Introduction
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Depression is an illness that affects both the mind and the body and is a leading cause of disability, workplace absenteeism, decreased productivity and high suicide rates.\textsuperscript{1,2} Depression is the most common psychiatric disorder in general practice and about one in ten patients seen in the primary care settings suffer from some form of depression.\textsuperscript{3,4} It is an important global public health problem due to both its relatively high lifetime prevalence and the significant disability that it causes. Depression accounted for 4.5\% of the worldwide total burden of disease (in terms of disability-adjusted life years). It is also responsible for the greatest proportion of burden attributable to non-fatal health outcomes, accounting for almost 12\% of total years lived with disability worldwide. Without treatment, depression has the tendency to assume a chronic course, to recur, and to be associated with increasing disability over time.

In a study by the World Health Organization (WHO) conducted at 14 sites, the most common diagnosis in primary care was depression.\textsuperscript{5} Depression is estimated to affect 340 million people globally.\textsuperscript{6} The prevalence of psychiatric disorders is reported to differ between countries and within countries, across various ethnicities.\textsuperscript{7} Most studies on depression are from the developed world and there are few studies from developing countries. The World Mental Health Survey Initiative carried out cross-national research in mental health, especially in developing countries. There have been a few population based studies from India but most have been done on selected groups.\textsuperscript{8-12} The prevalence of depression in a population based study conducted in urban Pakistan was 45.9\%,\textsuperscript{12} while in rural Bangladesh, it was reported to be 29\%,\textsuperscript{13} and in a peri-urban clinic based study in Uganda, it was reported to be 6.1\%.\textsuperscript{14} Earlier Indian studies have reported prevalence rates of depression that vary from 21–83\% in primary care practices.\textsuperscript{15-18}

In news of The Financial Express, 11 March 2012 it was mentioned that a study based on the World Health Organization’s World Mental Health Survey Initiative has said that India has the highest rate of major depression in the world. The study, ‘Cross-national epidemiology of DSM-IV major depressive episode, based on
interviews of nearly 90,000 subjects across 18 countries with different income levels was published in the peer-reviewed journal BMC Medicine by Biomed Central. The average lifetime rates of depression, according to the study, were found to be 14.6 per cent in ten high income countries, and 11.1 percent in eight low- to middle-income countries. But lifetime incidents of what was identified as Major Depressive Episodes (MDE) were highest among Indians at 35.9 percent, while China was at the lowest at 12 per cent. Average percentage of MDE was, however, considerably higher in high-income countries at 28.1 percent, compared to 19.8 percent in the low- to middle-income countries.

1.1 Types of depressive disorder

There are several forms of depression, each one with its own constellation of symptoms. Depression can be classified into two opposite poles: melancholic or somatic syndrome and atypical syndrome. The atypical syndrome can be characterized by reverse symptoms such as increased appetite, weight gain, hypersomnia, extreme fatigue\textsuperscript{19,20} and interpersonal rejection sensitivity.\textsuperscript{21,22} Melancholic depression includes major depressive disorder, dysthymia and subsyndromal depression.

![Depression and important factors](image_url)

Fig 1. Depression and important factors
Major depressive disorder is characterized by a heterogeneous group of behavioural, psychological and physiological symptoms, which include: psychomotor agitation or retardation, marked weight loss, disturbances of sleep, decreased appetite, fatigue, extreme feelings of guilt or worthlessness, difficulty in concentrating and suicidal ideation. Dysthymia is also associated with significant functional impairment, and it occurs in approximately 3% of people. Chronically depressed mood must be present during a period of two years or more, and during those periods, at least two of the following symptoms are present: appetite and sleep disturbances, decreased energy or fatigue, feelings of low self-esteem or hopelessness, and decreased concentration.

Subsyndromal depression is an acute mood disorder, although less severe than major depression. It has an increased risk for the development of major depression and decreased functioning. This form of depression is diagnosed when depressed mood and/or loss of interest or pleasure in nearly all activities, and one to three of the symptoms used to diagnose MDD last at least 2 weeks.

There are other forms of unipolar depressive disorder, which exhibit slightly different characteristics than those described above, and may only develop under certain medical conditions. Some examples of these situations are psychotic depression, postpartum depression and seasonal affective disorder (SAD). Besides the various forms of unipolar depression listed above, bipolar depression also exists, however not being as epidemiologically spread as major depression or dysthymia. Finally, another form of depression is iatrogenic depression, which is pathogenetically induced by the treatment of various disorders, including pharmacotherapy and surgical treatment, being more common in the elderly.
Despite the strong impact and prevalence of depression, there is still little knowledge about its pathogenesis. This might be due to several aspects, such as the difficulty in documenting the pathological changes in the brain rather than other organs. The available techniques for assessing the brain functions consist on post-mortem studies and neuroimaging, which provided important insights about brain regions involved in depression, although simple changes in brain activity cannot be considered sufficient to explain this complex syndrome in full. The regulation of emotions, reward and executive function implicates several brain regions and circuits, which are highly interconnected. Among these structures, the prefrontal cortex, ventral striatum (including nucleus accumbens), amygdala and the hippocampus play an important role (Figure 1). It is believed that impairment of these areas is related to depression; for this reason, these brain structures are considered to be targets of antidepressant treatment. Thus, brain regions listed above are thought to contribute to different mechanisms of depression. For example, neocortex and

**Fig 2. Types of depression**

1.2 Pathogenesis of depression

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hippocampus are believed to mediate cognitive symptoms of depression, such as memory impairments and feelings of worthlessness, hopelessness and guilt. Several studies of depressed patients demonstrated changes in blood flow, reductions in grey-matter volume and glia density in the prefrontal cortex and the hippocampus.\textsuperscript{28,31} The striatum and the amygdala are most likely responsible for emotional memory. Deficits in these structures may underlie anhedonia, anxiety and reduced motivation. Also, the hypothalamus may play a role in depression with neurovegetative symptoms such as sleep and appetite disturbances. Other subcortical structures (nucleus accumbens, amygdala) implicated in fear, reward and motivation, are also involved in this pathways.\textsuperscript{29}

Using functional magnetic resonance imaging (fMRI) and positron-emission tomography (PET), it has been shown that depressive symptoms associated with increased activity of the amygdala and subgenual cingulated cortex are related to dysphoric emotions.\textsuperscript{28}

The role of the serotonergic system and the hypothalamic-pituitary-adrenal (HPA) axis in mechanisms of depression is well documented, among other neuroregulatory systems of the brain. During depression, the HPA axis is characterized by its overactivity. Depressed patients were found to have elevated levels of corticotrophin-releasing hormone (CRH) and cortisol in 40\% of cases,\textsuperscript{32} an abnormality reversed by antidepressants and physical exercise.\textsuperscript{33} Low responses with the pituitary adrenocorticotropin (ACTH) secretion are another feature of depressed patients, which reflects altered functions of the HPA axis.\textsuperscript{34} The elevated CRH levels are believed to gradually desensitize the CRH receptors that attenuate pituitary response.

According to another hypothesis, the attenuated pituitary ACTH response may be due to the reduced sensitivity of serotonin receptors and reduced serotonergic neurotransmission. The release of ACTH is regulated by the serotonin-1A receptors, both at the pituitary and hypothalamus levels.\textsuperscript{35} Several studies indicate that serotonergic neurotransmission is impaired in depression.\textsuperscript{36,37} Pharmacological challenges and PET studies showed a decrease in serotonin-1A receptor-mediated
signaling in depressed patients.\textsuperscript{38} The monoamine hypothesis,\textsuperscript{39,40} states that depression is caused by impairment in monoamine function in the brain, characterized by a deficiency in neurotransmission mediated by serotonin (5-HT), norepinephrine and dopamine.

This hypothesis has been refined through the past years and more experimental and clinical evidence has come to light.\textsuperscript{28,38} The concentration of monoamines may be altered due to disruption in synthesis, storage or release, or it may remain normal, but the receptors and/or sub-cellular messenger’s activity may be altered. Monoamines are known to affect several aspects which are altered during depression, including sleep, vigilance, appetite, motivation, motor activity and reward. Another hypothesis is based on one of the core symptoms of depression, anhedonia. The brain reward system (BRS) is a neural pathway involved in eliciting rewarding experiences in animals, including humans, and it is thought that an altered function of this system may underlie brain mechanisms behind depression.\textsuperscript{41}

Finally, the role of neurotrophic factors on the etiology of depression has also been discussed.\textsuperscript{42} Neurotrophic factors are known for being potent regulators of the plasticity and survival of adult neurons and glia. Hence, the neurotrophic hypothesis states that a deficiency in neurotrophic support may contribute to hippocampal pathology during the development of depressive syndrome, being that this condition is reversed by antidepressant treatment and electroconvulsive shock (ECS).\textsuperscript{43,44} This hypothesis has focused on brain-derived neurotrophic factor (BDNF), one of the most prevalent neurotrophic factors in the brain,\textsuperscript{29} which concentration is decreased in major depression.\textsuperscript{45}

1.3 Theories of depression

1.3.1 Monamine/5-HT Hypothesis

Just as with schizophrenia, the most popular neurophysiological theory of depression follows from the drugs that are used to treat it. The evolution of antidepressant drugs has, in some ways, been the systematic narrowing down of monoamines to Serotonin. MAO inhibitors are Dopamine-Epinephrine-
Norepinephrine-Serotonin agonists. Tricyclic Antidepressants are Norepinephrine-Serotonin agonists, and, finally SSRIs act as Serotonin (5-HT) agonists.

Thus, the monoamine hypothesis has evolved in the same way, so that today one popular theory of depression, the Monoamine Hypothesis, is that depression is the result of underactivity of monoamines, especially 5-HT. Besides the fact that antidepression drugs are all monoamine agonists, there is other evidence that supports the theory. First, Reserpine, a monoamine antagonist, which was used to treat things like high blood pressure, is rarely used at the present time due to the fact that depression is a common side effect. Thus, not only can monoamine agonists decrease depression, but monoamine antagonists (Reserpine) can induce depression.

Another piece of evidence in support of the Monoamine Hypothesis is that levels of 5-HT, as measured by its metabolites, seem to be correlated with depression. For example, patients who have low levels of a 5-HT metabolite were found to be more likely to have committed suicide. It often takes two to three weeks for antidepressant drugs to effectively treat depression. This is a difficult phenomenon to explain within the context of the monoamine hypothesis. Presumably, in response to monoamine agonists these neurotransmitter levels increase right away, and, if depression is caused by low levels of the neurotransmitter, then depression should decrease as the levels of monoamines increase.

Two contrasting theories have been offered to explain this puzzling phenomenon, both of which rely on the supposition that the nervous system will compensate for large fluctuations in neurotransmitter levels with a compensatory increase or decrease in neurotransmitter or receptor release or sensitivity.

The first theory is the straightforward contention that the important effect of the antidepressant drugs is exactly the opposite of what the monoamine theory suggests. That is, over the course of three weeks the 5-HT post-synaptic receptors become subsensitive (as contrasted with the supersensitivity discussed in a schizophrenia module), so that the neurons actually become less responsive to 5-HT as a result of the antidepressants. Depression then, according to this interpretation is the result of
the overactivity of 5-HT neurons, and after the 5-HT agonists are taken for some time the 5-HT neurons respond less and less, and the depression goes away as a result. A second theory for this time-lag effect has now become more widely accepted, which is consistent with the monoamine hypothesis that depression is due to underactivity of 5-HT neurons. However, this theory still relies on the concepts of neural compensation and subsensitivity. The crucial difference is that according to what we’ll call the autoreceptor subsensitivity theory, it is not the post synaptic receptors that become subsensitive; rather it is 5-HT autoreceptors. Autoreceptors are typically located on the presynaptic or axonal membrane. These receptors, which serve a feedback function, are sensitive to the amount of neurotransmitter present in the intercellular fluid, and have an inhibitory effect on neurotransmitter production and/or release. Presumably, the initial effect of the monoamine agonists is to stimulate these autoreceptors such that they inhibit the increased release of monoamines resulting from the drug. After two or three weeks, however, the autoreceptors become subsensitive due to continued stimulation, and quit sending their inhibitory signal, so the monoamine agonists then have the effect that we would expect.

Another theory of depression also follows from successful treatment. The fact that sleep deprivation can effectively treat depression has led some to the conclusion that abnormal sleep patterns may play a role in depression. Also, most antidepressant drugs decrease or eliminate REM sleep, and those who suffer depression have been found to have abnormal sleep cycles. More specifically they have more REM, a shorter onset until REM, and generally more disrupted cycles.

Finally, those who have been successfully treated with sleep deprivation will often revert back to their depressed state as a result of a short nap. This has even leaded some to suggest that some type of depressogenic substance is released during sleep, and that the substance is deactivated during wakefulness. In fact, a typical pattern for some diagnosed with a major depression disorder is for the perception of depression to peak in the morning and decrease as the day goes one, which would be consistent with the idea that such a substance was being depleted as the day progresses.
1.3.2 The Serotonin Hypothesis

In 1965, Joseph Schildkraut put forth the hypothesis that depression was associated with low levels of norepinephrine, and later researchers theorized that serotonin was the neurotransmitter of interest. In subsequent years, there were numerous attempts to identify reproducible neurochemical alterations in the nervous systems of patients diagnosed with depression. For instance, researchers compared levels of serotonin metabolites in the cerebrospinal fluid of clinically depressed suicidal patients to controls, but the primary literature is mixed and plagued with methodological difficulties such as very small sample sizes and uncontrolled confounding variables. In a recent review of these studies, the chairman of the German Medical Board and colleagues stated, Reported associations of subgroups of suicidal behavior (e.g. violent suicide attempts) with low CSF–5HIAA (serotonin) concentrations are likely to represent somewhat premature translations of findings from studies that have flaws in methodology.

Attempts were also made to induce depression by depleting serotonin levels, but these experiments reaped no consistent result. Likewise, researchers found that huge increases in brain serotonin, arrived at by administering high-dose L-tryptophan, were ineffective at relieving depression. Contemporary neuroscience research has failed to confirm many serotonergic lesion in any mental disorder, and

![Diagram of serotonin synthesis and action](image-url)
has in fact provided significant counterevidence to the explanation of a simple neurotransmitter deficiency. Modern neuroscience has instead shown that the brain is vastly complex and poorly understood. While neuroscience is a rapidly advancing field, to propose that researchers can objectively identify a “chemical imbalance” at the molecular level is not compatible with the extant science. In fact, there is no scientifically established ideal “chemical balance” of serotonin, let alone an identifiable pathological imbalance. To equate the impressive recent achievements of neuroscience with support for the serotonin hypothesis is a mistake.

1.4 Neurotransmitter and depression

Insufficient activity of the neurotransmitters serotonin and norepinephrine is a central element of the model of depression most widely held by neurobiologists today. Nearly all of the drugs used to treat depression appear to enhance neurotransmission in one or both of these systems. In fact, understanding of the mechanism of action of antidepressant drugs in part gave rise to the model of depression. The synthesis of most neurotransmitters is controlled within the brain. For some neurotransmitters, the amount of biochemical precursors present in the brain can influence their rate of synthesis. During the 1970s, researchers established a body of evidence indicating ingestion of dietary precursors of certain neurotransmitters could increase their levels in the brain. This research suggested precursors of norepinephrine and serotonin might be useful in treating depression. In the late 1970s and 1980s, numerous researchers studied the use of the serotonin precursors L-tryptophan and 5-hydroxytryptophan (5-HTP), and the norepinephrine precursors tyrosine and phenylalanine in depressed patients. Interest in neurotransmitter precursors was partly a function of the shortcomings of antidepressant medications (particularly in terms of side-effects) available prior to Prozac and other selective serotonin re-uptake inhibitor drugs (SSRIs). It was hoped dietary precursors of key neurotransmitters might provide a more easily tolerated way of treating depression.

The conceptual case for the effectiveness of neurotransmitter precursors builds upon the idea that depression is a result of an inadequate amount or insufficient activity of
one or more neurotransmitters. Within limits, availability of the necessary precursors determines the amount of neurotransmitter synthesized. For example, serotonin production in the human brain can be increased two-fold by oral intake of L-tryptophan.\textsuperscript{51} Although administration of precursors appears to result in increased neurotransmitter synthesis, it is less clear whether it leads to augmentation of neurotransmitter release. An animal study found acute administration of L-tryptophan decreased the firing rate of serotonin neurons,\textsuperscript{52} which might tend to counteract increased synthesis. However, results from other animal studies suggest L-tryptophan administration can enhance serotonin release under some circumstances, and the same may be true in humans.\textsuperscript{53} Administration of 5-HTP has been associated with a significant increase in cerebrospinal fluid levels of 5-hydroxyindolacetic acid, the primary metabolite of serotonin, suggesting 5-HTP leads to an increased release of serotonin.

The key question which so far has not been answered is whether increased neurotransmitter release leads to ongoing stimulation of neurotransmitter activity; i.e., the strength of signaling. The latter depends not only on the amount of neurotransmitter released by the presynaptic neuron, but also on how long neurotransmitters remain in the synaptic cleft, and on factors which influence firing of the postsynaptic neuron. Neurotransmitter systems are characterized by feedback mechanisms that help maintain equilibrium with respect to neurotransmitter activity.

Thus, changed conditions in the short run, such as increased synthesis of a neurotransmitter, can be balanced by adaptations within the system. The time lag before appearance of a beneficial symptom-relieving effect of antidepressant drugs suggests late-developing factors may be more relevant to these drugs’ clinical efficacy than their acute effect (inhibiting re-uptake of neurotransmitters). Imipramine (a tricyclic antidepressant) can block neurotransmitter re-uptake in a matter of hours, but it takes several weeks for patients to begin to feel less depressed.
This time lag posed a challenge to the notion that the primary cause of depression was related to a reduced level of neurotransmitters in the synapse. In recent years, many studies have investigated long-term adaptive changes produced by antidepressant drugs on norepinephrine and serotonin systems.\textsuperscript{54} In addition, although most current antidepressant drugs are known to act on norepinephrine and serotonin systems, other factors appear to be involved in the mechanism of action of antidepressants. Dopamine, neuropeptides, neurohormones, intracellular second messengers, and modulation of gene expression may play a role in the mechanism of action of antidepressant drugs.\textsuperscript{55,42}

1.5 Herbs for Depression

**Folic Acid (folate) and Vitamin B12**

Folic acid and vitamin B12 are essential in several metabolic pathways in the central nervous system (CNS) and their metabolism is intimately related.\textsuperscript{56} Both folate and vitamin B12 participate in the synthesis of norepinephrine, dopamine, and serotonin and also act as coenzymes in the methylation pathways that synthesize S-adenosylmethionine (SAM).\textsuperscript{57,58} SAM is the main methyl donor in the brain and has
been shown to possess effective antidepressant properties.\textsuperscript{57} Folic acid is required for the synthesis of SAM by donating its methyl group to homocysteine to form methionine (a reaction catalyzed by vitamin B12), which is the immediate precursor of SAM.\textsuperscript{56,59-61} In folic acid and vitamin B12 deficiency this reaction may become severely impaired resulting in reduced SAM synthesis and a considerable elevation in plasma homocysteine levels.\textsuperscript{60} The neurotoxic effects of elevated homocysteine may also play a role in the neurologic and psychiatric disturbances that are associated with folate and vitamin B12 deficiency.\textsuperscript{62}

In a study of 100 consecutive patient admissions with severe depression, low serum folate and vitamin B12 concentrations were found in 24\% and 13\% of the cases, respectively.\textsuperscript{2} Low folate levels have been associated with mood disturbance, whereas low vitamin B12 has been more often associated with organic disorders and psychosis.\textsuperscript{62}

More than a dozen studies indicate that between one-third and one-half of psychiatric patients have a folate deficiency.\textsuperscript{57,56} Folate levels have been shown to be significantly lower in patients with major depressive disorder than normal controls or other psychiatric patients.\textsuperscript{61,58,64} Lower folate concentrations were associated with a greater severity of depression. In a recent study, folate levels were measured in 213 patients with major depressive disorder.\textsuperscript{65} These patients were then treated with the antidepressant fluoxetine (Prozac) for 8 weeks to measure treatment outcome. The study showed that patients with low folate levels were more likely to have melancholic depression and were significantly less likely to respond to fluoxetine treatment.

There is growing evidence that, irrespective of whether the deficiency is primary or secondary to the psychiatric disorder, folate supplementation may improve mental function.\textsuperscript{16} In a doubleblind, placebo-controlled trial of 24 patients with major depression and borderline or definite red blood cell folate deficiency (<200 \(\mu\)g/l), patients received 15 mg/day oral methylfolate or placebo for 6 months in addition to standard psychotropic treatment.\textsuperscript{66} The methylfolate group showed significant improvement in clinical and social recovery scores compared to the placebo group.
In an open study of 36 chronic alcoholics with major depression, patients were given 90 mg/day methylfolate for 4 weeks as an antidepressant.\textsuperscript{59} Significant improvement of depressive signs and symptoms was reported with no adverse side effects. While some studies have given patients up to 90 mg/day methylfolate without side effects, folate given in pharmacological doses (15 mg/day) to \textit{healthy} volunteers was associated with toxic effects such as altered sleep patterns, malaise, irritability, and over-activity.\textsuperscript{58}

However, most studies of depressed patients have used 15 mg/day without side effects. One researcher suggests that, because a significant effect was obtained with a dose of 0.2 mg/day in one study, doses below 1 mg/day may be appropriate for folic acid supplementation, which is more than the usual daily dietary intake.\textsuperscript{67} It appears that a dosage range of 0.2 mg to 15 mg of folic acid per day is appropriate.

\textbf{Inositol}

Inositol is a simple sugar-like compound present in the normal diet that acts as a messenger within cells.\textsuperscript{68} It is the precursor of the phosphatidylinositol (PI) second messenger system in the brain. The PI cycle is the second messenger system for numerous neurotransmitters including serotonin. It is hypothesized that inositol may be deficient in some brain systems in depression; low inositol levels could cause second-messenger dysfunction and thereby depression.\textsuperscript{68,69} In Europe, over-the-counter inositol has long been used as a folk remedy for anxiety and depression.\textsuperscript{68} However, inositol passes the blood-brain barrier poorly and doses needed for clinical studies are high (6 g to 12 g/day).\textsuperscript{69} After an encouraging open trial of 6 g/day of inositol for treatment-resistant depression, researchers performed a double-blind controlled trial for 28 depressed patients (12 g/day for 4 weeks). Significant overall benefit from treatment with inositol compared to placebo was found at week 4 but not week 2 on the Hamilton Depression Scale. A follow-up double-blind controlled trial of inositol treatment in panic disorders revealed significant benefit from the inositol (12 g/day for 4 weeks). No significant side effects were reported.
St. John’s Wort

St. John’s wort (*Hypericum perforatum*) has been widely researched for its antidepressant effects and recent research has focused on its potential as an antiviral agent. It has a long history of use for a multitude of indications: vulnerary (wound healing), diuretic, depression, neuralgic disorders, anti-inflammatory, and as a sedative.\(^{70,71}\)

St. John’s wort has become increasingly popular in Germany, where in 1994, 66 million daily doses were prescribed for use in the treatment of depression. Like synthetic antidepressants, St. John’s wort extract needs 2 to 4 weeks to develop its mood elevating effects. And while it is considered to be safe with no apparent side effects, it should not be taken with other psychoactive drugs. When consumed in large amounts, it has been associated with photosensitivity in animals; however, photosensitization does not usually occur when used within its recommended dosage range (0.5 to 3.0 mg hypericin/day).

St. John’s wort contains numerous compounds with documented biologic activity. Many researchers consider its effects to be due to a variety of constituents rather than any single component. These include hypericin, pseudohypericin, quercetin, hyperin, hyperforin, and xanthones. Continued research is needed to identify the constituents most responsible for St. John’s activity so that preparations can be optimally standardized. The current procedure of standardizing to hypericin content appears to result in an active product and may be better than no standardization.\(^{71}\) St. John’s has been tested in more than 3,000 patients against placebo and various active medications.

Researchers from Germany and the U.S. recently published a meta-analysis of 23 randomized trials of extracts of St. John’s wort with a total of 1,757 outpatients with mild to moderately severe depressive disorders.\(^{72}\) Comparisons were made of St. John’s wort alone, in combination with other plant extracts, to placebo, and/or a standard antidepressant. Twenty of the 23 trials were double-blind, one was single-blind, and two were open. Most were 4 to 8 weeks in duration and the total extract dose ranged from 300 to 1,000 mg/day (0.4 to 2.7 mg hypericin). In 13 studies
comparing a single St. John’s wort preparation with placebo, 55.1% of patients receiving the herb improved, compared with 22.3% responding to placebo.\textsuperscript{32} In the comparisons to standard antidepressants in 3 trials with single St. John’s wort preparations and 2 trials with combinations (St. John’s wort and \textit{Valeriana}), 63.9% of patients responded to single preparations compared with 58.5% to standard antidepressants and 67.7% responded to combination extract products compared with 50% to standard antidepressants. The researchers concluded that “\textit{Hypericum} extracts were significantly superior to placebo and similarly effective as standard antidepressants” while producing fewer side effects (19.8 % vs. 35.9%).\textsuperscript{75} While some of these studies were flawed and there is a lack of long-term studies, St. John’s wort definitely shows promise in the treatment of mild to moderate depression. The evidence suggests that St. John’s wort is more effective than placebo for the treatment of depressive disorders; however, evidence is inadequate to establish whether St. John’s wort is as effective as other antidepressants.

Studies are also needed for severely depressed patients, as are long-term studies to assess the risk of relapse and emergence of latent side effects.\textsuperscript{73} The mechanism of action for St. John’s for treating depression is unknown, although there are several theories.\textsuperscript{72} Hypericin and pseudohypericin may act on immune cells that secrete chemicals that cross the blood-brain barrier. St. John’s either lowers levels of cortisol or acts on GABA receptors on brain cells. Hyperforin, another compound found in the herb, may increase brain levels of serotonin. St. John’s wort extract may reduce cytokine expression (interleukin-6), the hypothesis being that interleukins can induce depression in susceptible individuals.

It is important to choose a St. John’s wort preparation standardized to a level of hypericin that permits a daily dose of approximately 2.7 mg hypericin. To be consistent with the form used in clinical trials, the preparation should include all of the other naturally occurring substances that are found in St. John’s wort and not just purified hypericin.
Other Herbs

There are numerous plants in addition to St. John’s wort that have been identified in ancient herbal texts from around the world to “settle the spirit,” “lift the mood,” and “clear the mind.” Several of these herbs have been found to possess potent naturally occurring chemicals that support these traditional observations by influencing various organ systems that play a role in mood, anxiety, and mental function. These include salvia root (Salvia miliorrhiza), rosemary herb (Rosamariam officinaridis), lavender (Lavandula spp.), California poppy (Eschscholzia californica), licorice root (Glycyrrhiza spp.), and jujube seed (Zizyphus spinosa). For example, jujuboside, one of the active ingredients of jujube seed, has been found to possess sedative and tranquilizing properties and protopine, found in California poppy, is an alkaloid found to act as a sedative and muscle relaxant.

In traditional Chinese medicine, managing emotional and mental problems involves choosing herbs that nourish the heart. It is interesting to note that two herbs frequently used in traditional Chinese medicine to enhance mood, salvia root and jujube seed, have also been found to influence the cardiovascular system. Because poor circulation, particularly evident in the elderly, can contribute to depression, herbs such as hawthorn berry (Crataegus pinnatifida) and Ginkgo biloba are useful because they increase blood flow to the brain. The addition of these herbs can enhance the targeted antidepressant effect of St. John’s wort by supporting multiple organ systems within the body, such as the adrenal glands, liver, cardiovascular system, and selected neurotransmitter functions.

1.6 Flavonoids in treatment of depression

Flavonoids have found to be ligands for GABA receptors in the central nervous system and it led to hypothesis that they act as benzodiazepine-like molecules. Many flavone derivatives were found to be ligands for the GABA receptors in the CNS; and thus they bind to the benzodiazepine binding site with resulting depressant actions in mice. These were also found to possess sedative action, tranquilizers, and anticonvulsant. Considering the sedative, the spontaneous locomotor activity and thiopental-induce sleeping time effects obtained with the flavonoid glycosides, the
following decreasing order of action results:

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<th>Compound</th>
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<tbody>
<tr>
<td>2S-hesperidin</td>
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<td>rutin</td>
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<tr>
<td>diosmin</td>
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<td>cong2S-neohesperidin</td>
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<td>gossypin</td>
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<tr>
<td>2S-naringin</td>
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Position of the sugar on the flavonoids nucleus seems relevant as well and position-7 is the most effective but the presence of a double bond between carbons 2 and 3, resulting in flavone derivatives with planar configuration (i.e. larinin) does not appear to be critical for activity. Flavonoid glycosides form the newest group within the growing family of flavonoids with activity on the CNS. Yi et al.; reviewed that dietary flavonoids possess multiple neuroprotective actions in Central nervous pathophysiological conditions including depression and it was reported that naringenin possess potent antidepressant-like property via central serotonergic and noradrenergic system. It was further suggested that dietary flavonoids possess a therapeutic potential in disorders especially where monoaminergic system is involve.

1.7 Polyphenols as neuroprotective

The neuroprotective effects of many polyphenols rely on their ability to permeate brain barrier and here directly scavenge pathological concentration of reactive oxygen and nitrogen species and chelate transition metal ions. Different polyphenolic compounds were shown to have scavenging activity and the ability to activate key antioxidant enzymes in the brain and thus breaking the vicious cycle of oxidative stress and tissue damage. Moreover, red wine polyphenolic compounds have been documented to increase endothelial NO synthase (eNOS), while downregulate inducible NO synthase (iNOS) activity in different structures. Documented that red wines strongly inhibit the synthesis of endothelin-1, wellknown vasoactive peptide, that is crucial in the development of coronary atherosclerosis. Previsously, the pathophysiological role of endothelin in the brain was documented as well. Attenuation of the release of cytokines, such as interleukin-1beta (IL-1beta) and tumor necrosis factor-alpha (TNF-alpha) was also ascriptius to different polyphenols. Thus, natural polyphenolic compounds may protect and/or improve physiological brain functions by different mechanisms that include mainly antioxidant activity, eNOS/iNOS balance, inhibition of endothelin production and attenuation of cytokine release.
1.8 Depression and animal models

Depression is a heterogeneous, multifaceted disorder with symptoms manifested at the psychological, behavioural and physiological level. This is perhaps why it is so difficult to mimic the disorder in the laboratory. Many of the human symptoms of depression, as described in the Diagnostic and Statistical Manual of the American Psychiatric Association (DSM IV) (such as recurring thoughts of death or suicide or having excessive thoughts of guilt) are impossible to be modeled in mice. The question, therefore, remains impenetrable as to whether we can ever assume a mouse is ‘depressed’. Evolutionary theories have been proposed for psychiatric disorders, which would plausibly predict that also lower animal species can exhibit behaviours useful in modeling human depression. However, such hypotheses are heavily debated and are difficult to address empirically.

Another difficulty in assessing depressive states in rodents is that the underlying pathophysiology in depression is still unresolved. Further, the mode of action of clinically effective antidepressants is not yet understood beyond the fact that they primarily alter monoamine neurotransmission. Despite the difficulties in translating the complexities of human affective disorders in its entire spectrum into relevant tests in mice, numerous attempts have been made to create so-called animal models of depression, or at least models of some of the core aspects of depression. Such models include those paradigms where various stress, pharmacological, lesion, environmental or genetic manipulations are applied.

Efforts to gain a better understanding of the underlying pathophysiology of affective disorders and the mechanism of action of antidepressant medications are most recently driven by technical advances in the field of molecular genetics. Indeed, it is hoped that the recent elucidation and the ongoing functionalization of the human genome may provide new insights into the etiology, course and hence treatment strategies for psychiatric illnesses such as major depression. Another avenue taken for better understanding psychiatric disorders has been the development of mice with genetically altered expression of a specific protein, be it a receptor, transporter, enzyme or signal transduction molecule (for recent reviews on various
technologies. These animals have the potential to give rise to novel targets for antidepressant activity for which pharmacological tools may not exist. Additionally, these mice will help to evaluate in vivo the validity of current molecular theories of depression. Presently, we are aware of over 40 distinct strains of genetically modified mice which have a phenotype possibly related to depression or antidepressant drug action.

1.8.1 Forced Swimming Test

Of all the experimental procedures used in preclinical depression research the forced swim test (FST) (a.k.a. Porsolt’s test; behavioural despair test) is probably the most widely and most frequently used. The FST changed the way that drug screening for antidepressants is carried out, largely due to its relative reliability across laboratories and its ability to detect activity of a broad spectrum of clinically effective antidepressants. Furthermore, this test is the most widely used paradigm to assess ‘depression’ and antidepressant-related phenotypes in genetically altered mice. The test is based on the observation that rodents, following initial escape-oriented movements, develop an immobile posture in an inescapable cylinder filled with water. If antidepressant treatments are given prior to the test, the subjects will actively persist engaging in escape-directed behaviours for longer periods of time than after vehicle treatment. For reasons not yet elucidated, in mice, one exposure is sufficient to generate a stable immobility readout that can be countered by acute pretreatment with antidepressant agents whereas generally two trials are required in the rat version.

The other important reason for the popularity of the FST as a model is its ability to detect the antidepressant-like effects of drugs from a variety of classes after acute administration which allows for rapid screening of novel drugs. The realization that meaningful data could be obtain from an acute animal assay that was not based on manipulation of a specific neurotransmitter system (as in the case of reserpine-induced hypothermia and yohimbine lethality assays), led to the development of other assays which involve assessing a behavioural response to a stressor. Chief among these was the introduction of the TST.
1.8.2 Tail Suspension Test (TST)

TST is based on the observation that rodents almost always mice although gerbils and rats have been used, after initial escape-oriented movements, develop an immobile posture when placed in an inescapable stressful situation. In the case of the TST the stressful situation involves the haemodynamic stress of being hung in an uncontrollable fashion by their tail whereas in the FST mice are placed in a cylinder filled with water. If antidepressant treatments are given prior to the test, the subjects will actively persist engaging in escape-directed behaviours for longer periods of time than after vehicle treatment. The test is usually quite short, 6 min, and the amount of time they spend immobile is recorded either manually or through an automated device. Acute antidepressant treatments decrease these immobility scores. An obvious advantage of this test is its ability to detect a broad spectrum of antidepressants irrespective of their underlying mechanism, it is inexpensive, methodologically unsophisticated and easily amenable to automation. Tomation also enables the assessment of additional parameters such as power of movement and more recently energy of movement. The advent of genetically modified mice has put ded emphasis on the use and development of murine models of depression. Thus, the use of the TST has substantially increased in recent years as a model for assessing antidepressant-relevant behaviour. Although both the FST and TST are similar in the constructs that they purport to assess they are probably different in terms of the biological substrates that underlie the observed behaviour although they often offer converging data on a potential antidepressant.

1.8.3 Immobility and animal models for depression

Many hypotheses have been advanced to explain the physical adaptation that is the immobility response observed in the FST and TST. The posture of immobility in the context of the FST was originally coined ‘behavioural despair’ by Porsolt (1978), largely based on the assumption that the animals have ‘given up hope of escaping’. In other words, the immobility represents a failure of persistence in escape-directed behaviour. Other investigators have contended that the behavioural responses comprise an evolutionary preserved coping strategy, in which immobility behaviours represent the psychological concept of “entrapment”
described in clinical depression.\textsuperscript{115-116} Thus, the development of immobility disengages the animal from active forms of coping with stressful stimuli.\textsuperscript{116} Further, immobility in the TST is due to inability or reluctance to maintain effort rather than a generalized hypoactivity, as evidenced by the fact that animals can adopt this posture quickly and drugs which may suppress activity (as is the case of many antidepressants) counter the immobility response. As such, this immobility may be analogous to the clinical observations that depressed patients often lack sustained expenditure of effort reflected in a pronounced psychomotor impairments.\textsuperscript{117}

Immobility is also part of the behavioural repertoire of animals in other contexts. One such response that has been well studied is the freezing behaviour following exposure to an aversive stimulus such as shock, the environmental context where the shock was previously given or a predator.\textsuperscript{118} However, this is considered a markedly different response to that seen in the TST, because it is largely defensive in nature and is more rapidly revealed upon re-exposure to the stimulus. Another form of immobility in animals, which is also largely a defensive response, is tonic immobility, which represents the unlearned reaction to brief manual restraint, which is characterized by a catatonic-like state of reduced responsiveness; its duration is thought to be positively related to the antecedent fear state.\textsuperscript{119} Tonic immobility has mainly been characterized in avian species but has been demonstrated in mammalian species such as the guinea pig.\textsuperscript{120} Another immobility-like behaviour is seen after repeated shocks in the learned helplessness procedure, a learned response.\textsuperscript{121} However, in contrast the TST-induced immobility is not a learned behaviour per se, as only one exposure is required to reliably detect antidepressant-like behaviours. Nevertheless, it is probable that the TST requires within session instrumental learning to engage in immobility.

1.8.3.1 Learned helplessness test

The learned helplessness (LH) animal model incorporates more closely disease etiology and predisposition and attempts to simulate a human depressive state in animals.\textsuperscript{122} It was first described in dogs,\textsuperscript{123} but currently it is used in rodents, both mice and rats. It is based on the observation that the animal develops deficits in
escape, cognitive and reward behaviours (sucrose preference) when subjected to repeated unavoidable and uncontrollable foot shocks, achieving the state of leaned helplessness. This state is defined as a failure to exhibit escape behaviour during subsequent exposure to the same stressful stimulus, only this time escape is possible.\textsuperscript{124} This acceptance of the uncontrollable situation was accepted as being analogous to the apathetic despair seen in human depression.\textsuperscript{125}

For yet unknown reasons, only a part of the animals subjected to this stress develop signs of helplessness, reflecting a potential environment interaction. Other symptoms present after exposure to this test are: loss of appetite and weight, decreased locomotor activity and poor performance in appetitively and aversively motivated tasks, equivalent to depressed human symptoms.\textsuperscript{98}

1.8.3.1 Brain lesion models

These paradigms are based on the assumption that depression might be caused by regulatory deficits in neuronal circuits, which causes a constellation of behavioural and neurochemical alterations. Bilateral olfactory bulbectomy is a widely used procedure which induces changes in behaviour, and in the endocrine, immune and neurotransmitter functions, which simulate many of the changes, observed in depressed patients. Since the olfactory system in rodents is part of the limbic region, along with the hippocampus and the amygdala, which contribute to memory and emotion, the removal of the bulbs result in a disruption of the limbichypothalamic axis causing all the alterations mentioned before.\textsuperscript{126}

In this model, the observed behavioural alterations are not just a consequence of loss of smell, since peripheral anosmia does not produce such a syndrome. Therefore, the behavioural syndrome connected to olfactory bulb must be the result of a major dysfunction of the cortical-hippocampal-amygda circuit. These areas are also impaired in depressed patients.\textsuperscript{126} This procedure has been predominantly applied in rats, but mice can also be used. After performing and olfactory bulbectomy in rats, marked changes in all major neurotransmitter systems occur.
Hyperactive response in the open field paradigm is the most consistent behavioural change observed, and it can be reversed by antidepressant medications.\textsuperscript{127} As chronic, but not acute, administration of antidepressants corrects the majority of the alterations caused by olfactory bulbectomised, this model is considered not only a model for detecting antidepressant activity\textsuperscript{95,126} but also one for exploring the connection between these systems.\textsuperscript{126} As this model mimics the slow onset of antidepressant action (chronic action), it shows high face validity, thus having one of the best preclinical profiles for assessing the effects of novel antidepressants.\textsuperscript{95}