant	Combination of buprenorphine and samidorphan	Phase 3	

^a Abbreviations: Ant: antagonist; Ago: agonist; Inh: inhibitor; NAM: negative allosteric modulator.

of response by stimulating monoaminergic receptors with opposing activities (e.g. see regulation of 5-HT transmission via pre- vs. postsynaptic stimulation of 5-HT_{1A} receptors as discussed below), targeting a single receptor may be inadequate due to system redundancies (e.g. see selective peptide receptor targets discussed in Section 4). Hence, multitarget (≥ 2 targets) or multimodal (≥ 2 targets from different target classes [12]) approaches that involve additive or synergistic effects of selected biological targets may be an attractive strategy for refining and improving efficacy and/or tolerability of currently used antidepressants. These ideas have been extensively discussed in the literature by several research groups [13–16] and are an overarching theme for this section. The authors wish to highlight in particular the very comprehensive reviews by Millan on this topic [16–19].

2.1. Add-ons to SSRIs and SNRIs in MDD

2.1.1. Augmentation mechanisms

For patients with an inadequate response to SSRI/SNRI therapy, a common strategy has been to prescribe a second drug as an adjunctive treatment in an attempt to either increase the efficacy of the first antidepressant or to treat residual symptoms. This strategy has been followed empirically by clinicians for many years based on a patient's symptoms. One of the first hypothesis-driven efforts that came out of preclinical research was to add a 5-HT_{1A} receptor partial agonist (or antagonist) to an SSRI [20]. This was based on data showing that desensitization of 5-HT_{1A} autoreceptors located in the raphe nuclei, a center for 5-HT producing cells, was necessary for the maximum effect of SSRIs [20]. The interpretation was that simultaneous application of a 5-HT_{1A}

receptor antagonist with an SSRI would instantaneously inhibit the negative feedback mechanism and allow for an immediate and sufficient increase in 5-HT levels to produce a fast antidepressant response. However, since enhanced serotonergic neurotransmission through stimulation of postsynaptic 5-HT_{1A} receptors was hypothesized to be essential for the antidepressant effect of SSRIs, it was important to only block presynaptic 5-HT_{1A} receptors [21]. Encouraging results from clinical studies using combinations of SSRIs and pindolol, a 5-HT_{1A} receptor partial agonist and a β -adrenoceptor antagonist, showed a faster onset of antidepressant effect than SSRIs alone and further fueled the interest in this research area (e.g. [20]). Although pindolol was not developed as an adjunctive treatment to SSRIs, the principle led to the initiation of major efforts by pharmaceutical companies in the 1990s to find compounds that interacted with both 5-HT_{1A} receptors and SERT. Two novel multimodal antidepressants, vilazodone and vortioxetine, that are currently on the market for treatment of MDD, target both 5-HT_{1A} receptors and the SERT in their mechanism of action (Section 2.3, Table 2).

Several additional 5-HT receptors (5-HT_{1B/1D}, 5-HT₂, 5-HT₃, 5-HT₄, 5-HT₆ and 5-HT₇ receptors) have also been investigated pre-clinically and were shown to increase 5-HT neurotransmission beyond the level achieved with SSRIs by impacting negative neuronal feedback loops [22]. For instance, antagonism at postsynaptic 5-HT_{2A/2C} receptors was shown to increase 5-HT levels produced by SSRIs, most likely via inhibition of GABAergic interneurons in the dorsal raphe nucleus (DRN) [23]. Interestingly, the 5-HT₂ receptor antagonism is a prominent mode of action of atypical antipsychotics that are now prescribed as add-ons to SSRIs and SNRIs.

2.1.2. Atypical antipsychotics as an augmenting strategy

Several atypical antipsychotics (olanzapine, risperidone, quetiapine, aripiprazole, paliperidone, ziprasidone and amisulpride) have been tested in clinical MDD trials as a strategy to augment SSRIs/SNRIs and some have obtained label claim for the treatment of MDD [24]. Aripiprazole was the first of the atypical antipsychotics to be approved in 2007 as an adjunct treatment in MDD [25]. In 2009, quetiapine was also approved for augmentation in MDD [26], and a fixed dose combination of fluoxetine and olanzapine was approved for TRD the same year [27].

As described in Section 2.1.1, the synergistic effect between atypical antipsychotics and SSRIs/SNRIs most likely involves the inhibition of 5-HT reuptake and antagonism of 5-HT_{2A/C} receptors (Table 2), which leads to an increase in 5-HT levels beyond what is achieved with an SSRI. Interestingly, there is very limited clinical evidence for the use of selective 5-HT₂ receptor antagonists as standalone therapy [28]. Preclinical studies suggest that these compounds might only be useful in augmentation strategies, likely because an increased endogenous 5-HT tone, achieved through the blockade of SERT, is required for a 5-HT₂ receptor antagonist to exert its functional activities [29].

Several other compounds are currently being investigated as adjunctive treatments for MDD. A new drug application (NDA) was recently filed for the use of brexpiprazole (OPC-34712 [30], a serotonin-dopamine activity modulator that is a partial agonist at 5-HT_{1A} and D₂ receptors and an antagonist at 5-HT_{2A} and α _{1B/2C} adrenergic receptors [31] (Table 2). Furthermore, cariprazine, a DA D₂, DA D₃ and 5-HT_{1A} receptor partial agonist [32], is currently undergoing clinical testing as adjunctive treatment for MDD [33]. On the other hand, PNB-01, a combination of citalopram and a low dose of pipamperone (D₂, D₄ and 5-HT_{2A} receptor antagonist), has failed to show any advantages over the treatment with citalopram alone in one clinical study [34]. A follow-up phase 3 study has been registered at <http://clinicaltrials.gov>, but patients have not yet been recruited (<https://clinicaltrials.gov/show/NCT01312922>).

In conclusion, the add-on of antipsychotics to SSRIs/SNRIs has shown improved clinical efficacy over the first line treatment. However, the strategy has obvious limitations due to the potential risk of drug–drug interactions, which may complicate achieving the right therapeutic levels, as well as the stigmatization associated with the use of antipsychotics in the treatment of MDD [12].

2.1.3. Additional augmentation strategies

Several other augmentation strategies have been pursued, but with limited success. The selective NE reuptake inhibitor, edivoxetine, and the dextroamphetamine prodrug, lisdexamfetamine, showed no augmentation effect with SSRIs in the clinic, and the programs were recently discontinued [35,36]. An add-on of the 5-HT_{1B/1D} receptor antagonist elzasonan (CP-448187) to sertraline was also discontinued in 2008 with no published data on its clinical efficacy (<http://clinicaltrials.gov/show/NCT00275197>).

2.2. Triple reuptake inhibitors

As a consequence of the SNRI success in treatment of MDD, researchers have pursued the idea of developing compounds blocking all three monoamine transporters, the so-called 5-HT, NE, DA reuptake inhibitors (SNDRI), or triple reuptake inhibitors. This concept was based on the positive role of DA on the reward system and the fact that anhedonia is a prominent symptom in a subset of MDD patients, especially those with melancholic depression [37]. This hypothesis was further supported by positive clinical augmentation studies with DA-enhancing drugs [37].

During the past decade, several SNDRI drug candidates have been developed and tested in the clinic. The main concerns with these drugs have been how to avoid exaggerated dopaminergic stimulation and abuse liability. Amitifadine (DOV-21947 or EB-1010), which was among the first SNDRI drug candidates, preferentially enhanced 5-HT with 1:2:8 potency rankings for the inhibition of SERT, NET, and DAT, respectively [38]. Amitifadine showed efficacy in a small clinical proof-of-concept (PoC) study. However, amitifadine did not meet the clinical endpoint in a subsequent larger double-blind placebo-controlled study in MDD patients who had failed one treatment with a first-line antidepressant [39]. Additional clinical studies in MDD are being planned with this compound at higher doses.

Other efforts to develop more balanced SNDRI with similar K_i values for SERT, NET and DAT have also failed, in most cases before reaching PoC studies. Two compounds, NS-2359 (GSK-372475) and liafensine (BMS-820836), were evaluated in phase 2 clinical studies, but both programs were terminated. In a large phase 2 program with 900 patients, NS-2359 was found neither efficacious nor well tolerated, whereas comparators paroxetine and venlafaxine separated significantly from placebo [40].

Thus, in spite of significant investments, the clinical value of SNDRI for the treatment of depression remains to be demonstrated. There are still several SNDRI in preclinical development (e.g. LPM570065, [41]) that might be clinically tested at a later time. However, the failure of the DA-releasing compound lisdexamfetamine to show efficacy as adjunctive treatment in MDD patients who responded inadequately to monotherapy with an SSRI or SNRI [36] is not supportive of a role of enhanced DA transmission in promoting antidepressant activity. SNDRI might have better efficacy in a well-defined subset of MDD patients with prominent symptoms of anhedonia (i.e. in melancholic depression). This remains to be substantiated in future clinical studies.

2.3. Multimodal antidepressants

Another strategy to augment 5-HT transmission beyond what is achieved with an SSRI has been to target 5-HT receptors in

combination with SERT inhibition in one molecule. The pharmacology behind these efforts is described in Section 2.1. This strategy has led to a new class of antidepressants with a multimodal mechanism of action [12]. Vilazodone, a 5-HT_{1A} receptor partial agonist and SERT inhibitor (Section 2.3.1) and vortioxetine, a 5-HT_{1A} receptor agonist, 5-HT_{1B} receptor partial agonist and 5-HT₃, 5-HT₇ and 5-HT_{1D} receptor antagonist and SERT inhibitor (Section 2.3.2), are two multimodal antidepressants that have recently received market authorization for the treatment of MDD. In addition, a positive dose-finding clinical study has been performed in MDD patients with tedatioxetine (Lu AA24530), a 5-HT₃ and 5-HT_{2C} receptor antagonist and SERT inhibitor, results from which were published in a press release form [42].

2.3.1. Vilazodone

Vilazodone (EMD 68843) has been approved in the United States (US) for the treatment of MDD in adults. The drug originates from a drug discovery program undertaken in the mid 1990s and was taken through a phase 2 clinical development program in MDD patients in the late 1990s until the early 2000s [43]. The program was discontinued due to disappointing results [44]. However, later the clinical development program was reinitiated and developed in the US only. After completion of two positive placebo-controlled phase 3 studies the FDA granted a market authorization in 2011.

In spite of a long development time, the preclinical and the clinical literature on vilazodone are limited. In preclinical studies, vilazodone acts as a partial agonist at 5-HT_{1A} receptors and a SERT inhibitor. It increases extracellular 5-HT beyond levels seen with SSRIs in microdialysis studies in rats without affecting cortical NE and DA levels [45]. The potentiating effect on 5-HT levels has been ascribed to vilazodone's partial agonism at 5-HT_{1A} receptors [46]. The direct effect at 5-HT_{1A} receptors was also shown in electrophysiology recordings in which vilazodone acutely suppressed the firing rate of serotonergic DRN neurons in 5-HT depleted rats where SSRIs do not work [47]. Interestingly and unexpectedly (given the rationale for its pharmacological profile [Section 2.1.1]), the time course for desensitization of 5-HT_{1A} autoreceptors was similar to that of an SSRI and still required 14 days administration of vilazodone [47].

The lack of effect of vilazodone on NE and DA levels in the cortex is puzzling since selective 5-HT_{1A} receptor agonists (both full and partial agonists) are known to increase cortical levels of NE and DA [48,49]. However, increased 5-HT release has been shown to attenuate cortical release of NE and DA [50–52]. It is therefore possible that these two opposing effects result in a neutral effect of vilazodone on NE and DA. Whereas SSRIs have been associated with 5-HT-mediated cognitive and emotional blunting and weaker effects on anhedonia than drugs that also enhance NE and DA release [53–57], there are no data for vilazodone on these measures. Furthermore, there is no information on vilazodone's effect on NE and DA release after repeated dosing.

Vilazodone is active in classical behavioral models predictive of antidepressant and anxiolytic activity, but in some instances the dose-response curve was biphasic, possibly due to its partial agonistic activity at 5-HT_{1A} receptors (a partial agonist will act as an antagonist in a system with an elevated 5-HT tone) and the involvement of both pre- and post-synaptic 5-HT_{1A} receptors in mediating these effects [58]. Since a clinical dose response relation has not been established (see below), it is unknown whether this biphasic dose response relation translates into the clinic.

Five double-blind randomized placebo-controlled phase 2 studies and two phase 3 studies in adult MDD patients were the basis for efficacy evaluation of the NDA [59]. Vilazodone did not separate from placebo on the primary endpoint in any of the phase 2 studies. In three of the phase 2 studies that included an active reference, the reference also failed to separate from placebo, suggesting that

these were failed studies [59]. Two short-term phase 3 studies compared vilazodone at 40 mg/day to placebo, and both studies met the primary endpoint [59]. The most common adverse effects were related to the gastrointestinal tract and sleep quality, which are well-known adverse effects of SERT inhibitors and selective 5-HT_{1A} receptor agonists [60]. Hence, vilazodone which displays both activities has relatively high rates of gastrointestinal adverse effects such as diarrhea, nausea and vomiting and requires dose titration over two weeks in order to reach the daily target dose of 40 mg [59,61].

According to <https://clinicaltrials.gov>, several clinical studies with vilazodone are still ongoing, some of which are phase 4 commitments related to the FDA approval (e.g., long-term efficacy and safety, efficacy in adolescents and in a geriatric population in MDD), others are directed toward investigating the efficacy of vilazodone in other indications, including generalized anxiety disorder, social anxiety disorder and post-traumatic stress disorder. In conclusion, vilazodone is a new multimodal antidepressant with interesting pharmacology, but it is too soon to evaluate its impact in everyday clinical practice.

2.3.2. Vortioxetine

The drug discovery program that led to the development of vortioxetine (Lu AA21004) also had its origins in the 5-HT augmentation hypothesis (Section 2.1.1). However, during the drug discovery phase of the project the target profile was redirected toward a combination of SERT inhibition, 5-HT_{1A} receptor agonism and 5-HT₃ receptor antagonism [62], in part because it was found that the 5-HT₃ receptor antagonism potentiated the increase in extracellular 5-HT levels produced by SERT inhibition [63]. Vortioxetine has been approved in major markets since 2013, including the US, European Union (EU), Canada, South Africa, Australia, Mexico and South Korea, for the treatment of MDD.

Vortioxetine is a 5-HT₃, 5-HT₇ and 5-HT_{1D} receptor antagonist, a 5-HT_{1B} receptor partial agonist, a 5-HT_{1A} receptor agonist and a SERT inhibitor in cellular assays and shows antidepressant and pro-cognitive activities in preclinical animal models [62,64]. It increases 5-HT (beyond that of an SSRI), NE, DA, acetylcholine (ACh), and HA levels in rat brain regions associated with MDD, such as the PFC and the ventral hippocampus [63,65]. Furthermore, vortioxetine enhances glutamatergic neurotransmission, most likely through inhibiting GABA interneurons [66,67]. The 5-HT₃ receptor antagonism plays a prominent role in the pharmacology of vortioxetine since the 5-HT₃ receptor agonist SR57227 can reverse the potentiating effect of vortioxetine on glutamatergic and serotonergic transmission [68]. However, the modulation of other 5-HT receptor subtypes, including antagonism of 5-HT₇ receptors and partial agonism at 5-HT_{1B} receptors, might also contribute to vortioxetine's overall pharmacological effects [69,70].

Vortioxetine exhibits antidepressant- and anxiolytic-like properties in classical monoamine-sensitive behavioral models of depression, but also in models that are insensitive to SSRIs/SNRIs, such as the progesterone withdrawal model [71] and in aged mice [72]. Interestingly, and different from any other antidepressants tested including SSRIs, vortioxetine had no effect on sucrose drinking in a chronic mild stress model of depression/anhedonia [64]. The mechanism underlying this lack of effect is currently not understood and it does not translate into reduced clinical efficacy on anhedonia related symptoms, as measured by clinical rating scales [64]. To date there are no studies that specifically assess the effect of vortioxetine on emotional blunting and given vortioxetine's complex modulation of multiple neurotransmitter systems, this question can only be addressed with empirical testing. Finally, in line with its mechanistic data and different from SSRIs,

vortioxetine enhances a broad range of cognitive functions, including attention/vigilance measured by quantitative electroencephalography [73], executive function, memory, and learning in animal models (reviewed in [64]). Taken together, these preclinical observations indicate that vortioxetine has a complex pharmacological profile that differs from those of the SSRIs and SNRIs.

The implications of vortioxetine's modulation of multiple neurotransmitter systems on its antidepressant and pro-cognitive potential are complex and the net effect can only be evaluated through empirical data. For instance, the increased 5-HT, NE and DA neurotransmission after acute and chronic dosing would, in line with the monoamine hypothesis, favor vortioxetine's antidepressant potential. On the other hand, increased 5-HT level may dampen, whereas the enhanced NE and DA levels may favor a pro-cognitive profile. Vortioxetine's increased ACh transmission would, in line with the cholinergic hypothesis of depression, disfavor its antidepressant activity. However, microdialysis studies indicate that the magnitude of the effect produced by vortioxetine on this neurotransmitter system is much less pronounced than that obtained with the cholinesterase inhibitor donepezil (*G. Smagin, unpublished observation*), and there is no effect on ACh release after chronic dosing with vortioxetine [63,65]. Thus, the cholinergic enhancing properties of vortioxetine may therefore contribute to its pro-cognitive activities, but may have less of an impact after repeated dosing. In addition, vortioxetine produces a sustained increase in cortical and hippocampal HA transmission after chronic dosing [74]. The brain HA system plays an important role in promoting cognitive function and attention. Thus, vortioxetine-induced increase of HA levels is likely to contribute to its pro-cognitive effects. The effect of HA signaling on mood is poorly defined. However, we cannot exclude the possibility that HA-dependent enhancement in cognition can indirectly make positive contributions to mood. Finally, vortioxetine's modulation of glutamate neurotransmission should favor its antidepressant and pro-cognitive properties since several measures indicate that vortioxetine promotes glutamate dependent neuronal plasticity to a degree that is superior to SSRIs. Vortioxetine, unlike the SSRI escitalopram, disinhibits 5-HT-induced suppression of pyramidal neuron activity in rat hippocampal slices and consequently increases long-term potentiation, a measure of increased synaptic plasticity [66]. Furthermore, gene expression studies show that vortioxetine, unlike fluoxetine, acutely activates genes associated with neuroplasticity in rats (i.e. mammalian target of rapamycin (mTOR), metabotropic glutamate 1 (mGlu1) receptor, spinophilin, protein kinase C (PKC) α , Homer1, Homer3) [75]. Similarly, after chronic dosing, vortioxetine activates neuronal plasticity related genes, improves visual spatial memory deficits, and shows antidepressant-like activity in 12 months old mice, whereas fluoxetine has no effects on any of these measures in old mice [76]. Vortioxetine also increases cell proliferation in the hippocampal dentate gyrus faster than fluoxetine (3 days for vortioxetine compared to 10 days for fluoxetine) [77] in rats, and produces a larger degree of dendritic branching than fluoxetine after two weeks of dosing in mice [78].

Vortioxetine's neuroplasticity enhancing properties support a notion that its antidepressant and pro-cognitive profiles are mediated, at least to some extent, through increased glutamate neurotransmission and neuroplasticity. It is important to note that although enhanced glutamate neurotransmission is thought to favor plasticity and subsequent antidepressant and pro-cognitive effects, it is also clear that exaggerated glutamate release (for instance, in relation to stress) can be neurotoxic (reviewed in [11,79]). Vortioxetine's effects on glutamate appear to be limited to enhanced neuronal function. This has been shown in microdialysis studies where no change in extracellular glutamate was detected in the ventral hippocampus and PFC upon the treatment with vortioxetine [80].

In a comprehensive clinical program for MDD, vortioxetine showed dose-dependent efficacy and was well tolerated (e.g., [81]). The short-term (6 or 8 weeks) program consisted of 12 randomized double-blind placebo-controlled studies and one 12-week study in MDD patients showing an inadequate response to SSRIs or SNRIs vs. agomelatine [64]. On the pre-specified primary efficacy endpoint, vortioxetine was statistically significantly superior to placebo in seven of the placebo-controlled studies, and the observed effect size was clinically relevant [64]. Vortioxetine was also superior to agomelatine [64]. Long-term efficacy of vortioxetine was shown in a placebo-controlled relapse-prevention study [82] and in two open-label extension studies [83,84]. Finally, in line with preclinical support for vortioxetine having pro-cognitive activities, vortioxetine showed a positive effect on pre-defined cognition outcome measures in three double-blind randomized placebo-controlled studies of cognitive dysfunction in MDD patients, two of which included the active reference duloxetine [85–87].

Vortioxetine was well tolerated in short-term as well as long-term clinical studies, with a low level of discontinuation symptoms, likely due to its relatively long half-life. The adverse effect with the highest incidence was nausea, which in general was of mild or moderate intensity and transient. There was a relatively low incidence of sexual dysfunction and sleep disruption [88,89]. Overall, vortioxetine showed strong evidence for antidepressant efficacy with good tolerability [90]. In addition, it showed improvement of cognitive dysfunction in patients with MDD.

Vortioxetine's clinically effective dose range of 5–20 mg/day spans from approximately 50 to >80% SERT occupancy [91]. Given that effective doses of SSRIs typically correspond to at least 80% SERT occupancy [92], the results at <80% SERT occupancy support the hypothesis that the antidepressant effects of vortioxetine arise from both receptor modulation and inhibition of the SERT. However, it has been difficult to determine vortioxetine's occupancy at 5-HT receptors in humans. An attempt to determine 5-HT_{1A} receptor occupancy using the 5-HT_{1A} receptor antagonist radiotracer [¹¹C]WAY100-635 in a positron emission tomography (PET) study failed to demonstrate occupancy of vortioxetine at this receptor [93]. However, using a 5-HT_{1A} receptor antagonist radiotracer may not be an appropriate method of estimating receptor occupancies for a drug like vortioxetine that acts as a full agonist. The 5-HT_{1A} receptor complex can exist in a coupled or decoupled state with its G protein signaling complex. The decoupled state is associated with low agonist affinity, whereas the antagonist affinity is unaffected by the coupling state [94–96]. Thus, vortioxetine might not displace 5-HT_{1A} antagonist binding at 5-HT_{1A} receptors that are in decoupled state. Furthermore, even though selective 5-HT_{1A} receptor agonist and 5-HT_{1B} receptor antagonist radiotracers have become available for human use over the last few years (i.e. [¹¹C]CUMI-101 and [¹¹C]AZ10419369, respectively [97,98]), they might not be useful for a drug like vortioxetine because these ligands are sensitive to changes in extracellular 5-HT. Since 5-HT_{1A} and 5-HT_{1B} receptors have a high affinity for 5-HT, elevated 5-HT levels produced by SERT inhibition would likely compete with vortioxetine's displacement of these radiotracers. There are no radiotracers for 5-HT₃ and 5-HT₇ receptors that have been validated for human use. Thus, vortioxetine's occupancy at 5-HT receptors in humans remains to be demonstrated.

In conclusion, even though vortioxetine's primary drug targets are serotonergic, the complexity of the 5-HT system and the fact that vortioxetine only modulates a subset of 5-HT receptors lead to its complex pharmacological profile that overlaps with a number of hypotheses of depression. Future clinical studies with a translational focus will hopefully provide further insights into how vortioxetine's preclinical profile may define its efficacy in subpopulations of patients with MDD.

2.4. Agomelatine

Another approach to treat the symptoms of depression has been to target 5-HT beyond reuptake. An example of this is agomelatine, a melatonin MT₁ and MT₂ receptor agonist and a 5-HT_{2C} receptor antagonist [99]. 5-HT is catabolized into melatonin in the pineal gland via a two-step enzymatic process [100]. Dysregulation in 5-HT production and catabolism may affect melatonin levels and thus be a cause of the sleep disturbances often observed in depressed patients. Agomelatine bypasses melatonin production by targeting MT₁ and MT₂ receptors and affects the 5-HT system directly by antagonizing 5-HT_{2C} receptors. The drug was developed in the 1990s and has been approved in several regions, including EU, South Africa, Australia and Canada for the treatment of MDD. A phase 3 clinical program was also initiated in the US, but then discontinued possibly due to a potential requirement of liver function monitoring in patients taking the drug [101].

An extensive preclinical and clinical program has been conducted on agomelatine and almost 500 research papers and multiple reviews have been published. Preclinically, agomelatine has been shown to resynchronize disrupted circadian rhythms in rodents, by activating the MT₁ and MT₂ receptors in the suprachiasmatic nucleus (SCN) and by inhibiting the 5-HT_{2C} receptors (reviewed in [99,102]). Agomelatine has demonstrated antidepressant-like effects in various classical animal models of depression, including the forced swim test [103], chronic mild stress [104] and learned helplessness [105]. Again, in several of these models the antidepressant effect depended on its combined action at MT receptors and antagonism at 5-HT_{2C} receptors [103,105]. Acute treatment with agomelatine was shown to increase extracellular levels of DA and NE without increasing 5-HT levels in the PFC [106]. On the other hand, chronic treatment with agomelatine increased neuronal firing of both dopaminergic cells in the ventral tegmental area and of serotonergic cells in the DRN and thereby enhanced DA and 5-HT release [107], possibly via indirect connections of the SCN with midbrain monoaminergic nuclei [108]. Taken together, these results link agomelatine's mechanism of action to the modulation of monoaminergic transmission, but also differentiate its profile from those of SSRIs and SNRIs.

The above mentioned pharmacological properties of agomelatine were evident in the clinic. In multiple clinical trials, including three phase 2 double-blind placebo-controlled studies and several studies with active references, agomelatine was efficacious in reducing depressive symptoms, improving sleep quality and re-setting disrupted circadian rhythms [109]. Furthermore, it showed an ability to address anhedonia [110] and emotional blunting [111]. Agomelatine was well tolerated with no acute serotonergic side effects [112], lack of discontinuation syndrome [113] and minimal sexual dysfunction [114]. The main adverse effect was elevated levels of serum transaminase in some of the subjects, which might be indicative of liver damage [115]. In summary, agomelatine represents a novel and different mechanism for treating MDD with substantial positive preclinical and clinical data, but its approval and use have been quite limited. Intriguingly, no other compounds with a similar mechanism of action have been brought to the clinic. However, since agomelatine's profile is different from those observed with SSRIs and SNRIs [116], mechanisms underlying its responses and interactions between the melatonin and 5-HT systems perhaps should be explored further in antidepressant research.

3. Beyond monoamines – the glutamate track

3.1. Ketamine and other NMDA receptor modulators

Recent interest in the role of glutamatergic transmission in depression came from the seminal clinical study of Berman et al.

demonstrating that a single intravenous dose of the NMDA open channel blocker ketamine can produce immediate and long-lasting antidepressant effects in patients with TRD [10]. Follow-up studies confirmed this observation and now ketamine is used as a treatment option for difficult-to-treat MDD patients [117]. Despite its efficacy in difficult-to-treat patients, clinical use of ketamine is limited, mainly due to its psychotomimetic adverse effects and abuse liability [118]. Thus, considerable efforts have been made to elucidate ketamine's mechanism of action and to develop drugs with a similar clinical efficacy, but with a more favorable safety profile. Several such compounds are currently being investigated in the clinic and are discussed at the end of this section.

Ketamine is a non-competitive open-channel blocker of glutamatergic NMDA receptors. It binds to NMDA receptors in their open state, gets trapped inside the channel pore after the receptor closes, and slowly dissociates after the receptor is reactivated by its endogenous ligand [119]. A prominent hypothesis is that ketamine preferentially targets NMDA receptors located on GABAergic interneurons because these cells are more metabolically active than pyramidal neurons and have a higher probability of NMDA receptors being in an open state. Blockade of NMDA receptors by ketamine would prevent interneurons from firing which in turn would disinhibit pyramidal cells producing a targeted burst of glutamate in the brain [120]. In support of this hypothesis, the microdialysis study by Moghaddam et al. demonstrated that administration of ketamine causes a rapid and transient increase in extracellular glutamate in the medial PFC in rats [121]. The glutamate burst is thought to mediate the antidepressant effects of ketamine, in part by activating intracellular pathways relevant for neuronal plasticity and synaptogenesis [120,122–124]. For instance, Li et al. showed that treatment with ketamine enhances signaling through post-synaptic α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors and increases the number of dendritic spines in cortical layer V pyramidal neurons in the PFC [123]. Ketamine also reverses deficits in the spine number and reduces anhedonia produced by exposure to chronic unpredictable stress [124]. These effects are at least in part mediated by activation of the mTOR signaling pathway and release of brain-derived neurotrophic factor (BDNF), both of which have been linked to the formation of new synapses [122,123]. However, ketamine has also been shown to activate the immediate early activity gene *Arc* (activity regulated cytoskeleton associated protein) [125], which is known to down-regulate expression of AMPA receptors [126]. Thus, ketamine might not only be increasing synaptic activity, but rather normalizing it and bringing it to an optimal level.

In summary, preclinical research has started to provide insights into the molecular mechanisms of action of ketamine. However, ketamine has also been shown to produce antidepressant-like behavioral responses in mice lacking NMDA receptors in parvalbumin positive interneurons [127]. This study questions the dependence of ketamine's effect on the blockade of NMDA receptors on GABAergic cells. Furthermore, ketamine does not produce antidepressant effects in 5-HT deprived animals [128], suggesting that 5-HT tone is required for ketamine's antidepressant activity. Additional targets of ketamine, such as sigma receptors [129], monoamine transporters [130], and 5-HT_{1B} receptors [131], as well as pharmacological activities of ketamine's metabolites, might collectively contribute to its antidepressant effect. Thus, additional research is needed to elucidate ketamine's mechanism of action.

Clinical data on the efficacy of ketamine in depression came mostly from small open-label studies and case reports, with only a few crossover studies [10,132,133]. These studies reported a rapid acting antidepressant effect of ketamine in difficult-to-treat or TRD patients. A single intravenous infusion of a low sub-anesthetic dose

of ketamine produced a rapid antidepressant response within 2–4 h and this response was sustained for several days or even weeks [133–135]. In about 50–80% of patients, depressive symptoms and even suicidal ideation weakened after a single infusion of ketamine, but repeated administration of the drug was required to maintain its effect [135]. Blier et al. have recently reported that several monthly infusions with ketamine can lead to remission without the development of addiction or other adverse effects [136]. However, the consequences of long-term treatment with ketamine are still largely unknown which limits its clinical use. The (S)-enantiomer of ketamine, esketamine, is currently being tested in phase 2 clinical trials in MDD after both intravenous and intranasal administration, but no data on its efficacy have been reported yet according to <https://clinicaltrials.gov>.

As mentioned in the beginning of the section, ketamine's psychotomimetic adverse effects and abuse liability largely prevent its use in the general population of MDD patients. Thus, several strategies have been put forward to develop NMDA receptor modulators with the rapid ketamine-like antidepressant effect, but a better tolerability. One such strategy has been to develop compounds with a faster dissociation rate, resulting in a lower amount of trapping inside the NMDA channel [119]. An example of this is lanicemine (AZD6765), initially developed for the treatment of stroke. Lanicemine's efficacy in TRD was recently assessed in two small clinical trials [137,138]. Since the trapping rate of NMDA open channel blockers is thought to be correlated with psychotomimetic and dissociative effects [119], it was hypothesized that lanicemine would exhibit ketamine-like antidepressant efficacy without psychotomimetic symptoms. As predicted, lanicemine did not induce psychotomimetic or dissociative effects in clinical trials [137,138]. However, a single infusion of lanicemine failed to produce a sustained antidepressant effect in TRD patients [138]. Interestingly, in the study by Sanacora et al., repeated dosing with lanicemine (three times a week for three weeks) had a sustained antidepressant response after one to two weeks of treatment [137]. Thus, additional research with multiple dosing regimens is needed to investigate the efficacy of lanicemine in TR MDD.

Memantine is another low-trapping NMDA receptor antagonist that has been investigated for antidepressant activity. Memantine is approved for the treatment of moderate-to-severe Alzheimer's disease. However, it did not show a significant antidepressant response in patients with MDD in one clinical study [139] where memantine was administered orally over several weeks. It would be interesting to investigate whether an intravenous route of administration of memantine, which achieves steady state drug levels much faster than the oral route, would be efficacious.

Another strategy to develop an improved ketamine-like agent has been to target selective NMDA receptor subunits. NMDA receptors are tetramers composed of two major subunits, GluN1 and GluN2. There are eight splice variants of GluN1 subunits and four different GluN2 subunits (GluN2A, GluN2B, GluN2C and GluN2D) [140]. Ketamine is a non-selective NMDA receptor antagonist. Based on preclinical work, it was proposed that a selective GluN2B antagonist would retain antidepressant activity without having psychotomimetic effects [117,140]. Two GluN2B antagonists have been tested in the clinic with published results. Traxoprodil (CP-101,606) was tested in a small augmentation study in patients who had responded inadequately to paroxetine. A single intravenous infusion of traxoprodil showed a significant improvement in the paroxetine response five days after the infusion, and the response lasted for one week [141]. Traxoprodil was not tested as a stand-alone therapy. Some psychotomimetic effects were still observed with traxoprodil, but they were reduced when the dose was decreased. Another GluN2B antagonist CERC-301 (MK-0657) failed to show antidepressant efficacy on the

primary outcome measure, but showed significant effects on the secondary measures in one clinical trial [142]. It was a small study of 21 TRD patients and only five patients completed the study. A follow-up phase 2 study with CERC-301 as an adjunctive treatment in TRD has been recently completed, but its results have not been reported yet (<https://clinicaltrials.gov/show/NCT01941043>). It will be interesting to know whether CERC-301 shows antidepressant efficacy in that large clinical trial.

A different way to modulate NMDA receptors is through their glycine co-agonist site. An example of such an approach is GLYX-13 (rapastinel), a tetrapeptide that functions as a partial agonist at the glycine site [143]. GLYX-13 is currently being developed as an adjunctive therapy for MDD. The compound showed antidepressant activity in a phase 2 clinical study, results from which have been published in a review paper [143]. Administration of a single intravenous dose of GLYX-13 in patients resistant to at least one antidepressant agent resulted in improved depression scores without producing psychotomimetic side effects [143]. As with ketamine, the improvement was sustained, lasting for several days. However, there was no dose response relationship and the highest dose tested (30 mg/kg) did not differentiate from placebo. It will be important to know whether these encouraging findings will be replicated in larger clinical programs.

In conclusion, several NMDA receptor modulators have shown antidepressant activity, but these results came from relatively small clinical studies. While promising, these findings need to be confirmed in large, adequately powered clinical trials at doses where the target engagement ideally is ensured. It still remains to be determined whether the NMDA receptor antagonism is the only target for ketamine and whether other NMDA receptor modulators can be as efficacious as ketamine in the treatment of MDD, optimally without producing the psychotomimetic adverse effects.

3.2. Metabotropic glutamate receptor modulators

Another approach that has been taken to modulate glutamate neurotransmission is via the targeting of metabotropic glutamate (mGlu) receptors. The mGlu5 receptors are functionally and physically linked to NMDA receptors and may offer an alternative and possibly a safer way of regulating NMDA receptor function. mGlu5 receptors are also involved in regulation of AMPA receptor internalization, a key mechanism for the regulation of synaptic plasticity which is thought to be important in depression [144]. Another type of mGlu receptors, the mGlu2/3 receptors, are expressed presynaptically and involved in regulation of glutamate release. Two mGlu receptor modulators have been recently brought to the clinic: decogluturant (RG1578), an mGlu2 receptor negative allosteric modulator (NAM), and basimgluturant (RG7090), an mGlu5 receptor NAM, for the treatment of depression and TRD, respectively. Results from the first clinical study with basimgluturant as an adjunctive treatment to an SSRI have been recently reported [145]. Although the primary outcome measure was not met in this study, adjunctive treatment with basimgluturant showed antidepressant effect in several secondary measures and needs to be explored further.

4. Other target profiles with clinical validation data

Several other mechanisms have been investigated in clinical trials of MDD, but unfortunately with limited success. Extensive literature suggesting a link between stress and depression has prompted the so-called "stress hypothesis of depression", and substantial efforts have been made to identify drugs that can modulate the HPA-axis for the treatment of MDD [146,147] (Table 1). One theory has been that stress-induced dysregulation of the HPA-axis leads to depressive episodes via loss of synapses in

the PFC and hippocampus and reduction in neuroplasticity [148]. Mifepristone, an antagonist of glucocorticoid receptors that are activated during stress, has been investigated in a comprehensive clinical program for psychotic depression for more than 10 years [149]. However, due to disappointing clinical results the program was recently discontinued [150].

Based on clinical research showing that some MDD patients have a hyperactive adrenal gland, exhibit elevated responses to stress, and have increased levels of CRF in the cerebrospinal fluid, it was hypothesized that CRF₁ receptor antagonists could be efficacious in the treatment of MDD [146]. Several CRF₁ receptor antagonists have been tested in the clinic. The first small open-label study with the CRF₁ receptor antagonist NBI-30775/R121919 had positive results [151]. However, a later double-blind randomized placebo-controlled phase 2 study with the CRF₁ receptor antagonist CP-316,311 failed to demonstrate its efficacy in MDD [152]. As a result, several drug companies removed CRF₁ receptor antagonists from their drug pipelines. Thus, despite of a strong preclinical rationale and the availability of biomarkers to ensure CRF₁ receptor target engagement [152,153], CRF₁ receptor antagonists have failed to live up to their promise in the clinic.

There are several possible reasons for the failure of these programs. One obvious reason could be that the disease hypothesis has proven to be wrong, and that CRF hyper secretion is not directly associated with MDD. Alternatively, given the biological heterogeneity within the MDD diagnosis, it is possible that a CRF₁ receptor antagonist could have been efficacious in a well-defined subpopulation of MDD patients selected by applying a CRF specific biomarker.

Other peptide targets that originated from the stress/HPA-axis hypothesis are neurokinin (NK)₁ and NK₂ receptors that are activated by substance P and neurokinin A, respectively. NK₁ receptor antagonists have had conflicting results in clinical trials. Two NK₁ receptor antagonists, aprepitant (MK869) and L759274, reduced depressive symptoms in two PoC placebo-controlled randomized double-blind phase 2 clinical trials [154,155]. However, the efficacy of aprepitant was not confirmed in five subsequent phase 2 studies [156] and the program was discontinued. Several other NK₁ receptor antagonists, including orvepitant (GW823296) and casopitant (GW679769), were also tested for the treatment of MDD. The phase 2 study with orvepitant was terminated in order to assess the incidence of isolated seizure events [157]. Casopitant showed efficacy in one of the two clinical trials, but only at the highest dose, which translated to >99% occupancy at the target [158]. Finally, the NK₂ receptor antagonist saredutant (SR48968) was tested in ten clinical trials, either as stand-alone therapy, or in combination with paroxetine (<http://clinicaltrials.gov/show/NCT00629551>) or escitalopram (<http://clinicaltrials.gov/show/NCT00531622>). Results from these clinical trials have not been published, but the development program was discontinued in 2009 [159].

Hence, despite substantial investments in these biological targets by several pharmaceutical companies, NK receptor antagonists did not yield new antidepressant treatments. As discussed with the CRF₁ receptor antagonist programs, it cannot be ruled out that a subset of MDD patients identified by means of an appropriate biomarker might have benefited from drugs with these mechanisms. However, given the modulatory role of neuropeptides on neurotransmission, one possibility is that compounds that selectively target a peptide receptor are insufficient to treat MDD due to biological redundancies in the system. This idea was explored using NK receptor antagonism as an adjunctive treatment to an SSRI, but so far with limited success. A recently published study of aprepitant with paroxetine vs. each treatment alone did not show any advantages for the combination in MDD patients [160].

Other neuropeptide targets that have been brought forward for evaluation in the clinic are vasopressin and orexin receptor antagonists [161,162]. The orexin receptor antagonist florexant (MK-6096) was tested as an adjunctive therapy in patients responding inadequately to an SSRI/SNRI treatment. However, this clinical trial was terminated with no published results (<https://clinicaltrials.gov/show/NCT01554176>). A clinical trial with the vasopressin-1B receptor antagonist ABT-436 was also terminated according to <https://clinicaltrials.gov> due strategic decisions not associated with safety concerns (<https://clinicaltrials.gov/show/NCT01741142>). Thus, targeting neuropeptide receptors has so far failed to deliver new antidepressant treatments.

Several cholinergic mechanisms have been explored for the treatment of MDD based on the cholinergic hypothesis of depression [163] (Table 1). The hypothesis was founded on the clinical observation that cholinomimetics can precipitate depression and on pharmacological studies in animals suggesting that the state of depression/mania is associated with a disturbed balance between cholinergic and adrenergic brain activity [163]. The muscarinic cholinergic antagonist scopolamine has been recently tested as adjunct treatment or monotherapy in clinical trials of MDD [164–166]. It showed a rapid antidepressant response comparable to that of ketamine. Interestingly, scopolamine's antidepressant activity was suggested to be mediated via increased neuroplasticity, which would mechanistically converge with the proposed downstream effects of ketamine [164]. As expected, scopolamine produced typical anticholinergic adverse side effects such as dry mouth, blurred vision and drowsiness [166], which imply a limitation to its broad clinical use.

Another cholinergic mechanism that has been explored for the treatment of MDD relied on the modulation of nicotinic cholinergic receptors [167]. Both nicotinic antagonists and partial agonists have been tested in clinical trials (Table 1), but with limited success. While the first clinical study of the α 4 β 2 nicotinic cholinergic receptor antagonist TC-5214 (the S-enantiomer of mecamylamine) as an add-on treatment to SSRIs was promising, subsequent clinical studies failed to reproduce this finding and the development program was stopped [168].

The close relation between depressed mood and hedonic deficits has led to drug profiles that target the reward circuitry. The triple reuptake inhibitor programs discussed in Section 2.2 tie into this hypothesis via their DA enhancing properties of DAT inhibitors. Another approach that has recently obtained substantial interest is kappa opioid antagonists (Table 1). Stress-induced increase of dynorphin, the endogenous ligand for the kappa opioid receptor, has been found to cause dysphoria in humans [169]. Furthermore, preclinical studies have demonstrated that dynorphin can reduce DA release in the nucleus accumbens and thereby cause anhedonia-like behavior [169]. ALKS-5461, a combination of buprenorphine (partial μ opioid receptor agonist, kappa opioid receptor antagonist) and samidorphan (μ opioid antagonist), has shown positive results in a phase 2 study [170] and is currently in phase 3 as an augmentation therapy for the treatment of TRD (<https://clinicaltrials.gov/show/NCT02158533>). The strategy behind ALKS-5461 has been to reduce the μ opioid receptor mediated euphoric effect of buprenorphine by combining it with the μ opioid receptor antagonist samidorphan, and thus achieve the antidepressant efficacy through kappa opioid antagonism. A recently published study in MDD patients that investigated different ratios between the two compounds indicates that a 1:1 ratio may be optimal [171]. The positive results reported in a clinical trial of ALKS-5461 are very encouraging and it will be important to know whether they will repeat in the larger ongoing clinical program.

Neuroinflammation, another biology with linkage to MDD, has been an area of interest for some time. The interest has been fueled by several studies demonstrating that depressed patients have

increased serum levels of pro-inflammatory cytokines, notably IL-6 and TNF- α [172], and dysregulation of the kynurenic pathway [173]. Cyclooxygenase-2 (COX-2) inhibitors that block production of prostaglandine E(2) and pro-inflammatory cytokines have been investigated in several clinical trials of MDD. The most studied drug celecoxib showed antidepressant efficacy as an add-on therapy to reboxetine [174], fluoxetine [175] and sertraline [176] in three small double-blind placebo controlled trials. In one study, the adjunctive treatment with celecoxib reduced IL-6 serum levels in depressed patients, and there was a significant correlation between the reduction in Hamilton Depression Rating Scale scores and IL-6 serum levels after six weeks of treatment [176]. In preclinical research, celecoxib was shown to potentiate the effects of reboxetine and fluoxetine on cortical NE and 5-HT levels in rats which might provide mechanistic support for its antidepressant efficacy [177].

In addition to combined treatments with COX-2 inhibitors, the potential of a TNF- α antibody as monotherapy has been assessed in TRD patients in one clinical trial [178]. In that study, the TNF- α antibody infliximab did not separate from placebo, but did show a trend toward antidepressant activity in patients with elevated levels of inflammatory biomarkers at baseline [178]. Thus, this PoC study suggests that although therapies targeting TNF- α might not have generalized efficacy in TRD, they could be used to treat patients with high baseline inflammation. Taken together, these results indicate that anti-inflammatory agents should be further investigated as treatment options for MDD, especially in patients with co-morbid inflammatory diseases and high levels of inflammatory cytokines.

5. Target profiles with preclinical validation only

A substantial number of drug target profiles that originated from the above discussed hypotheses of depression have been under preclinical evaluation (Table 1). Unfortunately, there has been a low translatability of these studies to the clinic. This could partly be due to the fact that the majority of preclinical models rely on behavioral changes induced by only applying physical stressors.

In spite of the long-lasting interest in monoaminergic mechanisms, preclinical research is still ongoing in that field. An interesting, novel approach has been to refine the action of monoaminergic compounds based on the concept of selective signaling bias. This concept originates from the observations that a given receptor can couple to different intracellular effector systems depending on the receptor localization, and that compounds with selectivity for a given effector system can be found. One such example is the 5-HT_{1A} receptor agonist F15599 which stimulates postsynaptic 5-HT_{1A} receptors while having no effect on 5-HT_{1A} somatodendritic autoreceptors [179]. As discussed in Section 2.1.1, this profile would predict a rapid antidepressant effect since it would not dampen neuronal firing in the DRN, but selectively stimulate 5-HT_{1A} receptors in terminal areas including the cortex and hippocampus. Another novel and not so well explored monoaminergic mechanism is to target the intracellularly located presynaptic traceamine-associated receptor-1 (TAAR1), which is a key regulator of the release of brain monoamines [180]. Several TAAR1 agonists have shown antidepressant-like activity in preclinical models of depression and schizophrenia [180], but they have not been tested in the clinic.

As discussed in Section 3, the clinical research that emerged from the glutamate hypothesis of depression has been primarily focused on NMDA receptor modulation. The preclinical research has explored additional mechanisms, including targeting of AMPA receptors. Whereas enhanced AMPA receptor signaling plays a key role for the induction of neuroplasticity, it is also clear that direct stimulation of AMPA receptors can be neurotoxic [181]. One

approach to try to circumvent the toxicity issue has been to develop positive allosteric modulators, also known as AMPAkinases. However, their success has been limited, in part due to low bioavailability and toxicity issues for some of the compounds [182,183]. Thus, it remains to be demonstrated if AMPAkinases can have a potential as antidepressants.

There is growing evidence that the endocannabinoid (endogenous cannabinoid) system plays a critical role in coping with stress-related disorders, such as depression. For instance, the endocannabinoid system is closely linked to the glucocorticoid system. Preclinical literature suggests that facilitation of cannabinoid signaling, either through stimulation of cannabinoid-1 receptors (CB1) or by inhibition of the endocannabinoid degrading enzyme fatty acid amide hydrolase (FAAH) can produce antidepressant-like effects [184]. Furthermore, the CB1 receptor inverse agonist rimonabant, which was developed and approved for weight loss, had to be subsequently withdrawn from the market due to risk of severe depression and suicide [185]. There have been positive clinical observations on the antidepressant efficacy of cannabinoids, such as tetrahydrocannabinol or cannabidiol. It will be interesting to know whether these findings can be replicated in larger well-controlled clinical trials or by using selective CB1 receptor agonists and/or FAAH inhibitors.

Another emerging area in drug discovery research has been to target mechanisms underlying neuronal plasticity [186]. It originates from the evidence that neuronal plasticity is important for the recovery from depression [187]. BDNF and its receptor TrkB (tropomyosin receptor kinase B) have been investigated extensively in this regard, and positive modulators of the TrkB receptor have been hypothesized to have antidepressant activity [188,189]. Thus, there are several ongoing efforts to identify small molecules with selectivity for the TrkB receptor [190]. However, there are also preclinical reports of depressogenic-like effects of BDNF via its negative modulation of the reward circuitry [191]. These findings question whether a TrkB receptor agonist is the optimal profile for an antidepressant, or whether an antagonist or partial agonist would be more appropriate. Thus, more work is needed to explore the opportunities for new antidepressant mechanisms from this line of research.

Finally, another potentially interesting mechanism for which no CNS drug-like molecules are available yet is chromatin remodeling through histone deacetylase inhibition. Histone deacetylase 2 (HDAC2) inhibitors have shown antidepressant-like effects and pro-cognitive actions in rodents [192–194]. Furthermore, an increase in H3 acetylation and decreased levels of HDAC2 in the nucleus accumbens have been observed in postmortem brain tissue from depressed patients [194]. Therefore, HDAC inhibitors should be explored further as potential targets for antidepressants, especially once centrally penetrable tool compounds become available [19].

6. Conclusions and future directions

Targeting the monoamine system has hitherto been the most successful approach to treat MDD. Going beyond inhibition of MAO and monoamine transporters has, in spite of some disappointments, produced novel and differentiated antidepressant therapies, including agomelatine, vilazodone and vortioxetine, as well add-on therapy with atypical antipsychotics. Research on glutamate targets may hold promise, but is still in its early stages and much more work is needed before we will know whether this is a viable approach for treating MDD in the general population or whether it will be limited to the use of ketamine in a subpopulation of TRD patients. The preclinical observation that ketamine's rapid antidepressant-like activity is 5-HT-dependent [128] suggests that a better understanding of the interface between the 5-HT and

glutamate systems is needed in MDD. It could be a valuable research strategy to pursue clinically by testing of a combination of approved drugs that each target either the glutamate or 5-HT systems. Several approaches (e.g., CRF, NK receptor antagonists) have largely failed in the clinic; however these failures have provided important knowledge about MDD. While these mechanisms may be important in the etiology of depression, targeting them in isolation is probably not sufficient to treat the disorder. Kappa opioid antagonists and neuroplasticity related targets that are coming out of preclinical research offer promising opportunities for the treatment of MDD, but additional research is needed to validate these targets. It is possible that all of these apparently distinct antidepressant target profiles ultimately converge on the modulation of glutamate neurotransmission and its downstream neuronal plasticity (Fig. 1). However, there are large gaps in our understanding of the relation between glutamate physiology and MDD, and a substantial amount of empirical data is needed to address this hypothesis.

A rather obvious, but to some extent still neglected issue in antidepressant research is of the importance of ensuring the presence of therapeutic drug levels at relevant targets in the brain. There have been multiple instances in which projects were terminated because drugs were not tested at high enough concentrations to reach therapeutic levels [195]. To overcome this issue, an integrated understanding of plasma exposure, target engagement and pharmacodynamic response is gaining momentum in modern drug development. However, depending on the target, for example the NMDA receptors, there are still methodological limitations to this approach, including difficulties in finding PET ligands [196].

A shortening of the time to obtain antidepressant efficacy would be a major achievement in antidepressant research. However, antidepressant drugs are often selected to allow for once a day oral dosing, which implies an elimination half-life of 24 h or more. This may by default set a limit to the time to onset of a therapeutic effect, at least if it is assumed that the therapeutic steady-state drug level is important for the drug's therapeutic action at the target. Thus, ways to accelerate steady-state drug levels and increase the initial exposure (e.g., via intravenous bolus injection or intranasal application followed by oral maintenance) may potentially provide faster relief of depressive symptoms and should be clinically explored.

Even though monotherapy has been the goal for antidepressant drug development in general, clinical practice shows the frequent use of adjunctive therapy (pharmacological or non-pharmacological) and several add-on therapies with atypical antipsychotics have recently been approved for MDD. This strategy should be further pursued during preclinical and clinical drug development, similar to what is done in epilepsy and attention deficit hyperactivity disorder. One example could be an adjunctive treatment with N-acetyl cysteine, which is a powerful antioxidant and an activator of the cystine–glutamate antiporter [197]. It has shown antidepressant-like properties in rodents [198] and clinical efficacy in bipolar and unipolar depression [199,200]. Another example is inhibitors of the recently described low affinity, high capacity transporters for 5-HT in the brain, i.e. organic cation transporters (OCTs) and plasma membrane monoamine transporters (PMATs) [201]. These transporters are involved in the uptake of 5-HT when extracellular 5-HT levels are high and, as shown in preclinical studies, can limit the ability of SSRIs to increase 5-HT. This field of research is still ongoing and to our knowledge, no OCT and PMAT inhibitors have been tested in the clinic. Needless to say, pharmacological adjunctive treatment imposes a need to address possible drug–drug interactions early on.

Improvement in response rates remains the major challenge in antidepressant research. This might be accomplished through a

better understanding of the biological mechanisms underlying the heterogeneity of MDD. Research in human genetics has so far not succeeded in identifying biologically well-defined subpopulations in depression, but it is a rapidly advancing field that at some point is likely to deliver a break-through [202]. Another way forward would be to integrate several clinical research methods to refine drug treatments, for example, by using physiological stress response tests, measuring hormone levels (thyroid, sex hormones) and blood biomarkers, and recording brain circuitry dynamics in depression-relevant brain regions with imaging and electrophysiology techniques. However, this integrated approach may be considered more of a long-term aspirational goal for such a complex and heterogeneous disorder with largely unknown etiologies as MDD.

In the area of drug discovery research, the dogma for many years has been to first identify a drug target and then discover and optimize a druggable molecule. Even though more challenging from a drug screening perspective, there are other approaches to consider, including phenotypic screening to identify compounds that modulate specific brain circuitries or molecules that interact with several targets and/or receptor heterodimers. For instance, it is known that 5-HT₃ receptors form heterodimers with α_4 nicotinic receptors [203], and 5-HT_{2A} receptors form heterodimers with mGlu2 receptors [204]. Furthermore, even for well-known drug targets, new opportunities may appear due to the realization that a given receptor may couple to multiple intracellular effector systems. Compounds can now be made with a selective signaling bias that may allow for the targeting of specific brain areas [179].

Development and refinement of non-pharmacological methods for treatment of depression (e.g. deep brain stimulation, transcranial magnetic stimulation, and cognitive behavioral therapy), which have not been the focus of this review, have made important advances over recent years [205,206]. Combination of pharmacological and non-pharmacological procedures is therefore a promising therapeutic approach that could benefit from larger more systematic studies. Thus, a wealth of possibilities still exists with regard to the development of new drug therapies for the treatment of MDD. However, in the near term these discoveries will likely continue to rely on empirical data and serendipity rather than being based on a thorough understanding of the disease etiology. Hence, basic research in understanding mechanisms of depression should remain a high priority.

Conflict of interest

E. Dale, B. Bang-Andersen and C. Sanchez are full-time employees of Lundbeck.

References

- [1] C. Sanchez, P.B. Bergqvist, L.T. Brennum, S. Gupta, S. Hogg, A. Larsen, et al., Escitalopram, the S-(+)-enantiomer of citalopram, is a selective serotonin reuptake inhibitor with potent effects in animal models predictive of antidepressant and anxiolytic activities, *Psychopharmacology (Berl.)* 167 (2003) 353–362.
- [2] M.J. Owens, D.L. Knight, C.B. Nemeroff, Second-generation SSRIs: human monoamine transporter binding profile of escitalopram and R-fluoxetine, *Biol. Psychiatry* 50 (2001) 345–350.
- [3] C. Sanchez, E. Meier, Behavioral profiles of SSRIs in animal models of depression, anxiety and aggression. Are they all alike? *Psychopharmacology (Berl.)* 129 (1997) 197–205.
- [4] A. Cipriani, M. Koesters, T.A. Furukawa, M. Nose, M. Purgato, I.M. Omori, et al., Duloxetine versus other anti-depressive agents for depression, *Cochrane Database Syst. Rev.* 10 (2012) Cd006533.
- [5] C. Sanchez, E.H. Reines, S.A. Montgomery, A comparative review of escitalopram, paroxetine, and sertraline: are they all alike, *Int. Clin. Psychopharmacol.* 29 (2014) 185–196.
- [6] A.J. Rush, M.H. Trivedi, S.R. Wisniewski, A.A. Nierenberg, J.W. Stewart, D. Warden, et al., Acute and longer-term outcomes in depressed outpatients requiring one or several treatment steps: a STAR*D report, *Am. J. Psychiatry* 163 (2006) 1905–1917.

- [7] H.D. Anderson, W.D. Pace, A.M. Libby, D.R. West, R.J. Valuck, Rates of 5 common antidepressant side effects among new adult and adolescent cases of depression: a retrospective US claims study, *Clin. Ther.* 34 (2012) 113–123.
- [8] J.J. Schildkraut, The catecholamine hypothesis of affective disorders: a review of supporting evidence, *Am. J. Psychiatry* 122 (1965) 509–522.
- [9] A. Carlsson, H. Corrodi, K. Fuxe, T. Hokfelt, Effect of antidepressant drugs on the depletion of intraneuronal brain 5-hydroxytryptamine stores caused by 4-methyl-alpha-ethyl-meta-tyramine, *Eur. J. Pharmacol.* 5 (1969) 357–366.
- [10] R.M. Berman, A. Cappiello, A. Anand, D.A. Oren, G.R. Heninger, D.S. Charney, et al., Antidepressant effects of ketamine in depressed patients, *Biol. Psychiatry* 47 (2000) 351–354.
- [11] G. Sanacora, G. Treccani, M. Popoli, Towards a glutamate hypothesis of depression: an emerging frontier of neuropsychopharmacology for mood disorders, *Neuropharmacology* 62 (2012) 63–77.
- [12] J. Zohar, D.J. Nutt, D.J. Kupfer, H.J. Moller, S. Yamawaki, M. Spedding, et al., A proposal for an updated neuropsychopharmacological nomenclature, *Eur. Neuropsychopharmacol.* 24 (2014) 1005–1014.
- [13] F. Artigas, A. Adell, P. Celada, Pindolol augmentation of antidepressant response, *Curr. Drug Targets* 7 (2006) 139–147.
- [14] A.F. Carvalho, J.R. Machado, J.L. Cavalcante, Augmentation strategies for treatment-resistant depression, *Curr. Opin. Psychiatry* 22 (2009) 7–12.
- [15] F. Lopes Rocha, C. Fuzikawa, R. Riera, M.G. Ramos, C. Hara, Antidepressant combination for major depression in incomplete responders – a systematic review, *J. Affect. Disord.* 144 (2013) 1–6.
- [16] M.J. Millan, Multi-target strategies for the improved treatment of depressive states: conceptual foundations and neuronal substrates, drug discovery and therapeutic application, *Pharmacol. Ther.* 110 (2006) 135–370.
- [17] M.J. Millan, Dual- and triple-acting agents for treating core and co-morbid symptoms of major depression: novel concepts, new drugs, *Neurotherapeutics: J. Am. Soc. Exp. Neurother.* 6 (2009) 53–77.
- [18] M.J. Millan, The role of monoamines in the actions of established and novel antidepressant agents: a critical review, *Eur. J. Pharmacol.* 500 (2004) 371–384.
- [19] M.J. Millan, On 'polypharmacy' and multi-target agents, complementary strategies for improving the treatment of depression: a comparative appraisal, *Int. J. Neuropsychopharmacol.: Off. Sci. J. Coll. Int. Neuropsychopharmacol.* 17 (2014) 1009–1037.
- [20] F. Artigas, V. Perez, E. Alvarez, Pindolol induces a rapid improvement of depressed patients treated with serotonin reuptake inhibitors, *Arch. Gen. Psychiatry* 51 (1994) 248–251.
- [21] M.C. Scorza, L. Llado-Pelfort, S. Oller, R. Cortes, D. Puigdemont, M.J. Portella, et al., Preclinical and clinical characterization of the selective 5-HT_{1A} receptor antagonist DU-125530 for antidepressant treatment, *Br. J. Pharmacol.* 167 (2012) 1021–1034.
- [22] F. Artigas, Serotonin receptors involved in antidepressant effects, *Pharmacol. Ther.* 137 (2013) 119–131.
- [23] L.J. Boothman, S.N. Mitchell, T. Sharp, Investigation of the SSRI augmentation properties of 5-HT₂ receptor antagonists using in vivo microdialysis, *Neuropharmacology* 50 (2006) 726–732.
- [24] C. Han, S.M. Wang, M. Kato, S.J. Lee, A.A. Patkar, P.S. Masand, et al., Second-generation antipsychotics in the treatment of major depressive disorder: current evidence, *Expert Rev. Neurother.* 13 (2013) 851–870.
- [25] R.M. Berman, R.N. Marcus, R. Swanink, R.D. McQuade, W.H. Carson, P.K. Corey-Lisle, et al., The efficacy and safety of aripiprazole as adjunctive therapy in major depressive disorder: a multicenter, randomized, double-blind, placebo-controlled study, *J. Clin. Psychiatry* 68 (2007) 843–853.
- [26] M. Bauer, H.W. Pretorius, E.L. Constant, W.R. Earley, J. Szamosi, M. Brecher, Extended-release quetiapine as adjunct to an antidepressant in patients with major depressive disorder: results of a randomized, placebo-controlled, double-blind study, *J. Clin. Psychiatry* 70 (2009) 540–549.
- [27] M.E. Thase, S.A. Corya, O. Osuntokun, M. Case, D.B. Henley, T.M. Sanger, et al., A randomized, double-blind comparison of olanzapine/fluoxetine combination, olanzapine, and fluoxetine in treatment-resistant major depressive disorder, *J. Clin. Psychiatry* 68 (2007) 224–236.
- [28] D. Bakish, Y.D. Lapiere, R. Weinstein, J. Klein, A. Wiens, B. Jones, et al., Risperidone, imipramine, and placebo in the treatment of dysthymic disorder, *J. Clin. Psychopharmacol.* 13 (1993) 409–414.
- [29] G.J. Marek, R. Martin-Ruiz, A. Abo, F. Artigas, The selective 5-HT_{2A} receptor antagonist M100907 enhances antidepressant-like behavioral effects of the SSRI fluoxetine, *Neuropsychopharmacology* 30 (2005) 2205–2215.
- [30] Otsuka and Lundbeck submit New Drug Application for brexpiprazole for the treatment of schizophrenia and as adjunctive therapy for the treatment of major depression, Press release 14 July 2014. <http://investor.lundbeck.com/releasedetail.cfm?ReleaseID=8593572014>.
- [31] K. Maeda, H. Sugino, H. Akazawa, N. Amada, J. Shimada, T. Futamura, et al., Brexpiprazole I: in vitro and in vivo characterization of a novel serotonin-dopamine activity modulator, *J. Pharmacol. Exp. Ther.* 350 (2014) 589–604.
- [32] B. Kiss, A. Horvath, Z. Nemethy, E. Schmidt, I. Laszlovszky, G. Bugovics, et al., Cariprazine (RGH-188), a dopamine D(3) receptor-preferring, D(3)/D(2) dopamine receptor antagonist-partial agonist antipsychotic candidate: in vitro and neurochemical profile, *J. Pharmacol. Exp. Ther.* 333 (2010) 328–340.
- [33] Forest Laboratories, Inc; Gedeon Richter, Plc announce positive Phase IIb topline results for cariprazine as adjunctive therapy in the treatment of major depressive disorder, Press release 21 March 2014. <http://investor.frx.com/press-release/r-and-d-news/forest-laboratories-inc-and-gedeon-richter-plc-announce-positive-phase-ii2014>.
- [34] A.G. Wade, G.M. Crawford, C.B. Nemeroff, A.F. Schatzberg, T. Schlaepfer, A. McConnell, et al., Citalopram plus low-dose pipamperone versus citalopram plus placebo in patients with major depressive disorder: an 8-week, double-blind, randomized study on magnitude and timing of clinical response, *Psychol. Med.* 41 (2011) 2089–2097.
- [35] Lilly announces Edivoxetine did not meet primary endpoint of Phase III clinical studies as add-on therapy for major depressive disorder, Press release 5 December 2013. <https://investor.lilly.com/releasedetail.cfm?releaseid=8117512013>.
- [36] Shire reports top-line results from two Phase 3 studies for Vyvanser (lisdexamfetamine dimesylate) capsules (CII) as an adjunctive treatment for adults with major depressive disorder. <http://www.shire.com/shireplc/en/investors/investorsnews/irshirennews?id=9212014>.
- [37] G. Stotz, B. Woggon, J. Angst, Psychostimulants in the therapy of treatment-resistant depression Review of the literature and findings from a retrospective study in 65 depressed patients, *Dialogues Clin. Neurosci.* 1 (1999) 165–174.
- [38] P. Skolnick, P. Popik, A. Janowsky, B. Beer, A.S. Lippa, Antidepressant-like actions of DOV 21,947: a triple reuptake inhibitor, *Eur. J. Pharmacol.* 461 (2003) 99–104.
- [39] Euthymics reports top-line results from TRIADE trial of amitifadine for major depressive disorder. <http://euthymics.com/press-releases/euthymics-reports-top-line-results-from-triade-trial-of-amitifadine-for-major-depressive-disorder/2013>.
- [40] S. Learned, O. Graff, S. Roychowdhury, R. Moate, K.R. Krishnan, G. Archer, et al., Efficacy, safety, and tolerability of a triple reuptake inhibitor GSK372475 in the treatment of patients with major depressive disorder: two randomized, placebo- and active-controlled clinical trials, *J. Psychopharmacol. (Oxf.)* 26 (2012) 653–662.
- [41] R. Zhang, X. Li, Y. Shi, Y. Shao, K. Sun, A. Wang, et al., The effects of LPM570065, a novel triple reuptake inhibitor, on extracellular serotonin, dopamine and norepinephrine levels in rats, *PLOS ONE* 9 (2014) e91775.
- [42] Lundbeck announced headline result. Lu AA24530 shows positive results in major depressive disorder phase II study, Press release 2 July 2009. <http://investor.lundbeck.com/releasedetail.cfm?ReleaseID=6086192009>.
- [43] T. Heinrich, H. Bottcher, R. Gericke, G.D. Bartoszyk, S. Anzali, C.A. Seyfried, et al., Synthesis and structure-activity relationship in a class of indolebutylpiperazines as dual 5-HT_{1A} receptor agonists and serotonin reuptake inhibitors, *J. Med. Chem.* 47 (2004) 4684–4692.
- [44] L.A. Dawson, The discovery and development of vilazodone for the treatment of depression: a novel antidepressant or simply another SSRI, *Expert Opin. Drug Discov.* 8 (2013) 1529–1539.
- [45] Z.A. Hughes, K.R. Starr, C.J. Langmead, M. Hill, G.D. Bartoszyk, J.J. Hagan, et al., Neurochemical evaluation of the novel 5-HT_{1A} receptor partial agonist/serotonin reuptake inhibitor, vilazodone, *Eur. J. Pharmacol.* 510 (2005) 49–57.
- [46] C. van Amsterdam, C.A. Seyfried, Mechanism of action of the bimodal antidepressant vilazodone: evidence for serotonin_{1A}-receptor-mediated auto-augmentation of extracellular serotonin output, *Psychopharmacology (Berl.)* 231 (2014) 2547–2558.
- [47] P. Blier, A. Crnic, M. El Mansari, Effects of acute and sustained administration of the antidepressant vilazodone on monoaminergic systems: in vivo electrophysiological studies, *Neuropsychopharmacology* 38 (2013) S8–S367.
- [48] L. Diaz-Mataix, M.C. Scorza, A. Bortolozzi, M. Toth, P. Celada, F. Artigas, Involvement of 5-HT_{1A} receptors in prefrontal cortex in the modulation of dopaminergic activity: role in atypical antipsychotic action, *J. Neurosci.: Off. J. Soc. Neurosci.* 25 (2005) 10831–10843.
- [49] M. Suzuki, T. Matsuda, S. Asano, P. Somboonthum, K. Takuma, A. Baba, Increase of noradrenaline release in the hypothalamus of freely moving rat by postsynaptic 5-hydroxytryptamine_{1A} receptor activation, *Br. J. Pharmacol.* 115 (1995) 703–711.
- [50] M. Di Mascio, G. Di Giovanni, V. Di Matteo, S. Prisco, E. Esposito, Selective serotonin reuptake inhibitors reduce the spontaneous activity of dopaminergic neurons in the ventral tegmental area, *Brain Res. Bull.* 46 (1998) 547–554.
- [51] Y. Mateo, J.A. Ruiz-Ortega, J. Pineda, L. Ugedo, J.J. Meana, Inhibition of 5-hydroxytryptamine reuptake by the antidepressant citalopram in the locus coeruleus modulates the rat brain noradrenergic transmission in vivo, *Neuropharmacology* 39 (2000) 2036–2043.
- [52] M. Matsumoto, M. Yoshioka, H. Togashi, M. Tochihiro, T. Ikeda, H. Saito, Modulation of norepinephrine release by serotonin receptors in the rat hippocampus as measured by in vivo microdialysis, *J. Pharmacol. Exp. Ther.* 272 (1995) 1044–1051.
- [53] D. Nutt, K. Demyttenaere, Z. Janka, T. Aarre, M. Bourin, P.L. Canonico, et al., The other face of depression, reduced positive affect: the role of catecholamines in causation and cure, *J. Psychopharmacol. (Oxf.)* 21 (2007) 461–471.
- [54] T. Renoir, T.Y. Pang, L. Lanfumey, Drug withdrawal-induced depression: serotonergic and plasticity changes in animal models, *Neurosci. Biobehav. Rev.* 36 (2012) 696–726.
- [55] A.M. Gardier, E. Lepoul, J.H. Trouvin, E. Chanut, M.C. Dessalles, C. Jacquot, Changes in dopamine metabolism in rat forebrain regions after cessation of long-term fluoxetine treatment: relationship with brain concentrations of fluoxetine and norfluoxetine, *Life Sci.* 54 (1994) P151–P156.
- [56] R.A. Sansone, L.A. Sansone, SSRI-induced indifference, *Psychiatry (Edgmont (PA: Township))* 7 (2010) 14–18.
- [57] R.M. Lane, Restoration of positive mood states in major depression as a potential drug development target, *J. Psychopharmacol. (Oxf.)* 28 (2014) 527–535.
- [58] M.E. Page, J.F. Cryan, A. Sullivan, A. Dalvi, B. Saucy, D.R. Manning, et al., Behavioral and neurochemical effects of 5-[4-(5-Cyano-3-indolyl)-butyl]-butyl]-1-piperazinyl]-benzofuran-2-carboxamide (EMD 68843): a combined selective

- inhibitor of serotonin reuptake and 5-hydroxytryptamine(1A) receptor partial agonist, *J. Pharmacol. Exp. Ther.* 302 (2002) 1220–1227.
- [59] T.P. Laughren, J. Gobburu, R.J. Temple, E.F. Unger, A. Bhattaram, P.V. Dinh, et al., Vilazodone: clinical basis for the US Food and Drug Administration's approval of a new antidepressant, *J. Clin. Psychiatry* 72 (2011) 1166–1173.
- [60] J.F. Heiser, C.S. Wilcox, Serotonin 5-HT_{1A} receptor agonists as antidepressants: pharmacological rationale and evidence for efficacy, *CNS Drugs* 10 (1998) 343–354.
- [61] S.M. Wang, C. Han, S.J. Lee, A.A. Patkar, P.S. Masand, C.U. Pae, A review of current evidence for vilazodone in major depressive disorder, *Int. J. Psychiatry Clin. Pract.* 17 (2013) 160–169.
- [62] B. Bang-Andersen, T. Ruhland, M. Jorgensen, G. Smith, K. Frederiksen, K.G. Jensen, et al., Discovery of 1-[2-(2,4-dimethylphenylsulfanyl)phenyl]piperazine (Lu AA21004): a novel multimodal compound for the treatment of major depressive disorder, *J. Med. Chem.* 54 (2011) 3206–3221.
- [63] A. Mork, A. Pehrson, L.T. Brennum, S.M. Nielsen, H. Zhong, A.B. Lassen, et al., Pharmacological effects of Lu AA21004: a novel multimodal compound for the treatment of major depressive disorder, *J. Pharmacol. Exp. Ther.* 340 (2012) 666–675.
- [64] C. Sanchez, K.E. Asin, F. Artigas, Vortioxetine, a novel antidepressant with multimodal activity: review of preclinical and clinical data, *Pharmacol. Ther.* 145 (2015) 43–57.
- [65] A.L. Pehrson, T. Cremers, C. Betry, M.G. van der Hart, L. Jorgensen, M. Madsen, et al., Lu AA21004, a novel multimodal antidepressant, produces regionally selective increases of multiple neurotransmitters – a rat microdialysis and electrophysiology study, *Eur. Neuropsychopharmacol.: J. Eur. Coll. Neuropsychopharmacol.* 23 (2013) 133–145.
- [66] E. Dale, H. Zhang, S.C. Leiser, Y. Xiao, D. Lu, C.R. Yang, et al., Vortioxetine disinhibits pyramidal cell function and enhances synaptic plasticity in the rat hippocampus, *J. Psychopharmacol. (Oxf.)* 28 (2014) 891–902.
- [67] M.S. Riga, P. Celada, C. Sanchez, F. Artigas, Role of 5-HT₃ receptors in the mechanism of action of the investigational antidepressant vortioxetine, *Eur. Neuropsychopharmacol.* 23 (2013) S4–S393.
- [68] M.S. Riga, P. Celada, C. Sanchez, F. Artigas, Cortical and hippocampal microcircuits involved in the mechanism of action of the new antidepressant drug vortioxetine, *Neuropsychopharmacology* 39 (2014) S632.
- [69] M. El Mansari, M. Lecours, P. Blier, Effects of acute and sustained administration of vortioxetine on the serotonin system in the hippocampus: electrophysiological studies in the rat brain, *Psychopharmacology (Berl.)* (2015) (Epub ahead of print).
- [70] L. Westrich, N. Haddjeri, O. Dkhissi-Benyahya, C. Sanchez, Involvement of 5-HT receptors in vortioxetine's modulation of circadian rhythms and episodic memory in rodents, *Neuropharmacology* 89C (2014) 382–390.
- [71] Y. Li, K.F. Raaby, C. Sanchez, M. Gulino, Serotonergic receptor mechanisms underlying antidepressant-like action in the progesterone withdrawal model of hormonally induced depression in rats, *Behav. Brain Res.* 256 (2013) 520–528.
- [72] Y. Li, C. Sanchez, M. Gulino, Memory impairment in old mice is differentially sensitive to different classes of antidepressants, *Eur. Neuropsychopharmacol.* 23 (2013) S282.
- [73] S.C. Leiser, A.L. Pehrson, P.J. Robichaud, C. Sanchez, Multimodal antidepressant vortioxetine increases frontal cortical oscillations unlike escitalopram and duloxetine – a quantitative EEG study in rats, *Br. J. Pharmacol.* 171 (2014) 4255–4272.
- [74] G. Smagin, D. Song, D.P. Budac, A. Pehrson, Y. Li, C. Sanchez, Chronic treatment with vortioxetine activates the central histaminergic system: a microdialysis study in rats, *Biol. Psychiatry* 75 (2014) 391S.
- [75] K.G. Du Jardin, N. Liebenberg, H. Müller, C. Sanchez, G. Wegener, B. Elfving, Single dose vortioxetine or ketamine but not fluoxetine increases expression of neuroplasticity related genes in the rat prefrontal cortex, *Eur. Neuropsychopharmacol.* 23 (2013) S392.
- [76] Y. Li, C. Sanchez, A. Abdourahman, J.A. Tamm, M. Gulino, Reversal by vortioxetine of age-induced memory impairment is associated with changes in specific gene expressions in female mice, *Eur. Neuropsychopharmacol.* 24 (2014) S370.
- [77] C. Betry, A. Etievant, A. Pehrson, C. Sanchez, N. Haddjeri, Effect of the multimodal acting antidepressant vortioxetine on rat hippocampal plasticity and recognition memory, *Prog. Neuro-psychopharmacol. Biol. Psychiatry* 58 (2014) 38–46.
- [78] J.P. Guilloux, I. Mendez-David, A. Pehrson, B.P. Guiard, C. Reperant, S. Orvoen, et al., Antidepressant and anxiolytic potential of the multimodal antidepressant vortioxetine (Lu AA21004) assessed by behavioural and neurogenesis outcomes in mice, *Neuropharmacology* 73 (2013) 147–159.
- [79] G. Sanacora, M. Banasr, From pathophysiology to novel antidepressant drugs: glial contributions to the pathology and treatment of mood disorders, *Biol. Psychiatry* 73 (2013) 1172–1179.
- [80] A.L. Pehrson, C. Sanchez, Vortioxetine reverses social recognition memory impairments induced by acetylcholine or glutamate dysregulation in rats, *Eur. Neuropsychopharmacol.* 24 (2014) S369.
- [81] E. Alvarez, V. Perez, M. Dragheim, H. Loft, F. Artigas, A double-blind, randomized, placebo-controlled, active reference study of Lu AA21004 in patients with major depressive disorder, *Int. J. Neuropsychopharmacol.: Off. Sci. J. Coll. Int. Neuropsychopharmacol.* 15 (2012) 589–600.
- [82] J.P. Boulenger, H. Loft, I. Florea, A randomized clinical study of Lu AA21004 in the prevention of relapse in patients with major depressive disorder, *J. Psychopharmacol. (Oxf.)* 26 (2012) 1408–1416.
- [83] M.Y. Alam, P.L. Jacobsen, Y. Chen, M. Serenko, A.R. Mahabeshwarkar, Safety, tolerability, and efficacy of vortioxetine (Lu AA21004) in major depressive disorder: results of an open-label, flexible-dose, 52-week extension study, *Int. Clin. Psychopharmacol.* 29 (2014) 36–44.
- [84] D.S. Baldwin, T. Hansen, I. Florea, Vortioxetine (Lu AA21004) in the long-term open-label treatment of major depressive disorder, *Curr. Med. Res. Opin.* 28 (2012) 1717–1724.
- [85] C. Katona, T. Hansen, C.K. Olsen, A randomized, double-blind, placebo-controlled, duloxetine-referenced, fixed-dose study comparing the efficacy and safety of Lu AA21004 in elderly patients with major depressive disorder, *Int. Clin. Psychopharmacol.* 27 (2012) 215–223.
- [86] R.S. McIntyre, S. Lophaven, C.K. Olsen, A randomized, double-blind, placebo-controlled study of vortioxetine on cognitive function in depressed adults, *Int. J. Neuropsychopharmacol.: Off. Sci. J. Coll. Int. Neuropsychopharmacol.* (2014) 1–11.
- [87] A. Mahabeshwarkar, J. Zajecka, W. Jacobson, Y. Chen, K. R., Efficacy of vortioxetine on cognitive function in adult patients with major depressive disorder: results of a randomized, double-blind, active-referenced, placebo-controlled trial, *Int. J. Neuropsychopharmacol.: Off. Sci. J. Coll. Int. Neuropsychopharmacol.* (2014) 17.
- [88] D.S. Baldwin, M. Serenko, W. Palo, S. Lophaven, J. Matz, The safety and tolerability of vortioxetine (Lu AA21004) in the treatment of adults with major depressive disorder (MDD): a pooled analysis, *Int. J. Psychiatry Clin. Pract.* 17 (2013) 16–17.
- [89] P.L. Jacobsen, L. Harper, M. Serenko, S. Chan, A.R. Mahabeshwarkar, A phase 3, long-term, open-label extension study evaluating the safety and tolerability of 15 and 20 mg vortioxetine in subjects with major depressive disorder, *Int. J. Neuropsychopharmacol.: Off. Sci. J. Collegium Int. Neuropsychopharmacol.* 17 (2014) 97–98.
- [90] J. Zhang, M.V. Mathis, J.W. Sellers, G. Kordzakhia, A.J. Jackson, A. Dow, et al., The US Food and Drug Administration's perspective on the new antidepressant vortioxetine, *J. Clin. Psychiatry* 76 (2015) 8–14.
- [91] J. Areberg, M. Luntang-Jensen, B. Sogaard, D.O. Nilausen, Occupancy of the serotonin transporter after administration of Lu AA21004 and its relation to plasma concentration in healthy subjects, *Basic Clin. Pharmacol. Toxicol.* 110 (2012) 401–404.
- [92] J.H. Meyer, A.A. Wilson, S. Sagrati, D. Hussey, A. Carella, W.Z. Potter, et al., Serotonin transporter occupancy of five selective serotonin reuptake inhibitors at different doses: an [¹¹C]DASB positron emission tomography study, *Am. J. Psychiatry* 161 (2004) 826–835.
- [93] P. Stenkrona, C. Halldin, J. Lundberg, 5-HTT and 5-HT(1A) receptor occupancy of the novel substance vortioxetine (Lu AA21004). A PET study in control subjects, *Eur. Neuropsychopharmacol.: J. Eur. Coll. Neuropsychopharmacol.* 23 (2013) 1190–1198.
- [94] B. Kobilka, Adrenergic receptors as models for G protein-coupled receptors, *Annu. Rev. Neurosci.* 15 (1992) 87–114.
- [95] M.B. Emerit, S. El Mestikawy, H. Gozlan, B. Rouot, M. Hamon, Physical evidence of the coupling of solubilized 5-HT_{1A} binding sites with G regulatory proteins, *Biochem. Pharmacol.* 39 (1990) 7–18.
- [96] M.B. Assie, C. Cosi, W. Koek, Correlation between low/high affinity ratios for 5-HT_{1A} receptors and intrinsic activity, *Eur. J. Pharmacol.* 386 (1999) 97–103.
- [97] M. Nord, S.J. Finnema, C. Halldin, L. Farde, Effect of a single dose of escitalopram on serotonin concentration in the non-human and human primate brain, *Int. J. Neuropsychopharmacol.: Off. Sci. J. Coll. Int. Neuropsychopharmacol.* 16 (2013) 1577–1586.
- [98] J.S. Kumar, R.V. Parsey, S.A. Kassir, V.J. Majo, M.S. Milak, J. Prabhakaran, et al., Autoradiographic evaluation of [³H]CUMI-101, a novel, selective 5-HT_{1A}R ligand in human and baboon brain, *Brain Res.* 1507 (2013) 11–18.
- [99] C. de Bodinat, B. Guardiola-Lemaitre, E. Mocaer, P. Renard, C. Munoz, M.J. Millan, Agomelatine, the first melatonergic antidepressant: discovery, characterization and development, *Nat. Rev. Drug Discov.* 9 (2010) 628–642.
- [100] J. Axelrod, The pineal gland: a neurochemical transducer, *Science* 184 (1974) 1341–1348.
- [101] Summary of Product Characteristics for Agomelatine (Valdoxan), 2009 Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Product_Information/human/000915/WC500046227.pdf.
- [102] V. Srinivasan, R. Zakaria, Z. Othman, E.C. Lauterbach, D. Acuna-Castroviejo, Agomelatine in depressive disorders: its novel mechanisms of action, *J. Neuro-psychiatry Clin. Neurosci.* 24 (2012) 290–308.
- [103] M. Bourin, E. Mocaer, R. Porsolt, Antidepressant-like activity of S 20098 (agomelatine) in the forced swimming test in rodents: involvement of melatonin and serotonin receptors, *J. Psychiatry Neurosci.* 29 (2004) 126–133.
- [104] M. Papp, P. Gruca, P.A. Boyer, E. Mocaer, Effect of agomelatine in the chronic mild stress model of depression in the rat, *Neuropsychopharmacology* 28 (2003) 694–703.
- [105] V. Bertaina-Anglade, C.D. la Rochelle, P.A. Boyer, E. Mocaer, Antidepressant-like effects of agomelatine (S 20098) in the learned helplessness model, *Behav. Pharmacol.* 17 (2006) 703–713.
- [106] M.J. Millan, A. Gobert, F. Lejeune, A. Dekeyne, A. Newman-Tancredi, V. Pasteau, et al., The novel melatonin agonist agomelatine (S20098) is an antagonist at 5-hydroxytryptamine_{2C} receptors, blockade of which enhances the activity of frontocortical dopaminergic and adrenergic pathways, *J. Pharmacol. Exp. Ther.* 306 (2003) 954–964.
- [107] F. Chenu, M. El Mansari, P. Blier, Electrophysiological effects of repeated administration of agomelatine on the dopamine, norepinephrine, and serotonin systems in the rat brain, *Neuropsychopharmacology* 38 (2013) 275–284.
- [108] C.A. McClung, How might circadian rhythms control mood? Let me count the ways, *Biol. Psychiatry* 74 (2013) 242–249.
- [109] S.H. Kennedy, S.J. Rizvi, Agomelatine in the treatment of major depressive disorder: potential for clinical effectiveness, *CNS Drugs* 24 (2010) 479–499.

- [110] M. Di Giannantonio, G. Di Iorio, R. Guglielmo, D. De Berardis, C.M. Conti, T. Acciavatti, et al., Major depressive disorder, anhedonia and agomelatine: an open-label study, *J. Biol. Regul. Homeost. Agents* 25 (2011) 109–114.
- [111] G. Martinotti, G. Sepede, F. Gambi, G. Di Iorio, D. De Berardis, M. Di Nicola, et al., Agomelatine versus venlafaxine XR in the treatment of anhedonia in major depressive disorder: a pilot study, *J. Clin. Psychopharmacol.* 32 (2012) 487–491.
- [112] K.L. Huang, W.C. Lu, Y.Y. Wang, G.C. Hu, C.H. Lu, W.Y. Lee, et al., Comparison of agomelatine and selective serotonin reuptake inhibitors/serotonin-norepinephrine reuptake inhibitors in major depressive disorder: a meta-analysis of head-to-head randomized clinical trials, *Aust. N. Z. J. Psychiatry* 48 (2014) 663–671.
- [113] S.A. Montgomery, S.H. Kennedy, G.D. Burrows, M. Lejoyeux, I. Hindmarch, Absence of discontinuation symptoms with agomelatine and occurrence of discontinuation symptoms with paroxetine: a randomized, double-blind, placebo-controlled discontinuation study, *Int. Clin. Psychopharmacol.* 19 (2004) 271–280.
- [114] A. Montejo, S. Majadas, S.J. Rizvi, S.H. Kennedy, The effects of agomelatine on sexual function in depressed patients and healthy volunteers, *Hum. Psychopharmacol.* 26 (2011) 537–542.
- [115] S.M. Stahl, M. Fava, M.H. Trivedi, A. Caputo, A. Shah, A. Post, Agomelatine in the treatment of major depressive disorder: an 8-week, multicenter, randomized, placebo-controlled trial, *J. Clin. Psychiatry* 71 (2010) 616–626.
- [116] B.H. Harvey, F.N. Slabbert, New insights on the antidepressant discontinuation syndrome, *Hum. Psychopharmacol.* 29 (2014) 503–516.
- [117] G. Serafini, M. Pompili, M. Innamorati, Y. Dwivedi, G. Brahmachari, P. Girardi, Pharmacological properties of glutamatergic drugs targeting NMDA receptors and their application in major depression, *Curr. Pharm. Des.* 19 (2013) 1898–1922.
- [118] C. Pittenger, G. Sanacora, J.H. Krystal, The NMDA receptor as a therapeutic target in major depressive disorder, *CNS Neurol. Disord. Drug Targets* 6 (2007) 101–115.
- [119] G.A. Mealing, T.H. Lanthorn, C.L. Murray, D.L. Small, P. Morley, Differences in degree of trapping of low-affinity uncompetitive N-methyl-D-aspartic acid receptor antagonists with similar kinetics of block, *J. Pharmacol. Exp. Ther.* 288 (1999) 204–210.
- [120] R.S. Duman, Pathophysiology of depression and innovative treatments: remodeling glutamatergic synaptic connections, *Dialogues Clin. Neurosci.* 16 (2014) 11–27.
- [121] B. Moghaddam, B. Adams, A. Verma, D. Daly, Activation of glutamatergic neurotransmission by ketamine: a novel step in the pathway from NMDA receptor blockade to dopaminergic and cognitive disruptions associated with the prefrontal cortex, *J. Neurosci.: Off. J. Soc. Neurosci.* 17 (1997) 2921–2927.
- [122] A.E. Autry, M. Adachi, E. Nosyreva, E.S. Na, M.F. Los, P.F. Cheng, et al., NMDA receptor blockade at rest triggers rapid behavioural antidepressant responses, *Nature* 475 (2011) 91–95.
- [123] N. Li, B. Lee, R.J. Liu, M. Banasr, J.M. Dwyer, M. Iwata, et al., mTOR-dependent synapse formation underlies the rapid antidepressant effects of NMDA antagonists, *Science* 329 (2010) 959–964.
- [124] N. Li, R.J. Liu, J.M. Dwyer, M. Banasr, B. Lee, H. Son, et al., Glutamate N-methyl-D-aspartate receptor antagonists rapidly reverse behavioral and synaptic deficits caused by chronic stress exposure, *Biol. Psychiatry* 69 (2011) 754–761.
- [125] A. de Bartolomeis, C. Sarappa, E.F. Buonaguro, F. Marmo, A. Eramo, C. Tomasetti, et al., Different effects of the NMDA receptor antagonists ketamine, MK-801, and memantine on postsynaptic density transcripts and their topography: role of Homer signaling, and implications for novel antipsychotic and pro-cognitive targets in psychosis, *Prog. Neuro-psychopharmacol. Biol. Psychiatry* 46 (2013) 1–12.
- [126] H. Okuno, K. Akashi, Y. Ishii, N. Yagishita-Kyo, K. Suzuki, M. Nonaka, et al., Inverse synaptic tagging of inactive synapses via dynamic interaction of Arc/Arg3.1 with CaMKIIbeta, *Cell* 149 (2012) 886–898.
- [127] L. Pozzi, I.P. Dorocic, X. Wang, M. Carlen, K. Meletis, Mice lacking NMDA receptors in parvalbumin neurons display normal depression-related behavior and response to antidepressant action of NMDAR antagonists, *PLOS ONE* 9 (2014) e83879.
- [128] V. Gigliucci, G. O'Dowd, S. Casey, D. Egan, S. Gibney, A. Harkin, Ketamine elicits sustained antidepressant-like activity via a serotonin-dependent mechanism, *Psychopharmacology (Berl.)* 228 (2013) 157–166.
- [129] M.J. Robson, M. Elliott, M.J. Seminerio, R.R. Matsumoto, Evaluation of sigma (sigma) receptors in the antidepressant-like effects of ketamine in vitro and in vivo, *Eur. Neuropsychopharmacol.: J. Eur. Coll. Neuropsychopharmacol.* 22 (2012) 308–317.
- [130] M. Nishimura, K. Sato, T. Okada, I. Yoshiya, P. Schloss, S. Shimada, et al., Ketamine inhibits monoamine transporters expressed in human embryonic kidney 293 cells, *Anesthesiology* 88 (1998) 768–774.
- [131] H. Yamanaka, C. Yokoyama, H. Mizuma, S. Kurai, S.J. Finnema, C. Hallidin, et al., A possible mechanism of the nucleus accumbens and ventral pallidum 5-HT1B receptors underlying the antidepressant action of ketamine: a PET study with macaques, *Transl. Psychiatry* 4 (2014) e342.
- [132] J.W. Murrough, D.V. Iosifescu, L.C. Chang, R.K. Al Jurdi, C.E. Green, A.M. Perez, et al., Antidepressant efficacy of ketamine in treatment-resistant major depression: a two-site randomized controlled trial, *Am. J. Psychiatry* 170 (2013) 1134–1142.
- [133] C.A. Zarate Jr., J.B. Singh, P.J. Carlson, N.E. Brutsche, R. Ameli, D.A. Luckenbaugh, et al., A randomized trial of an N-methyl-D-aspartate antagonist in treatment-resistant major depression, *Arch. Gen. Psychiatry* 63 (2006) 856–864.
- [134] L. Ibrahim, N. Diazgranados, D.A. Luckenbaugh, R. Machado-Vieira, J. Baumann, A.G. Mallinger, et al., Rapid decrease in depressive symptoms with an N-methyl-D-aspartate antagonist in ECT-resistant major depression, *Prog. Neuro-psychopharmacol. Biol. Psychiatry* 35 (2011) 1155–1159.
- [135] R.B. Price, M.K. Nock, D.S. Charney, S.J. Mathew, Effects of intravenous ketamine on explicit and implicit measures of suicidality in treatment-resistant depression, *Biol. Psychiatry* 66 (2009) 522–526.
- [136] P. Blier, D. Zigman, J. Blier, On the safety and benefits of repeated intravenous injections of ketamine for depression, *Biol. Psychiatry* 72 (2012) e11–e12.
- [137] G. Sanacora, M.A. Smith, S. Pathak, H.L. Su, P.H. Boeijinga, D.J. McCarthy, et al., Lanicemine: a low-trapping NMDA channel blocker produces sustained antidepressant efficacy with minimal psychotomimetic adverse effects, *Mol. Psychiatry* 19 (9) (2013) 978–985.
- [138] C.A. Zarate Jr., D. Mathews, L. Ibrahim, J.F. Chaves, C. Marquardt, I. Ukoh, et al., A randomized trial of a low-trapping nonselective N-methyl-D-aspartate channel blocker in major depression, *Biol. Psychiatry* 74 (2013) 257–264.
- [139] C.A. Zarate Jr., J.B. Singh, J.A. Quiroz, G. De Jesus, K.K. Denicoff, D.A. Luckenbaugh, et al., A double-blind, placebo-controlled study of memantine in the treatment of major depression, *Am. J. Psychiatry* 163 (2006) 153–155.
- [140] B. Pochwat, A. Palucha-Poniewiera, B. Szewczyk, A. Pilc, G. Nowak, NMDA antagonists under investigation for the treatment of major depressive disorder, *Expert Opin. Investig. Drugs* (2014) 1–12.
- [141] S.H. Preskorn, B. Baker, S. Kolluri, F.S. Menniti, M. Krams, J.W. Landen, An innovated design to establish proof of concept of the antidepressant effects of the NR2B subunit selective N-methyl-D-aspartate antagonist, CP-101,606, in patients with treatment-refractory major depressive disorder, *J. Clin. Psychopharmacol.* 28 (2008) 631–637.
- [142] L. Ibrahim, N. Diaz Granados, L. Jolkovsky, N. Brutsche, D.A. Luckenbaugh, W.J. Herring, et al., A randomized, placebo-controlled, crossover pilot trial of the oral selective NR2B antagonist MK-0657 in patients with treatment-resistant major depressive disorder, *J. Clin. Psychopharmacol.* 32 (2012) 551–557.
- [143] J.R. Moskal, R. Burch, J.S. Burgdorf, R.A. Kroes, P.K. Stanton, J.F. Disterhoft, et al., GLYX-13, an NMDA receptor glycine site functional antagonist enhances cognition and produces antidepressant effects without the psychotomimetic side effects of NMDA receptor antagonists, *Expert Opin. Investig. Drugs* 23 (2014) 243–254.
- [144] A. Pilc, J.M. Wieronska, P. Skolnick, Glutamate-based antidepressants: preclinical psychopharmacology, *Biol. Psychiatry* 73 (2013) 1125–1132.
- [145] J. Quiroz, P. Tamburri, D. Deputa, L. Banken, U. Beyer, P. Fontoura, et al., The efficacy and safety of basimglurant as adjunctive therapy in major depression: a randomized, double-blind, placebo controlled study, *Neuropsychopharmacology* 39 (2014) S376–S377.
- [146] F. Holsboer, M. Ising, Central CRH system in depression and anxiety – evidence from clinical studies with CRH1 receptor antagonists, *Eur. J. Pharmacol.* 583 (2008) 350–357.
- [147] R.B. Lloyd, C.B. Nemeroff, The role of corticotropin-releasing hormone in the pathophysiology of depression: therapeutic implications, *Curr. Top. Med. Chem.* 11 (2011) 609–617.
- [148] H.M. Sickmann, Y. Li, A. Mork, C. Sanchez, M. Gulino, Does stress elicit depression? Evidence from clinical and preclinical studies, *Curr. Top. Behav. Neurosci.* 18 (2014) 123–159.
- [149] C.M. Blasey, T.S. Block, J.K. Belanoff, R.L. Roe, Efficacy and safety of mifepristone for the treatment of psychotic depression, *J. Clin. Psychopharmacol.* 31 (2011) 436–440.
- [150] Corcept Therapeutics Announces First Quarter 2014 Financial Results. http://www.corcept.com/news_events/view/pr_1399472862014.
- [151] A.W. Zobel, T. Nickel, H.E. Kunzel, N. Ackl, A. Sonntag, M. Ising, et al., Effects of the high-affinity corticotropin-releasing hormone receptor 1 antagonist R121919 in major depression: the first 20 patients treated, *J. Psychiatr. Res.* 34 (2000) 171–181.
- [152] B. Binneman, D. Feltner, S. Kolluri, Y. Shi, R. Qiu, T. Stiger, A 6-week randomized, placebo-controlled trial of CP-316,311 (a selective CRH1 antagonist) in the treatment of major depression, *Am. J. Psychiatry* 165 (2008) 617–620.
- [153] M. Ising, U.S. Zimmermann, H.E. Kunzel, M. Uhr, A.C. Foster, S.M. Learned-Coughlin, et al., High-affinity CRF1 receptor antagonist NBI-34041: preclinical and clinical data suggest safety and efficacy in attenuating elevated stress response, *Neuropsychopharmacology* 32 (2007) 1941–1949.
- [154] M.S. Kramer, N. Cutler, J. Feighner, R. Shrivastava, J. Carman, J.J. Sramek, et al., Distinct mechanism for antidepressant activity by blockade of central substance P receptors, *Science* 281 (1998) 1640–1645.
- [155] M.S. Kramer, A. Winokur, J. Kelsey, S.H. Preskorn, A.J. Rothschild, D. Snavely, et al., Demonstration of the efficacy and safety of a novel substance P (NK1) receptor antagonist in major depression, *Neuropsychopharmacology* 29 (2004) 385–392.
- [156] M. Keller, S. Montgomery, W. Ball, M. Morrison, D. Snavely, G. Liu, et al., Lack of efficacy of the substance p (neurokinin1 receptor) antagonist aprepitant in the treatment of major depressive disorder, *Biol. Psychiatry* 59 (2006) 216–223.
- [157] R. Machado-Vieira, C.A. Zarate Jr., Proof of concept trials in bipolar disorder and major depressive disorder: a translational perspective in the search for improved treatments, *Depress. Anxiety* 28 (2011) 267–281.
- [158] E. Ratti, K. Bellew, P. Bettica, H. Bryson, S. Zamuner, G. Archer, et al., Results from 2 randomized, double-blind, placebo-controlled studies of the novel NK1 receptor antagonist casopitant in patients with major depressive disorder, *J. Clin. Psychopharmacol.* 31 (2011) 727–733.
- [159] Sanofi pulls plug on four Ph III drugs, Press release 29 April 2009. <http://www.in-pharmatechnologist.com/Regulatory-Safety/Sanofi-pulls-plug-on-four-Ph-III-drugs2009>.

- [160] W.A. Ball, D.B. Snively, R.J. Hargreaves, A. Szegedi, C. Lines, S.A. Reines, Addition of an NK1 receptor antagonist to an SSRI did not enhance the antidepressant effects of SSRI monotherapy: results from a randomized clinical trial in patients with major depressive disorder, *Hum. Psychopharmacol.* 29 (2014) 568–577.
- [161] M. Nolle, S. Leman, Role of orexin in the pathophysiology of depression: potential for pharmacological intervention, *CNS Drugs* 27 (2013) 411–422.
- [162] M. Catena-Dell'Osso, A. Fagioli, D. Marazziti, S. Baroni, C. Bellantuono, Non-monoaminergic targets for the development of antidepressants: focus on neuropeptides, *Mini Rev. Med. Chem.* 13 (2013) 2–10.
- [163] D.S. Janowsky, M.K. el-Yousef, J.M. Davis, H.J. Sekerke, A cholinergic-adrenergic hypothesis of mania and depression, *Lancet* 2 (1972) 632–635.
- [164] W.C. Drevets, C.A. Zarate Jr., M.L. Furey, Antidepressant effects of the muscarinic cholinergic receptor antagonist scopolamine: a review, *Biol. Psychiatry* 73 (2013) 1156–1163.
- [165] R.J. Jaffe, V. Novakovic, E.D. Peselow, Scopolamine as an antidepressant: a systematic review, *Clin. Neuropharmacol.* 36 (2013) 24–26.
- [166] D. Khajavi, M. Farokhnia, A. Modabbernia, M. Ashrafi, S.H. Abbasi, M. Tabrizi, et al., Oral scopolamine augmentation in moderate to severe major depressive disorder: a randomized, double-blind, placebo-controlled study, *J. Clin. Psychiatry* 73 (2012) 1428–1433.
- [167] Y.S. Mineur, M.R. Picciotto, Nicotine receptors and depression: revisiting and revising the cholinergic hypothesis, *Trends Pharmacol. Sci.* 31 (2010) 580–586.
- [168] E. Vieta, M.E. Thase, D. Naber, B. D'Souza, E. Rancans, U. Lepola, et al., Efficacy and tolerability of flexibly-dosed adjunct TC-5214 (dexmecamylamine) in patients with major depressive disorder and inadequate response to prior antidepressant, *Eur. Neuropsychopharmacol.: J. Eur. Coll. Neuropsychopharmacol.* 24 (2014) 564–574.
- [169] A.T. Knoll, W.A. Carlezon Jr., Dynorphin, stress, and depression, *Brain Res.* 1314 (2010) 56–73.
- [170] Alkermes Presents Positive Results from Phase 2 Clinical Study of ALKS 5461 in Major Depressive Disorder at 53rd Annual NCDEU Meeting. <http://phx.corporate-ir.net/phoenix.zhtml?c=92211&p=irol-corporateNewsArticle&ID=18258172013>.
- [171] E. Ehrlich, R. Turncliff, Y. Du, R. Leigh-Pemberton, E. Fernandez, R. Jones, et al., Evaluation of opioid modulation in major depressive disorder, *Neuropsychopharmacology* (2014) (Epub ahead of print).
- [172] Y. Dowlati, N. Herrmann, W. Swardfager, H. Liu, L. Sham, E.K. Reim, et al., A meta-analysis of cytokines in major depression, *Biol. Psychiatry* 67 (2010) 446–457.
- [173] C. Bay-Richter, K.R. Linderholm, C.K. Lim, M. Samuelsson, L. Traskanman-Bendz, G.J. Guillemin, et al., A role for inflammatory metabolites as modulators of the glutamate N-methyl-D-aspartate receptor in depression and suicidality, *Brain Behav. Immun.* 43 (2015) 110–117.
- [174] N. Muller, M.J. Schwarz, S. Dehning, A. Douhe, A. Cerovecki, B. Goldstein-Muller, et al., The cyclooxygenase-2 inhibitor celecoxib has therapeutic effects in major depression: results of a double-blind, randomized, placebo controlled, add-on pilot study to reboxetine, *Mol. Psychiatry* 11 (2006) 680–684.
- [175] S. Akhondzadeh, S. Jafari, F. Raisi, A.A. Nasehi, A. Ghoreishi, B. Salehi, et al., Clinical trial of adjunctive celecoxib treatment in patients with major depression: a double blind and placebo controlled trial, *Depress. Anxiety* 26 (2009) 607–611.
- [176] S.H. Abbasi, F. Hosseini, A. Modabbernia, M. Ashrafi, S. Akhondzadeh, Effect of celecoxib add-on treatment on symptoms and serum IL-6 concentrations in patients with major depressive disorder: randomized double-blind placebo-controlled study, *J. Affect. Disord.* 141 (2012) 308–314.
- [177] D. Johansson, A. Falk, M.M. Marcus, T.H. Svensson, Celecoxib enhances the effect of reboxetine and fluoxetine on cortical noradrenaline and serotonin output in the rat, *Prog. Neuro-psychopharmacol. Biol. Psychiatry* 39 (2012) 143–148.
- [178] C.L. Raison, R.E. Rutherford, B.J. Woolwine, C. Shuo, P. Schettler, D.F. Drake, et al., A randomized controlled trial of the tumor necrosis factor antagonist infliximab for treatment-resistant depression: the role of baseline inflammatory biomarkers, *JAMA Psychiatry* 70 (2013) 31–41.
- [179] A. Newman-Tancredi, J.C. Martel, M.B. Assie, J. Buritova, E. Laussersergues, C. Cosi, et al., Signal transduction and functional selectivity of F15599, a preferential post-synaptic 5-HT_{1A} receptor agonist, *Br. J. Pharmacol.* 156 (2009) 338–353.
- [180] F.G. Revel, J.L. Moreau, B. Pouzet, R. Mory, A. Bradaia, D. Buchy, et al., A new perspective for schizophrenia: TAAR1 agonists reveal antipsychotic- and antidepressant-like activity, improve cognition and control body weight, *Mol. Psychiatry* 18 (2013) 543–556.
- [181] M.J. O'Neill, D. Bleakman, D.M. Zimmerman, E.S. Nisenbaum, AMPA receptor potentiators for the treatment of CNS disorders, *Curr. Drug Targets CNS Neurol Disord.* 3 (2004) 181–194.
- [182] F.S. Menniti, C.W. Lindsley, P.J. Conn, J. Pandit, P. Zagouras, R.A. Volkman, Allosteric modulators for the treatment of schizophrenia: targeting glutamatergic networks, *Curr. Top. Med. Chem.* 13 (2013) 26–54.
- [183] FDA's Psychiatric Division has Rejected Cortex's Request to Study CX717 in Phase IIb ADHD Study. http://www.cortexpharm.com/news/07/txt_101107.html#2007.
- [184] R.J. McLaughlin, G. Gobbi, Cannabinoids and emotionality: a neuroanatomical perspective, *Neuroscience* 204 (2012) 134–144.
- [185] K.H. Thomas, R.M. Martin, J. Potokar, M. Pirmohamed, D. Gunnell, Reporting of drug induced depression and fatal and non-fatal suicidal behaviour in the UK from 1998 to 2011, *BMC Pharmacol. Toxicol.* 15 (2014) 54.
- [186] T. Sharp, Molecular and cellular mechanisms of antidepressant action, *Curr. Top. Behav. Neurosci.* 14 (2013) 309–325.
- [187] G. Serafini, Neuroplasticity and major depression, the role of modern antidepressant drugs, *World J. Psychiatry* 2 (2012) 49–57.
- [188] E. Castren, T. Rantamaki, The role of BDNF and its receptors in depression and antidepressant drug action: reactivation of developmental plasticity, *Dev. Neurobiol.* 70 (2010) 289–297.
- [189] T. Rantamaki, E. Castren, Targeting TrkB neurotrophin receptor to treat depression, *Expert Opin. Ther. Targets* 12 (2008) 705–715.
- [190] M. Cazorla, J. Premont, A. Mann, N. Girard, C. Kellendonk, D. Rognan, Identification of a low-molecular weight TrkB antagonist with anxiolytic and antidepressant activity in mice, *J. Clin. Investig.* 121 (2011) 1846–1857.
- [191] A.J. Eisch, C.A. Colanos, J. de Wit, R.D. Simonak, C.M. Pudiak, M. Barrot, et al., Brain-derived neurotrophic factor in the ventral midbrain-nucleus accumbens pathway: a role in depression, *Biol. Psychiatry* 54 (2003) 994–1005.
- [192] J.J. Day, J.D. Sweatt, Epigenetic treatments for cognitive impairments, *Neuropsychopharmacology* 37 (2012) 247–260.
- [193] H.E. Covington 3rd, V. Vialou, E.J. Nestler, From synapse to nucleus: novel targets for treating depression, *Neuropharmacology* 58 (2010) 683–693.
- [194] H.E. Covington 3rd, I. Maze, Q.C. LaPlant, V.F. Vialou, Y.N. Ohnishi, O. Berton, et al., Antidepressant actions of histone deacetylase inhibitors, *J. Neurosci.: Off. J. Soc. Neurosci.* 29 (2009) 11451–11460.
- [195] P. Morgan, P.H. Van Der Graaf, J. Arrowsmith, D.E. Feltner, K.S. Drummond, C.D. Wegner, et al., Can the flow of medicines be improved? Fundamental pharmacokinetic and pharmacological principles toward improving Phase II survival, *Drug Discov. Today* 17 (2012) 419–424.
- [196] F. Sobrio, G. Gilbert, C. Perrio, L. Barre, D. Debruyne, PET and SPECT imaging of the NMDA receptor system: an overview of radiotracer development, *Mini Rev. Med. Chem.* 10 (2010) 870–886.
- [197] K. Hashimoto, Targeting of NMDA receptors in new treatments for schizophrenia, *Expert Opin. Ther. Targets* 18 (2014) 1049–1063.
- [198] F.R. Ferreira, C. Biojone, S.R. Joca, F.S. Guimaraes, Antidepressant-like effects of N-acetyl-L-cysteine in rats, *Behav. Pharmacol.* 19 (2008) 747–750.
- [199] M. Berk, D.L. Copolov, O. Dean, K. Lu, S. Jeavons, I. Schapkaitz, et al., N-acetyl cysteine for depressive symptoms in bipolar disorder – a double-blind randomized placebo-controlled trial, *Biol. Psychiatry* 64 (2008) 468–475.
- [200] M. Berk, O.M. Dean, S.M. Cotton, S. Jeavons, M. Tanious, K. Kohlmann, et al., The efficacy of adjunctive N-acetylcysteine in major depressive disorder: a double-blind, randomized, placebo-controlled trial, *J. Clin. Psychiatry* 75 (2014) 628–636.
- [201] R.E. Horton, D.M. Apple, W.A. Owens, N.L. Baganz, S. Cano, N.C. Mitchell, et al., Decynium-22 enhances SSRI-induced antidepressant-like effects in mice: uncovering novel targets to treat depression, *J. Neurosci.: Off. J. Soc. Neurosci.* 33 (2013) 10534–10543.
- [202] S.I. Shyn, S.P. Hamilton, The genetics of major depression: moving beyond the monoamine hypothesis, *Psychiatr. Clin. N. Am.* 33 (2010) 125–140.
- [203] J.A. van Hooff, A.D. Spier, J.L. Yakel, S.C. Lummis, H.P. Vijverberg, Promiscuous coassembly of serotonin 5-HT₃ and nicotinic alpha4 receptor subunits into Ca²⁺-permeable ion channels, *Proc. Natl. Acad. Sci. U. S. A.* 95 (1998) 11456–11461.
- [204] J. Gonzalez-Maeso, R.L. Ang, T. Yuen, P. Chan, N.V. Weisstaub, J.F. Lopez-Gimenez, et al., Identification of a serotonin/glutamate receptor complex implicated in psychosis, *Nature* 452 (2008) 93–97.
- [205] M. Crowe, B. Beaglehole, H. Wells, R. Porter, Non-pharmacological strategies for treatment of inpatient depression, *Aust. N. Z. J. Psychiatry* 49 (2015) 215–226.
- [206] A. Rau, N. Grossheinrich, U. Palm, O. Pogarell, F. Padberg, Transcranial and deep brain stimulation approaches as treatment for depression, *Clin. EEG Neurosci.* 38 (2007) 105–115.
- [207] J.J. Schildkraut, S.S. Kety, Biogenic amines and emotion, *Science* 156 (1967) 21–37.
- [208] E.J. Nestler, M. Barrot, R.J. DiLeone, A.J. Eisch, S.J. Gold, L.M. Monteggia, Neurobiology of depression, *Neuron* 34 (2002) 13–25.
- [209] T.J. Riedler, Inflammatory mechanisms in major depressive disorder, *Curr. Opin. Psychiatry* 24 (2011) 519–525.
- [210] A.L. Pehrson, C. Sanchez, Altered gamma-aminobutyric acid neurotransmission in major depressive disorder: a critical review of the supporting evidence and the influence of serotonergic antidepressants, *Drug Des. Dev. Ther.* 9 (2015) 603–624.