EVALUATION OF ANTIDEPRESSANT EFFECT OF*CYMBOPOGON CITRATUS* (LEMON GRASS) IN ALBINO MICE

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Madurai

.09.2016

CERTIFICATE

This is to certify that the dissertation entitled **"EVALUATION OF ANTIDEPRESSANT EFFECT OF *CYMBOPOGON CITRATUS* (LEMON GRASS) IN ALBINO MICE"** is a bonafide record of work done by **Dr.R.MANGALADEVI,** under the guidance and supervision of **Dr.R.SAROJINI,M.D.,** Professor, in the Institute of Pharmacology, Madurai Medical College, Madurai during the period of her postgraduate study of M.D Pharmacology from2014-2017.

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DECLARATION

I, **Dr. R. MANGALADEVI,** solemnly declare that the dissertation titled **"EVALUATION OF ANTIDEPRESSANT EFFECT OF*CYMBOPOGON CITRATUS* (LEMON GRASS) IN ALBINO MICE"** has been prepared by me under the able guidance and supervision of **Dr.R.PARAMESWARI M.D,** Director and Professor, Institute of Pharmacology, Madurai Medical College, Madurai, in partial fulfillment of the regulation for the award of M.D Pharmacology degree examination of the Tamilnadu Dr.MGR Medical University, Chennai to be held in April 2017.

This work has not formed the basis for the award of any degree or diploma to me, previously from any other university to anyone.

Place: Madurai **Dr. R. MANGALADEVI**

Date:

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CONTENTS

ANNEXURES

INTRODUCTION

Depression is a common psychiatric disorder, associated with poor concentration, lack of interest, loss of pleasure, guilt, altered sleep and appetite. There is also an increased suicidal tendency.

It is an oldest disease mentioned by Hippocrates in 400 BC as melancholia. Now the disease prevalence is so high. WHO estimates 121 million people are suffering from depression worldwide. It contributes to 4th leading cause of global disease burden. Women suffer from depression more than men with a point prevalence of 3.2% in women and 1.9% in men[1].

The outlook of management of depression was gloomy previously. Most of the psychiatric problems were treated in asylum. Now with a better understanding of etiopathogenesis of the disease, with advent of new drugs and other treatment modalities for depression, it is possible to treat the disease at primary health care set up.

The cornerstone in the management of depression is by drugs. By combining non pharmacological therapy like Cognitive Behavioral Therapy (CBT) and interpersonal therapy it is almost curable[2].

Pharmacotherapy is the most effective intervention in the management of Major Depressive Disorder (MDD). It helps to achieve remission and also prevents relapse. Among the drugs available Selective Serotonin Reuptake Inhibitors (SSRIs) and Serotonin Noradrenalin Reuptake Inhibitors (SNRIs) are the most commonly used drugs. They belong to second generation antidepressant drugs. Tricyclic Antidepressants

(TCAs) and Monoamine Oxidase Inhibitors (MAOIs) are referred to as first generation drugs but currently not preferred because of lack of selective targeting and an array of adverse effects like cardiac conduction abnormalities.

The drugs employed in the management of MDD either decrease the degradation or block the reuptake of noradrenaline and serotonin at the synapse. Among the many newer antidepressants available, SSRIs are claimed to be safe with better tolerability profile but they too have adverse effects[3].

Noncompliance is approximately 40% in patients seeking primary care, receiving drug therapy alone because of delayed onset of therapeutic effects unless otherwise supported by other modalities like cognitive behavioral therapy (CBT).In a good compliant patient too, who is adequately managed with drugs alone, remission is only in 60-70%[4]. However, no drug is free from side effects. As all classes of antidepressants have almost similar efficacy, it is the unwanted adverse effects which are important in choosing a drug for a particular individual.

Hence, the hunt for newer drugs in the management of depression is an ongoing topic of research. An ideal drug exhibiting better safety and tolerability, which cures depression most efficiently is sought for.

In order to bridge the defects in the modern medical system, complementary and alternative medicine helps. Natural products from plants have played an important role in the lives of human beings from time immemorial. It is a belief that traditional medicine

lack efficacy and evidence and are inferior to allopathic medicine. However, it is not so. Traditional medicine helps in conditions like sleep disorders, cardiovascular disorders, chronic pain, depression, obesity, erectile dysfunction, common cold, etc.,

Many effective drugs have come from a wide armamentarium of herbs. Drugs like morphine, aspirin, vincristine, vinblastine, artemisinin, atropine, taxanes etc., are derived from herbs. Natural products like St. John's wort from *Hypericum perforatum*, *Crocus sativus* (saffron), *Rhodiola rosea* (roseroot) are effective for treating mild to moderate depression[5].

As Hippocrates mentioned, "Let food be thy medicine and medicine be thy food", our daily consumption of vegetables have medicinal effects. Plant based diet and culinary herbs contain various phytochemicals with medicinal properties. Traditional medicine is easily accepted, less expensive, and can be self administered. Since no medicine is free from side effects, herbal medicines must be subjected to proper clinical trial.

In the pathogenesis of depression, stress is attributed to cause decreased neurotransmitters in brain, increased free radical formation and injury to the neurons. Phytochemicals extracted from natural products are good sources of antioxidants and act as stress adaptogens, and help in alleviating depression. Evaluation of antidepressant property is done in many herbs like roots of *Curcuma longa*, leaves of *Mimosa puolica*, roots of *Ocimum sanctum*, root of *Withania somnifera*.

Cymbogon citratus, which is known by its common name as lemon grass is often used as a flavouring agent in cooking and in aroma therapy. This plant has also got a wide range of medicinal properties and it is in use as a folk remedy in many parts of the world.

The essential oil prepared from lemon grass exhibits antifungal, antimalarial, larvicidal, antifungal, antibacterial, antiamoebic, antinociceptive, ascaricidal effects. Aqueous extract prepared from fresh leaves shows anticancer, antioxidant, hypoglycemic, hypocholesterolemic, antifilarial, antidiarrhoeal, anti-inflammatory effects and antidepressant property[6].

Since an ideal antidepressant is still awaited, and it is always an area of research, lemon grass, a common culinary herb having antidepressant property is selected for this study.

Screening methods for evaluation of antidepressant property are always difficult because of lack of a typical animal model. Of the many in vivo animal models available, we planned to study the antidepressant activity of lemon grass in Swiss albino mice by tail suspension method.

AIM

AND

OBJECTIVES

AIM

To evaluate of antidepressant effect of *Cymbogoncitratus* (Lemon Grass) in albino mice by tail suspension test.

REVIEW

OF

LITERATURE

Psychiatric disorders are common in medical practice. They are a group of disorders, present as a primary condition or associated with some other medical problems, characterized by altered regulation of mood, behavior and affect.

Among the psychiatric disorders depression is the most common one. It is a disorder of mood, with intact cognition or thought. Mood is an inner feeling which influences one's behavior whereas affect is external expression of mood. A spectrum of mood either normal or low or high is experienced by us. Healthy individuals have control over their mood and affect whereas when the control is lost, mood disorder occurs.

Mood disorders are classified broadly into unipolar depression and bipolar depression. Unipolar depression is characterized by depressive episodes only, whereas in bipolar depression, the manic episode alternates with depressive episodes. Other mood disorders include hypomania, cyclothymia and dysthymia[7].

Depression is actually a syndrome, may present with an array of heterogeneous symptoms, including physical, behavioral, cognitive, impulse control or vegetative symptoms. Mania is exactly opposite to depression, characterized by euphoria, flight of ideas, insomnia, aggressive behavior, grandiose delusions, increased physical and mental activity, and pleasure seeking behavior.

About 75% unipolar depression patients are non-familial, having a strong correlation with stressful life events; remaining 25% patients have no associated distress

called as endogenous depression. Bipolar depression appears in early adult life. There is a strong evidence of genetic predisposition.

Although Hippocrates described mental disturbances in 400 BC, as mania and melancholia, till date proper understanding of the disease per se is difficult. This is partly due to difficulty in creating animal models for depression which cannot express mood changes similar to human beings. Most of the times, therapeutic interventions and practical experience undertaken on trial and error basis had only thrown light in understanding depression rather by a methodical experimental way.

EPIDEMIOLOGY

Mood disorders contribute to one of the leading causes of morbidity all over the world ultimately resulting in a great economic loss to the society due to loss of productivity and utility of medical resources.

True prevalence of depression is underdiagnosed because of social stigma attached to it. It is estimated that approximately 121 million people are suffering from depression worldwide. Depression can occur in any age group. The most common age of onset of depression is around late twenties. First episode may occur at any age. Adults between age group of 18 to 29 years experience the highest rate of major depression during any given year.

The life time risk for a major depressive disorder is 10 to 25% for women and 5 to 12% for men. The point prevalence for adult men is 2 to 3 % while 5 to 9% for adult

women[8]. Women are at increased risk of depression from early adolescence until their mid 50's with a life time rate that is 1.7 to 2.7 times greater than that for men.

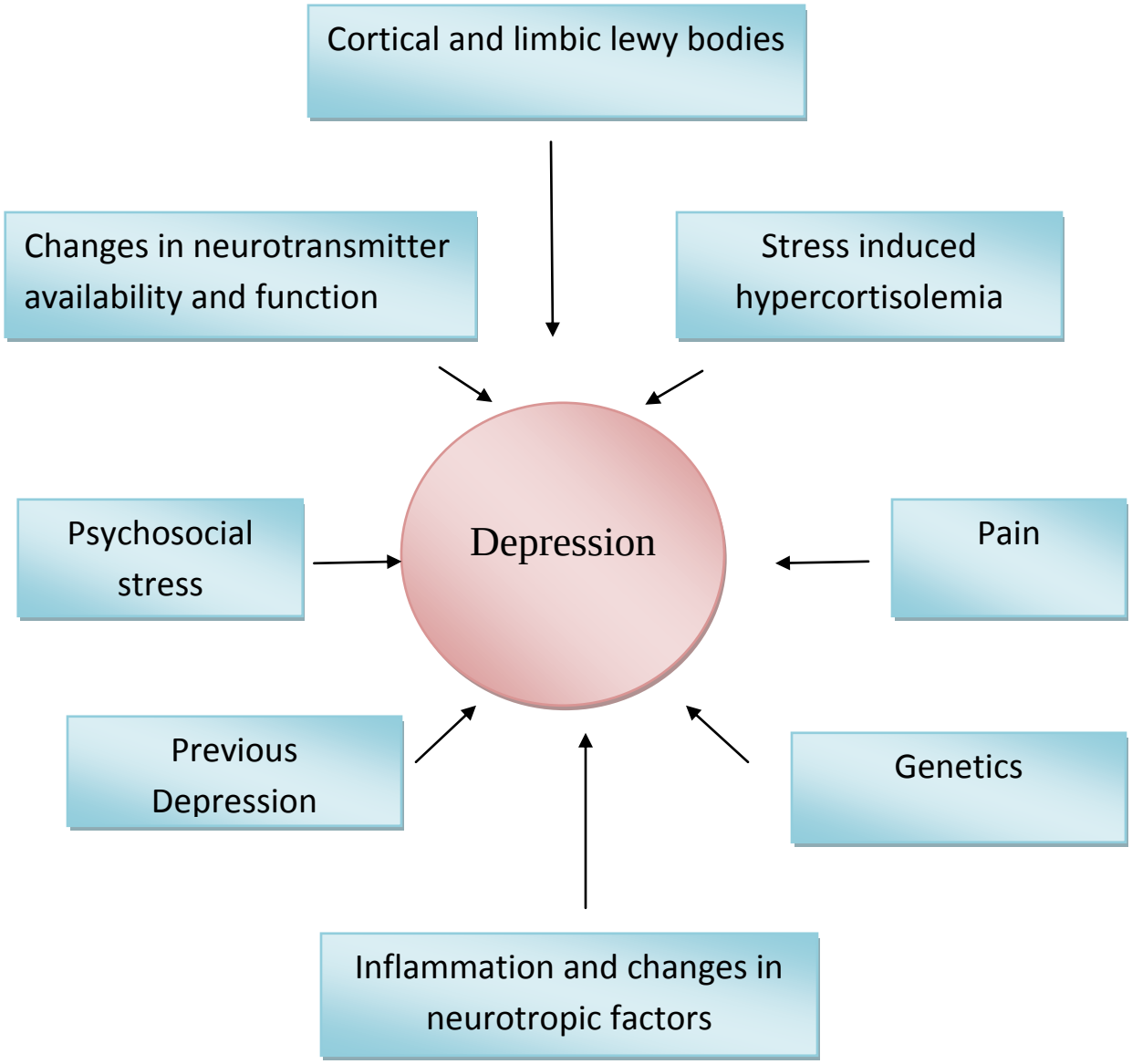
Genetic factor play a major role in causing depression. There is a relationship between parents suffering from depression and episodes of depression in their offspring. The chance for the off spring to get depression is 2.7 times more when one parent is suffering from depression whereas it is 3 times more if both the parents are suffering[9].

About 3 to 4% of Indians suffer from major mental disorders; 7 to 10% of the population suffers from minor depressive disorder. There is a high rate of suicidal tendency with depression. The primary diagnosis of depression is made retrospectively in 50% of patients who have committed suicide. In India 120,000 people commit suicide every year. Indian union health ministry has estimated that 37.8% of suicides were below 30 years of age[10].

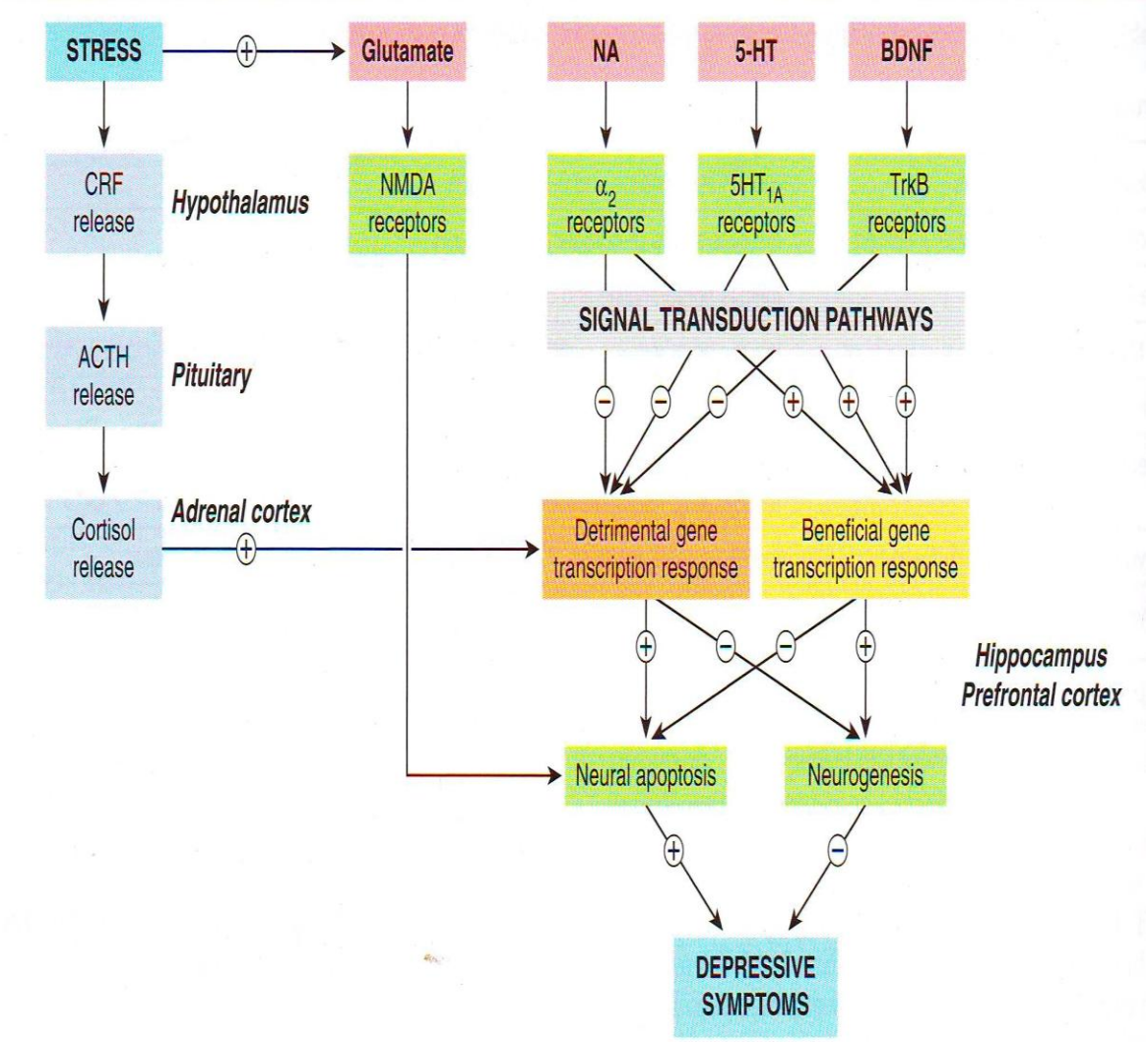
According to World Health Organization (WHO) it is estimated that the disease burden due to the major depression will go ahead than the disease burden due to road traffic accidents, cardiovascular and cerebrovascular diseases by the year 2030.

PATHOPHYSIOLOGY OF DEPRESSION

The pathophysiology behind the major depressive disorder has not been clearly identified. Preclinical and clinical studies have found altered monoamine neurotransmission in the central nervous system would be the basic pathology behind the development of depression[11]. Further researches has focused the deficiency of other neurotransmitters like noradrenaline, dopamine, glutamate may be linked to depression.



The role of central nervous system serotonergic activity in pathophysiology of major depressive disorder is suggested by the efficacy of serotonin reuptake inhibitors in the treatment of depression. Serotonergic neurons linked to depression are found in the dorsal raphe nucleus, limbic system and left prefrontal cortex



The concentration of serotonin metabolites is very low in the CSF of depressed patients. L-Tryptophan which is the precursor of serotonin is also decreased in depressed patients. Involvement of noradrenergic neurotransmission in depression is evident from

noradrenaline reuptake inhibitors such as reboxetine, nortryptiline producing antidepressant effect which is again confirmed by low levels of noradrenaline metabolite in the CSF of the depressed patients. In depression, dopaminergic neurotransmission is also altered; the pathognomonic symptom of depression – anhedonia is primarily mediated by dopaminergic neurons. Drug treatment which enhances dopamine neurotransmission that are selective dopamine receptor agonists may be a novel approach to treat SSRI nonresponders. There is also association of Parkinson's disease along with depression[12]. Dopamine deficiency is the main etiology in the development of Parkinson's disease and the metabolites of dopamine in CSF are also reduced in Parkinson's patients with depression.

Recent advances in the imaging techniques like MRI, has found a small reduction in hippocampal size.

PET SCAN FINDINGS IN DEPRESSION[13]

- Increased activation of the amygdala by negative stimuli
- Reduced activation of the nucleus accumbens by rewarding stimuli
- Decreased metabolic activity in neocortical structures
- Increased metabolic activity in limbic structures.
- Abnormally diminished activity in prefrontal cortex
- Small reduction in hippocampal size.

These areas are responsible for the regulation of noradrenergic, dopaminergic and serotonergic neurotransmission which have a prominent role in the regulation of mood.

There is also evidence of endocrine abnormalities. The prevalence of depression in postmenopausal women is high suggesting estrogen has a role in mood regulation. Low level of testosterone is associated with depression in elderly males[14].

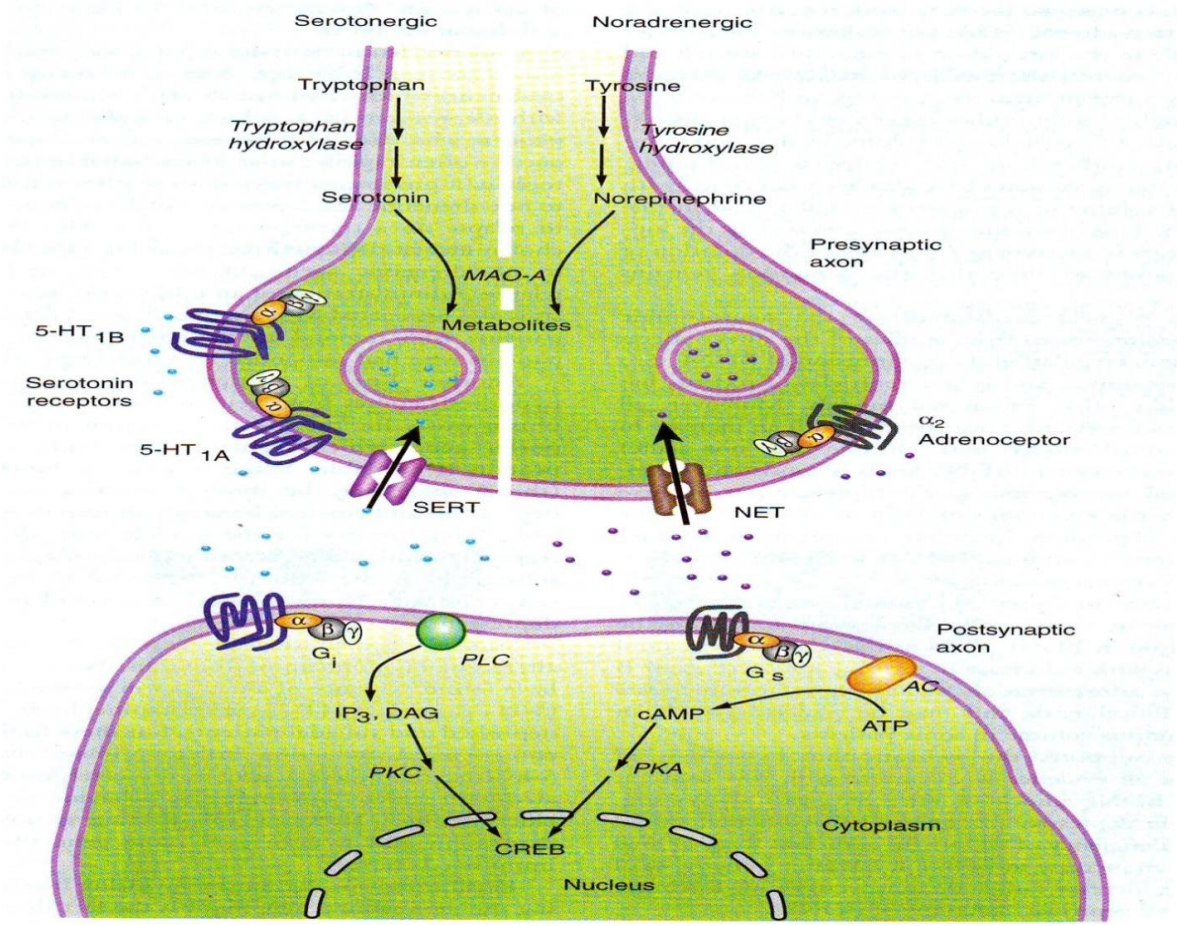
THEORIES OF DEPRESSION

BIOLOGICAL THEORIES

MONOAMINE HYPOTHESIS

The monoamine theory proposes that deficiency of monoamine neurotransmitters 5-hydroxytryptamine (5-HT) and noradrenaline (NA) at certain areas of brain including limbic system and cortical areas are linked to depression[15] whereas the excess leads to mania. This theory is supported strongly with the observations made after using drugs like reserpine, iproniazid and imipramine which resulted in unimaginable changes in mood.

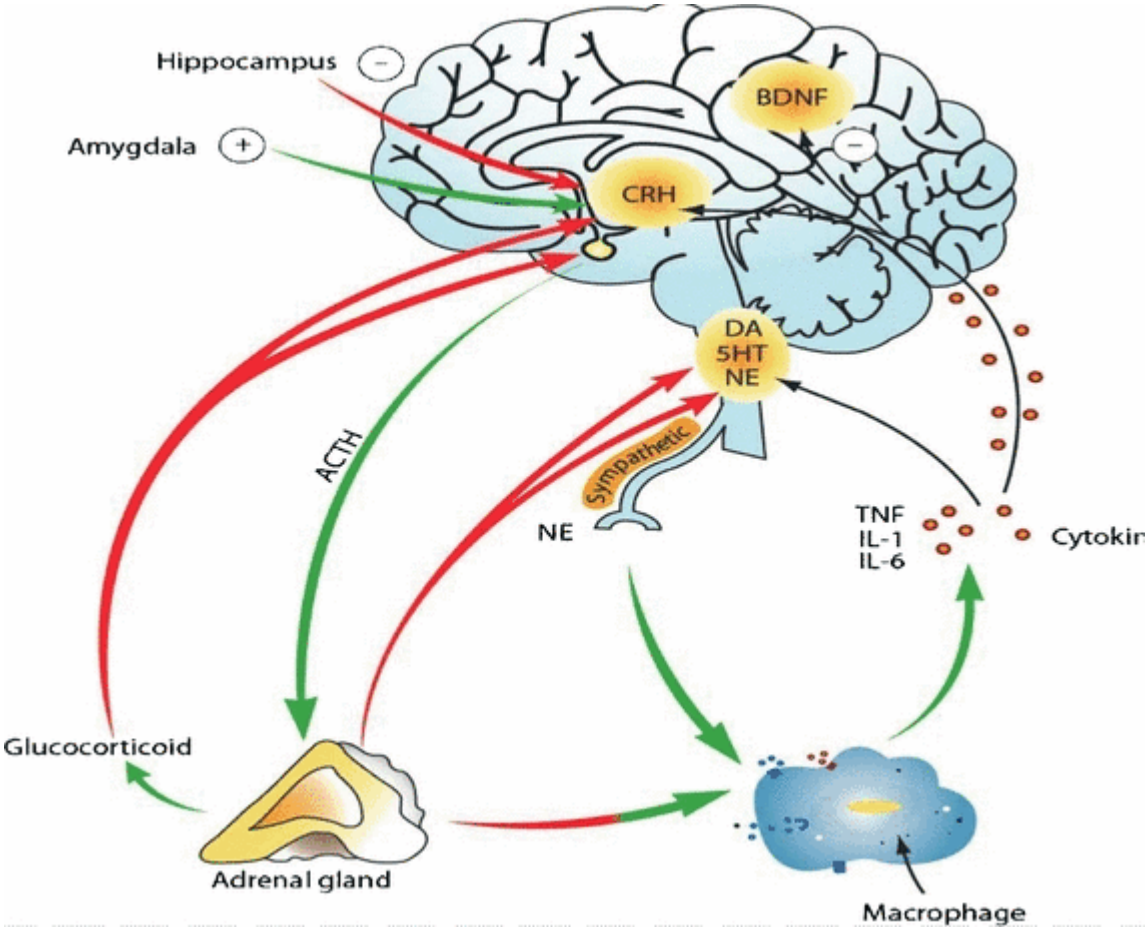
Imipramine which was introduced as an antipsychotic, was found to block 5-HT reuptake and its metabolite blocked noradrenaline reuptake thereby increasing the concentrations of 5-HT and NA at the synapse and increasing the postsynaptic activation of monoaminergic neurotransmission.



Iproniazid, which was used for the management of tuberculosis resulted in monoamine oxidase inhibition leading to excess neurotransmitter release like 5-HT, NA, dopamine and thus relieved symptoms of depression. Reserpine used as an antihypertensive caused depletion of monoamines. It binds to VMAT and blocked its function and the patients suffered from depression. Based on the above observations, monoamine hypothesis was postulated that major depression is a condition due to decreased monoamine neurotransmitters and depression could be reversed by increasing the neurotransmitters at the synapse.

Even though there is strong evidence, this theory has pitfalls. There is a time lag between the onset of action and antidepressant effect produced. Also some patients respond to drugs selectively increasing 5-HT and some patients to NA reuptake inhibitors. New antidepressants target other pathways and produce antidepressant effect. Monoamine hypothesis could not explain the above questions.

NEUROENDOCRINE FACTORS IN DEPRESSION

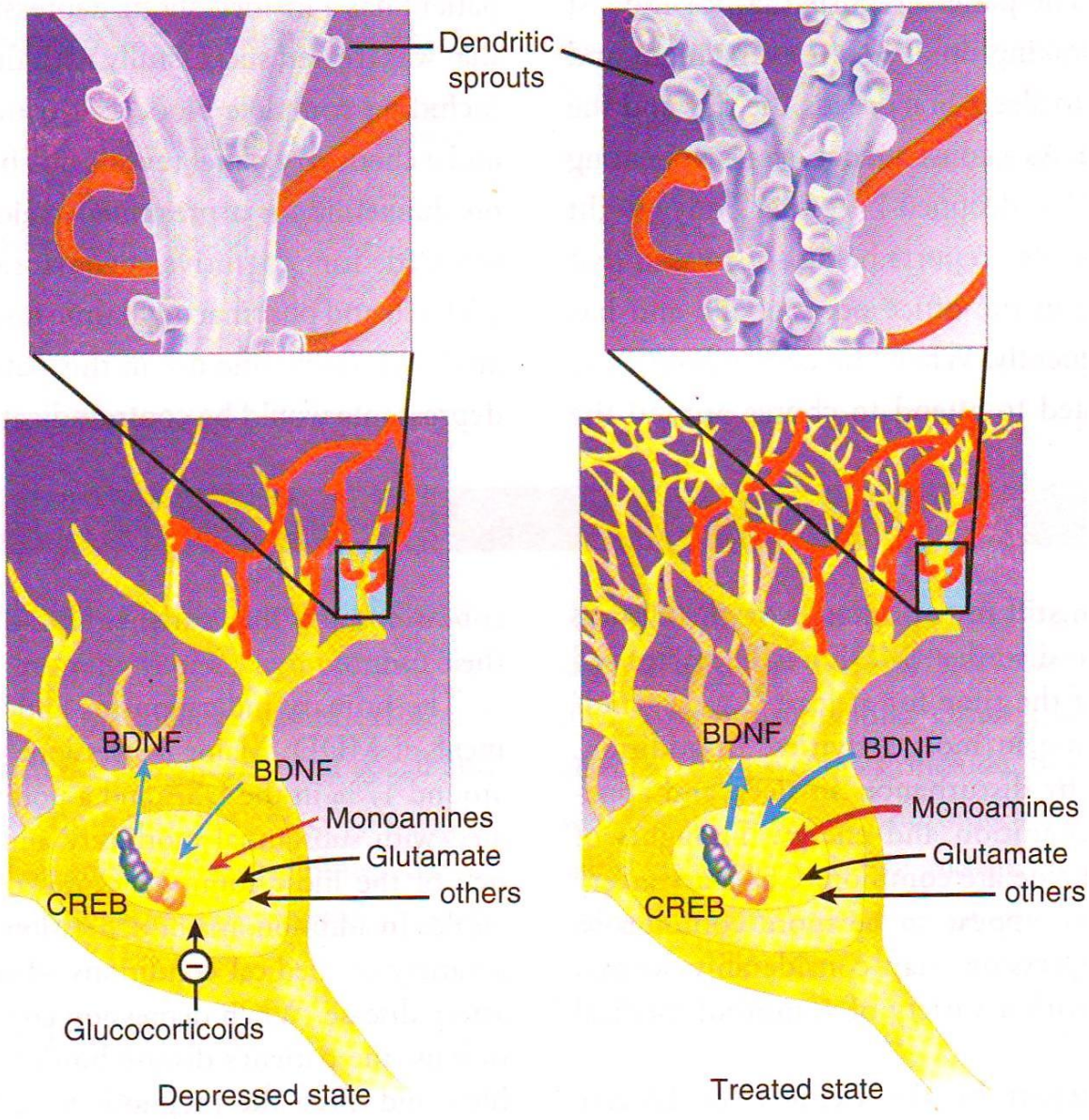


Patients with depression show abnormal HPA axis producing high CRH levels and high plasma cortisol levels which would not get suppressed with dexamethasone administration. Depressed patients have elevated CRH levels in CSF and brain[16].

Chronic stress, which is an important precipitating factor for depression, is also associated with high CRH levels.

B. NEUROTROPIC HYPOTHESIS

Brain Derived Neurotropic Factor (BDNF), a nerve growth factor, has a control over neurogenesis, neural plasticity and resilience[17]. It is proved that there is a direct association between development of depression and loss of neurotropic support.



With the effective antidepressant therapy, there is increased neurogenesis and synaptic connectivity in hippocampus. Stress and pain are associated with decreased levels of BDNF. Direct infusion of BDNF into midbrain, hippocampus and lateral ventricles of rodents has shown antidepressant activity. Chronic administrations of antidepressants have shown to increase the concentration of BDNF in hippocampus in animal models.

PSYCHOSOCIAL THEORIES

A. COGNITIVE BEHAVIOR THEORY

Depression results from maladaptation of cognitions which leads to distorted thoughts and judgments. A direct relationship is found between the amount and severity of someone's negative thoughts and severity of depressive symptoms[18]. The cognitive perspective as well as the contribution from the helplessness-hopelessness model forms an empirical basis for cognitive behavior theory.

PSYCHODYNAMIC THEORY OF DEPRESSION

According to Sigmund Freud, a person must successfully resolve early developmental conflicts in order to overcome depression and achieve mental health. Mental illness is a failure to resolve this conflict[19].

CLASSIFICATION OF MOOD DISORDERS

Based upon the criteria published by American Psychiatric Association, the diagnosis and classification of depression is done. According to the Diagnostic and Statistical Manual of Mental disorders, Fourth edition, Text Revision (DSM-IV-TR), depressive disorders are classified under mood disorders[20].

A. Depressive Disorders

1. Major Depressive Disorder, Single Episode
2. Major Depressive Disorder, Recurrent
3. Dysthymic Disorder
4. Depressive Disorder Not Otherwise Specified

B. Bipolar Disorders

1. Bipolar Disorder, Single Episode
2. Bipolar Disorder, Recurrent
3. Cyclothymic Disorder
4. Bipolar Disorder Not Otherwise Specified

C. Secondary Mood Disorder Due to Non psychiatric Medical Condition

D. Substance-Induced Mood Disorder

E. Mood Disorder Not Otherwise Specified

CLINICAL PRESENTATION OF DEPRESSION

The cardinal symptoms include loss of interest, low mood, fatigue. Depression must be diagnosed if any one of the above symptoms is present for at least a minimum period of 2 weeks along with other associated symptoms like sleep disturbance, guilt, lack of concentration, changes in appetite, suicidal tendency, agitation or physical slowness[21].

Many patients with depression present with physical symptoms which often confuse the primary care physician. Such patients express their emotional distress as bodily symptoms called as somatisation, leading to misdiagnosis of underlying depression. Depression can also results from many drugs and associated with other medical conditions[22]. So, before making the diagnosis of depression, always search for any underlying medical condition or drugs if any.

DIAGNOSIS OF MAJOR DEPRESSIVE DISORDER (MDD)

Presence of recurrent episodes of only depression is the feature of major depressive disorder (MDD). Whenever there is a single episode of either mania or hypomania with or without associated depression, diagnosis of bipolar disorder should be made. Hence while making a diagnosis of depression, a careful evaluation regarding previous episodes of mania or hypomania to be done.

The Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition, Text Revision (DSM-IV-TR) is the most widely accepted diagnostic reference. According to

DSM-IV, TR, MDD must be diagnosed after ruling out organic cause because depression will resolve if the medical cause is identified and treated or if the drug is withdrawn.

DRUGS CAUSING DEPRESSION

- Reserpine
- Clonidine
- Methyl dopa
- Hydralazine
- Guanethidine
- Steroids
- Oral contraceptives
- Isotretinoin
- L-Dopa
- Interferon-β
- Barbiturates
- Ethanol
- Varenicline

MEDICAL CONDITIONS ASSOCIATED WITH DEPRESSION

- Cancer
- Diabetes

- Multiple sclerosis
- Systemic lupus erythematosus
- Thyroid disease
- Addison's disease
- Viral infection
- Post stroke
- Post myocardial infarction
- Epilepsy
- Parkinson's disease

So before making the diagnosis of MDD, proper history taking, complete physical examination, psychiatric evaluation must be done. There is no single investigation or a biochemical parameter or imaging method to diagnose depression. It is based on patient's clinical symptoms. The standard diagnostic criteria by WHO- ICD – 10 criteria help to diagnose depression.\

ICD - 10 CRITERIA FOR DEPRESSION[23]

Cardinal Symptoms:

1. Persistent sadness or low mood
2. Loss of interest or pleasure
3. Fatigue or low energy

At least any one of these symptoms for most of the day and most of the time, for at least 2 weeks

Associated Symptoms

1. Disturbed sleep
2. Poor concentration or indecisiveness
3. Low self- confidence
4. Poor or increased appetite
5. Suicidal thoughts or acts
6. Agitation or slowing of movements
7. Guilt or self – blame

The above 10 symptoms define the degree of the depression.

Depression can be classified based on the symptoms as follows:

- No Depression - Fewer than four symptoms
- Mild Depression -Four symptoms
- Moderate Depression -Four to Six symptoms
- Severe Depression -Seven and above with or without psychotic symptoms

The symptoms should be present for at least a month or more and every symptom should be present for most of every day.

SCALES FOR DEPRESSION

A depression rating scale is a psychiatric measuring instrument having descriptive words and phrases that indicate the severity of depression symptoms for a time period[24].

1) Hamilton Depression Rating Scale

In 1960, Max Hamilton designed a rating scale which includes 17 questions. Each question is rated from 0 to 4 according to the severity. It is one of the most commonly used scales to assess the effects of drug therapy[25].

2) Montgomery - Asberg Depression Rating Scale

It is a questionnaire containing 10 items to be completed by researchers to assess the effects of drug therapy[26].

3) Raskin Depression Rating Scale

It rates the severity of the patients' symptoms in three areas: verbal reports, behavior, and secondary symptoms of depression.

PHARMACOTHERAPY

Pharmacotherapy is the most effective intervention in the management of MDD, helps to achieve remission and also prevents relapse. Among the drugs available, SSRIs and SNRIs are the most commonly used drugs now. They belong to second generation antidepressant drugs. TCAs and MAOIs are referred to as first generation drugs but not

preferred as first line drugs because of lack of selective targeting leading to an array of adverse effects including dreadful complications like cardiac conduction abnormalities.

Even though SSRIs and SNRIs are used commonly which have a high efficacy and safety in overdosage and better tolerability, with a good patient compliance, they too have adverse effects.

Any antidepressant needs to be administered in appropriate doses, at least for a period of 4-6 weeks to achieve remission. It is still more extended in case of older individuals up to 12 weeks. Antidepressants must be continued even after achieving remission for a period of 6 months if it happens to be a single episode and for a longer duration of up to 2 years if the patient has suffered from multiple episodes.

Withdrawal from the antidepressants must be done gradually so as to prevent withdrawal symptoms like headache, giddiness, gastrointestinal symptoms, sweating and sleeplessness. It is more common with short acting drugs like paroxetine and venlafaxine. Except for fluoxetine with a very long half life, abrupt cessation should not be done for other drugs[27].

Extrapyramidal syndrome in case of withdrawal from SSRIs and psychomotor agitation with MAOIs can occur. With drug therapy alone, non compliance is approximately 40%, if there is no symptomatic improvement within a month. Cognitive Behavioral Therapy (CBT) along with antidepressants helps in such patients. It is estimated that antidepressants lead to remission in a good compliant patient, adequately

managed over 6-8 weeks with correct dosage is only 60-70%. None of the antidepressants prove to be an ideal one with a high efficacy, less adverse effect profile and minimal drug interactions. As all classes of antidepressants have almost similar efficacy, the choice of the agent for a particular patient is based on the adverse effect profile of the antidepressant[28].

CLASSIFICATION OF ANTIDEPRESSANTS

Drugs are classified based on their chemical structure and their target actions as Reuptake inhibitors, Enzyme inhibitors and Receptor blockers.

I. Tricyclic antidepressants (TCAs)

(i) Nonselective reuptake inhibiton of both NA & 5HT

- Imipramine
- Amitryptiline
- Trimipramine
- Clomipramine
- Doxepin
- Dothiepin

(ii) Relatively selective NA reuptake inhibition

- Amoxapine
- Desipramine

- Nortriptiline
- Maprotiline
- Nordoxepin
- Reboxetine
- Lofepramine
- Norclomipramine

II. Selective Serotonin Reuptake Inhibitors (SSRIs)

- Fluoxetine
- Fluvoxamine
- Paroxetine
- Sertraline
- Citalopram
- Escitalolopram

III. Serotonin and Noradrenaline reuptake inhibitors

- Venlafaxine
- Desvenlafaxine
- Milnacipram
- Levomilnacipram
- Duloxetine

IV. MAO Inhibitors

(i) Reversible inhibitors

- Moclobemide
- Clorgyline
- Brofarmine
- Pirilindole
- Toloxatone
- Belfoxatone

(ii) Irreversible inhibitors

- Tranylcypromine
- Isocarboxazid
- Phenelzine

V. Atypical antidepressants

- Trazodone
- Mianserin
- Mirtazapine
- Bupropion
- Tianeptine
- Amineptine

- Nafazodone
- Atomoxetine

II. NON- PHARMACOLOGICAL TREATMENTS

1. PSYCHOTHERAPIES

- Cognitive Behavior Therapy (CBT)

  Cognitive behavior therapy is recommended as a first line therapy in managing mild to moderate depression. In severe depression, CBT along with drug therapy is effective.

- Inter Personal Therapy (IPT)

  Inter personal therapy helps to improve the social functioning of the patient.

2. BRAIN STIMULATION TECHNIQUES

- Electro- Convulsive Therapy (ECT)

  Electro- convulsive therapy helps to have a quick relief from symptoms of depression but for a short span only. So antidepressants must be started to avoid relapse. ECT is preferred for patients with suicidal tendency

- Vagus Nerve Stimulation

  An electrical device is implanted below the clavicle, in the subcutaneous plane which stimulates the cerebral cortex along the left vagus. FDA approved this technique for resistant cases of depression.

- Transcranial Magnetic Stimulation (TMS)

  Electrical stimulus is applied across the scalp in a noninvasive manner which creates an electrical field in the cerebral cortex.

- Light therapy

  Light therapy relieves fatigue and irritability associated with depression.

MECHANISM OF ACTION OF ANTIDEPRESSANTS

Drugs which increase the serotonin and noradrenaline at the synapse either by preventing the reuptake by SERT/NET (SSRIs, SNRIs, TCAs) or by preventing degradation of monoamines (MAOIs) or by directly blocking the α_2 or 5-HT_2 receptors, ultimately facilitate the serotonergic and adrenergic neurotransmission and thus help in depression. Antidepressants cause therapeutic effects only after 2 weeks and maximum effect at 8-12 weeks. The slow onset of therapeutic effects is attributed to the chronic effects of antidepressants rather than acute effects.

Neurotransmitter level in the synapse increases immediately after drug intake, but there will not be an anticipated level of increase in neurotransmitters, because of stimulation of autoreceptors (α_2 or 5-HT_2) in the presynaptic neurons[29]

ACUTE EFFECTS OF ANTIDEPRESSANTS

Antidepressants

↓

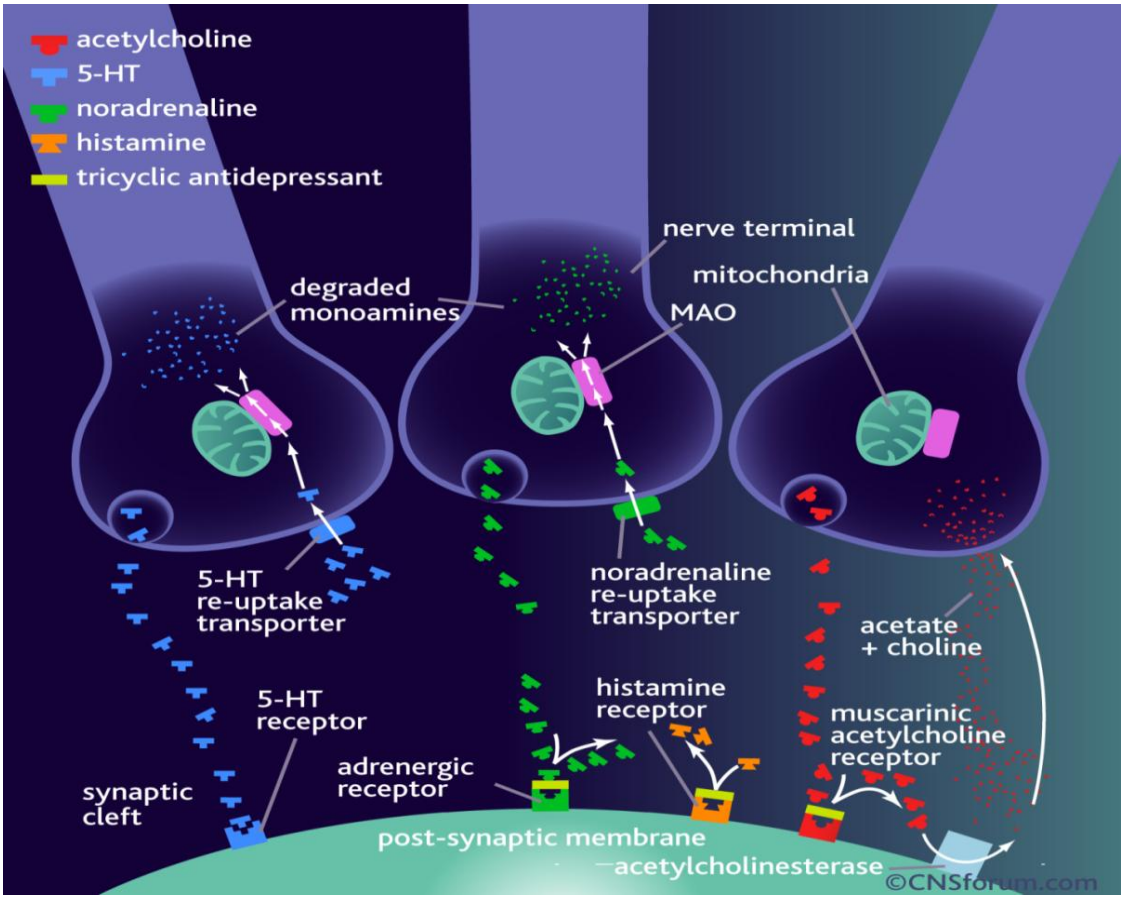
Increased neurotransmitters NA/5-HT or both

↓

Stimulation of autoreceptors (α_2 and 5-HT_2) in presynaptic neuron

↓

Decreased NA and 5-HT synthesis



CHRONIC ADMINISTRATION OF ANTIDEPRESSANTS

Down regulation of autoreceptors

↓

Increased sensitivity of postsynaptic receptors

↓

Enhanced neurotransmission

↓

Alteration of intracellular signaling pathways

↓

Increased cyclic GMP signaling

↓

Increased phosphorylation of CREB

↓

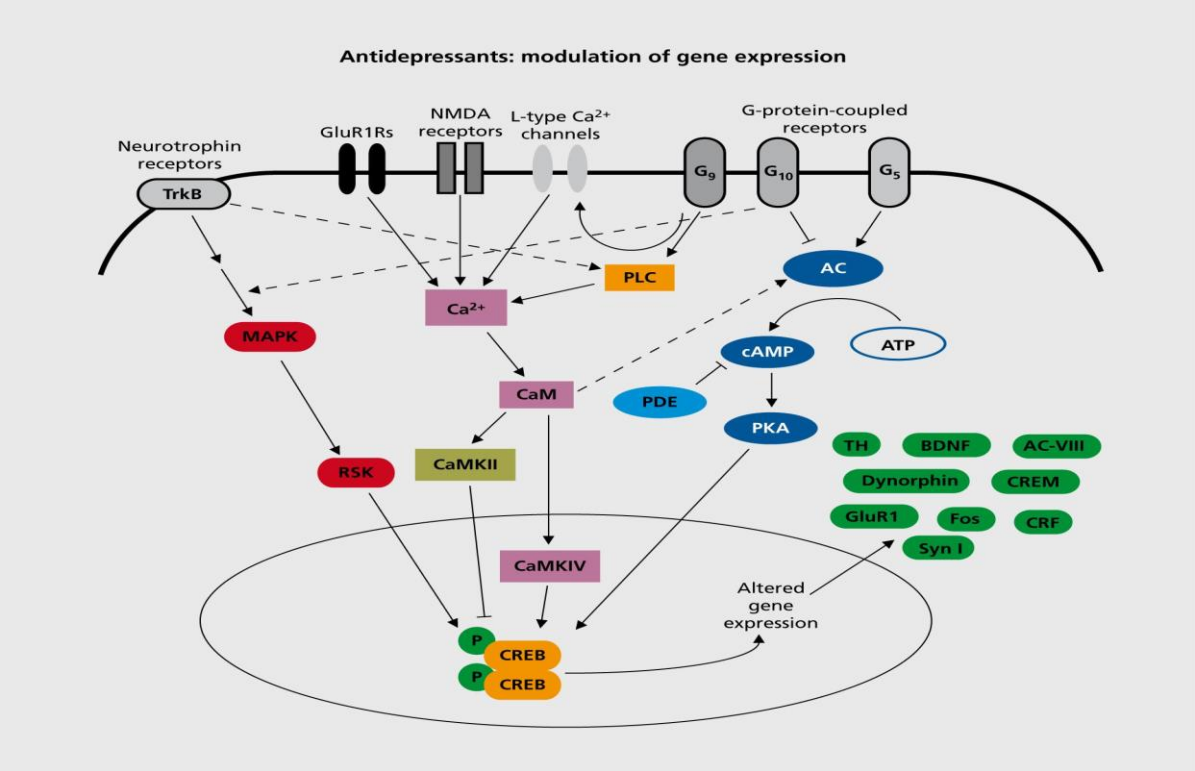
Induction of neurotrophic factor like BDNF

↓

Activation of progenitor cells

↓

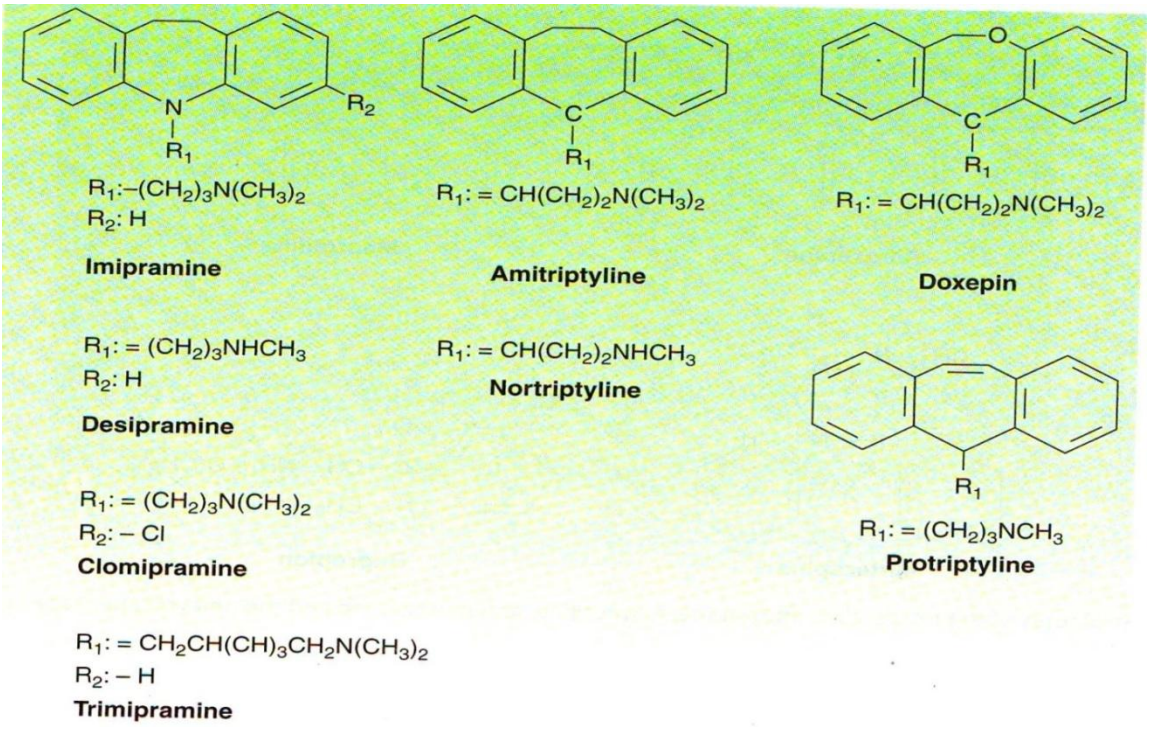
Neurogenesis at hippocampus and other areas



TRICYCLIC ANTIDEPRESSANTS

CHEMISTRY AND STRUCTURAL ACTIVITY RELATIONSHIP

Tricyclic antidepressants, as the name describes they are three ringed organic chemical structures which were initially used as antipsychotics. Later on serendipitous discovery of its antidepressive action was observed. Between 1950 and 1960, it was marketed as antidepressant. The initial development of TCA resulted from the psychopharmacological characterization of a series of structurally related analogues that had been developed as antihistamines, sedatives, analgesics and antiparkinson's drugs. Search for the compounds related to imipramine yielded many analogues[30].



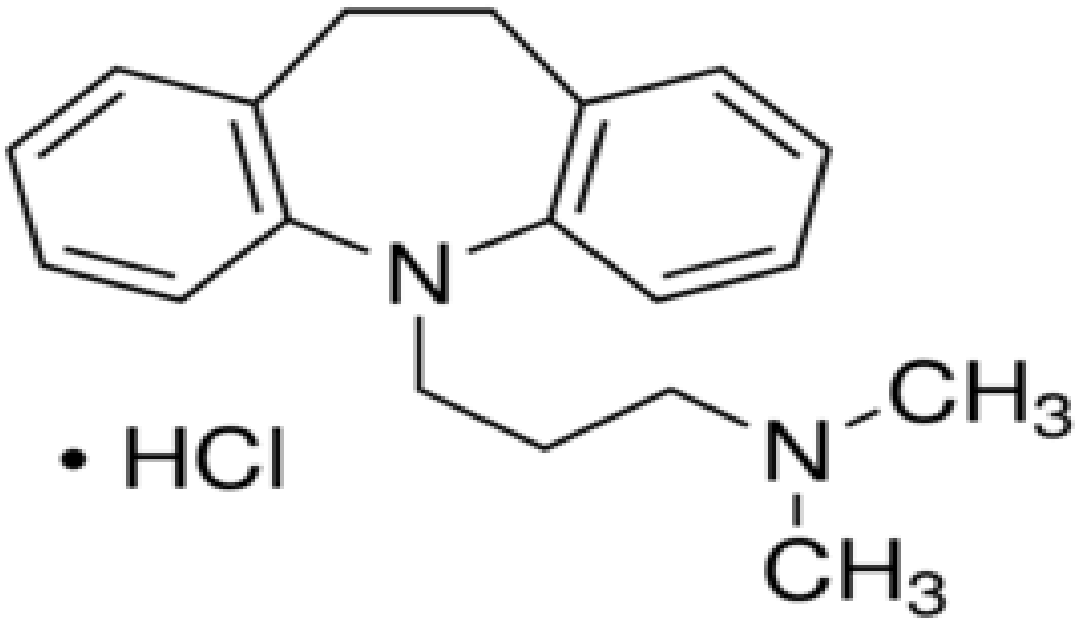
The tertiary amine tricyclics such as amitriptyline, imipramine has two methyl groups at the end of the side chain. These can be methylated to secondary amines such as desipramine and nortryptiline. The tetracyclic compounds maprotiline and amoxapine have four ringed central structure. Chemical modification of TCA structure led to earliest SSRI, Zimelidine which was withdrawn due to serious adverse effects.

Imipramine is a tricyclic antidepressant, with tertiary amine side chain, inhibits both norepinephrine and serotonin reuptake, thereby the availability of the serotonin and the noradrenaline is increased,producing antidepressant action. They also block histamine, adrenergic and muscarinic receptors and in high doses they may block sodium channels. The additional receptor blockade by imipramine leads to more side effects. The potency of blocking serotonin and noradrenaline by the TCA varies among

the individual drugs in the group. Desipramine, maprotiline block NET more selectively[31].

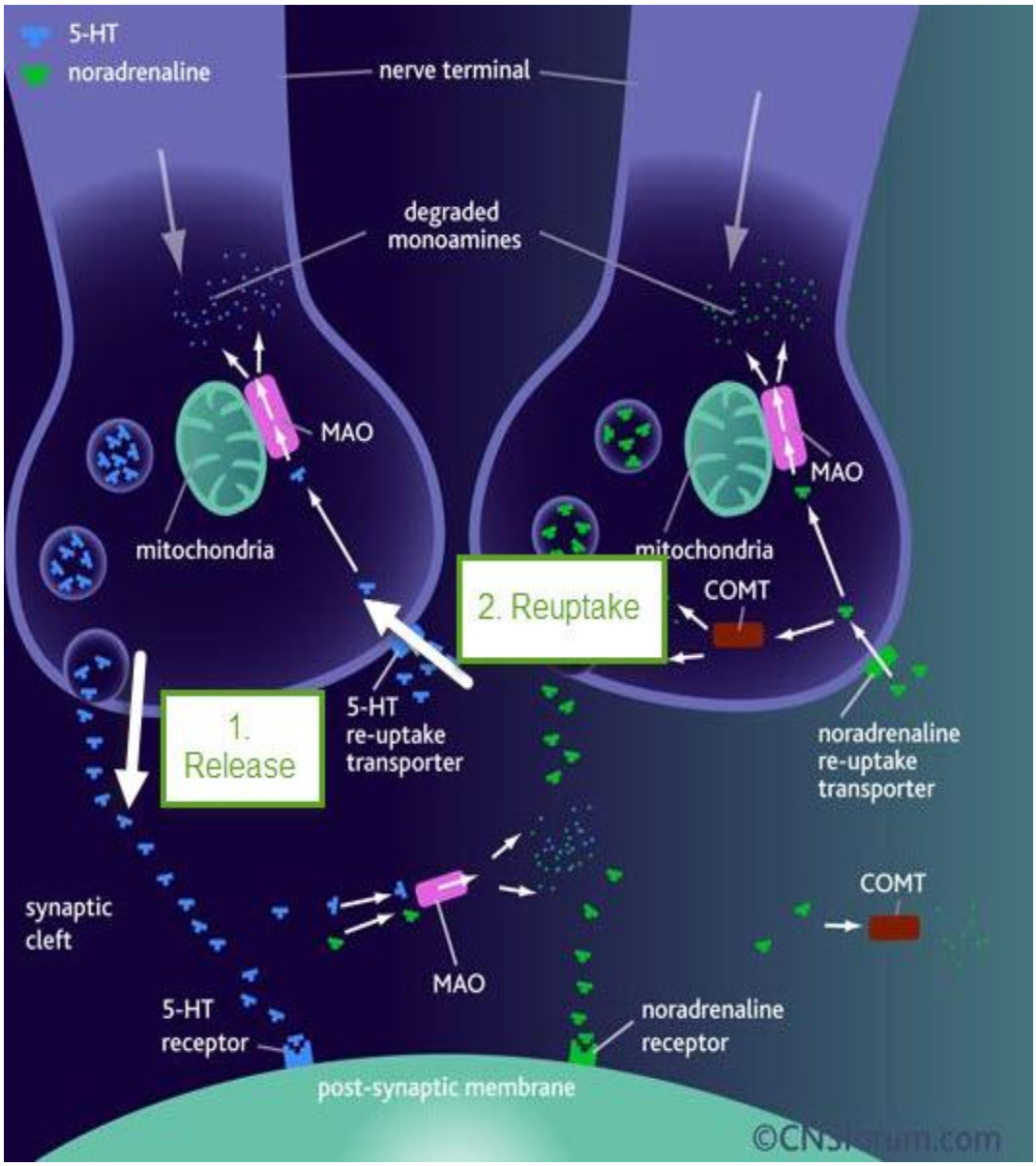
In this present study Imipramine is taken as a standard drug for comparing the antidepressant effect of lemon grass.

IMIPRAMINE[32]



3-(10,11-Dihydro-5*H*-dibenz[*b,f*]azepin-5-yl)propyldimethylamine.Imipramine Hydrochloride is a odourless, white to off-white, crystalline powder. It can be dissolved freely in alcohol and water. It is soluble in acetone also but insoluble in benzene or ether. This drug must be stored in air tight container.

MECHANISM OF ACTION OF IMIPRAMINE

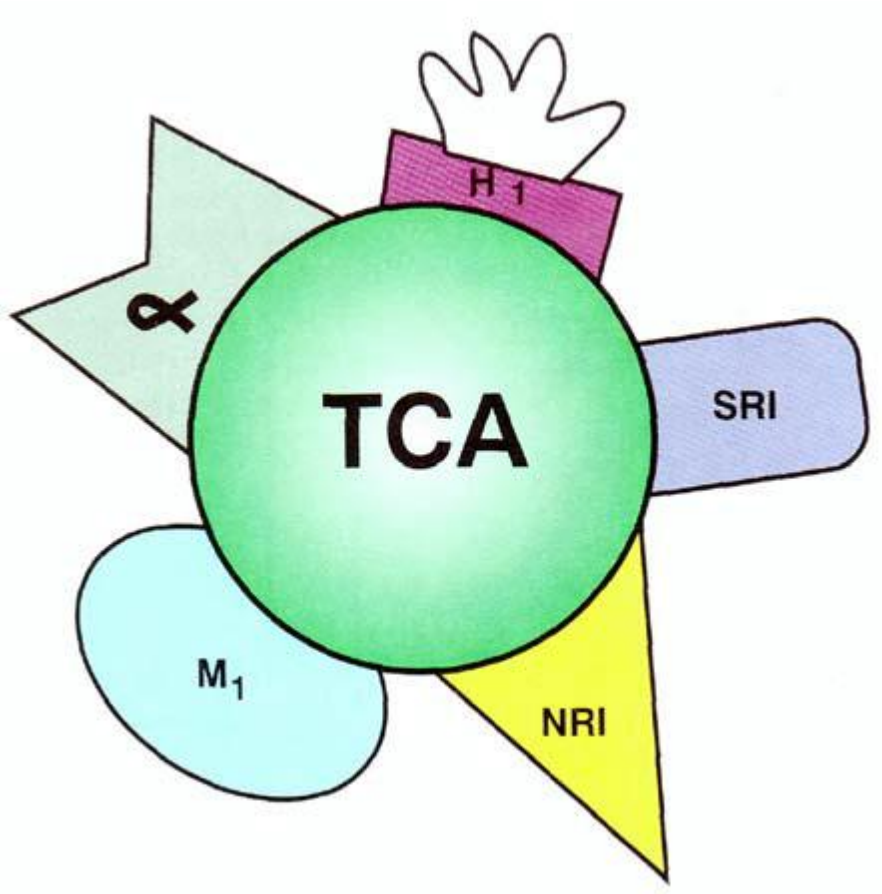


PHARMACOKINETICS

Imipramine is available as tablet and administered orally. The drug gets absorbed quickly. It undergoes high first pass metabolism in the liver, getting converted to

desipramine, an active metabolite, by demethylation. Desipramine is further metabolized by hydroxylation and N- oxidation and inactive metabolites are excreted mainly by kidneys and a small percent in faeces through bile. It is distributed widely throughout the body and bound to plasma proteins and tissue proteins extensively. It has a half life of 9 – 28 hours or more in over dosage.

IMIPRAMINE ADVERSE EFFECTS



Due to its nonselective action, imipramine causes dry mouth, constipation, paralytic ileus, urinary retention, blurring of vision, increased intraocular pressure, hyperthermia. Imipramine should be used cautiously in elderly patients with benign

prostatic hypertrophy, glaucoma, and in patients with chronic constipation. This can be prevented by starting in small doses, but the clinical response may be delayed.

It can cause adverse effects in central nervous system which include drowsiness, headache, peripheral neuropathy, tremor, ataxia, seizures, tinnitus, confusion, hallucination and delirium, especially with elderly individuals. Mania and behavioral disturbance can occur in children.

Gastrointestinal effects are metallic taste, nausea, vomiting, gastric irritation. Hypersensitivity reactions like photosensitization, angioedema, utricaria, rarely bone marrow depression, leucopenia, agranulocytosis and eosinophilia can occur with the intake of imipramine. Endocrine side effects include gynaecomastia, galactorrhoea, testicular enlargement, sexual dysfunction, SIADH and changes in blood sugar.

Most dreadful complications include cardiovascular adverse effects like tachycardia, postural hypotension, cardiomyopathy, QT prolongation progressing to torsades de pointes, increased risk of MI which occur even in therapeutic doses itself. Sudden cardiac death can occur in patients with preexisting cardiac disease, common in overdosage and in children.

PRECAUTIONS

1. Cautious use of imipramine in urinary retention, prostatic hypertrophy, chronic constipation, untreated angle closure glaucoma and pheochromocytoma.

2. It is epileptogenic and its usage in patient with history of seizures should be cautious.
3. Heart block, arrhythmias may be precipitated in patients having decompensated cardiac reserve. Arrhythmia may be precipitated in hyperthyroid subjects.
4. Blood sugar alteration may be produced in patients with diabetes; so blood sugar monitoring has to be done regularly.
5. Imipramine is metabolized in the liver, so patients with lower hepatic function need dose reduction.
6. Imipramine also possesses the risk of suicidal tendency like all other antidepressants.
7. Drowsiness may be there at the start of treatment and later tolerance may develop.
8. It is not recommended for treatment of depression in children.
9. It can be used for nocturnal enuresis in children but limited to short courses.
10. It is contraindicated in pregnancy. It is secreted in breast milk, so avoided in mothers during breast feeding.

WITHDRAWAL

Sudden stopping after 8 weeks or more may precipitate withdrawal symptoms. It presents with gastrointestinal disturbances, somatic symptoms, malaise, chills, headache anxiety, agitation, sleep disturbances, insomnia, vivid dreams, akathesia, hypomania or

mania. Many of the symptoms associated with stopping of TCA are due to cholinergic rebound and can be minimized by gradual reduction of dose.

It is recommended that any antidepressants, including TCA that has been given regularly for 8 weeks or more should be stopped gradually over a period of 4 weeks. After response to therapy, the maintenance phase of treatment should be initiated for the patient. The maintenance should be given for 4 to 6 months to avoid relapse on stopping therapy. Patient with history of recurrent depression should continue to receive maintenance therapy for a period of five years and possibly lifelong if there are more possibilities of developing depression in the future.

USES OF IMIPRAMINE

AS ANTIDEPRESSANT

Imipramine is started with a dose of 75mg daily initially and gradually increased upto 150 mg daily. It is given as a single dose or in divided doses. In severe depression doses upto 200- 300 mg daily is used. Additional doses are given in late afternoon or evening. In adolescents and elderly it should be started as low as 25mg as these age group patients tolerate imipramine poorly. Imipramine, as the hydrochloride, is also available in injection formulation which is administered by intramuscular route.

FOR NOCTURNAL ENURESIS

The use of imipramine hydrochloride is indicated for nocturnal enuresis after ruling out organic causes if any. In children 6-7 years of age, the dose needed is 25 mg

daily; for 8-11 years of age, 25-50 mg daily; for age more than 11 years, 50 - 75 mg daily. The drug needs to be administered at night.

OTHER USES

- Anxiety disorder
- Bulimia nervosa
- Interstitial cystitis
- Nocturnal enuresis
- Narcoleptic syndrome
- Painful neuralgias
- Attention deficit hyperactivity disorder
- Chronic pain

OTHER ANTIDEPRESSANTS

SELECTIVE SEROTONIN REUPTAKE INHIBITORS

They are more selective inhibitors of serotonin reuptake[33]. SSRIs increase the serotonin levels in the serotonergic neurons of the somatodendritic areas while chronic administration leads to desensitization of $5HT_{1A}$ auto receptors. The antidepressant activity of SSRI is due to the increased serotonergic neurotransmission between midbrain raphe nucleus to prefrontal cortex. Citalopram and fluoxetine are racemic mixtures while sertraline and paroxetine are separate enantiomers. SSRIs are the first line drugs for the treatment of depression as they lack many side effects of older antidepressants like MAO-A inhibitiors and TCAs. But still they do have some undesirable effects. The side

effects include anxiety, insomnia, gastrointestinal disturbances, sexual dysfunction, agitation due its effect on various 5- HT receptors like 5- HT_2, 5- HT_3.

SELECTIVE NOREPINEPHRINE REUPTAKE INHIBITORS

They produce dose dependent blockade of various transporters. At low dose they block serotonin transporters and at medium to high dose, they cause additional norepinephrine blockage and very high dose they cause dopamine reuptake blockade. Eg. Venlafaxine. Anticholinergic, sedative side effects are less with SNRIs. They have lesser side effects than TCAs. Duloxetine and Milnacipran are better tolerated and there is no cardiovascular toxicity[34].

SEROTONIN-2 RECEPTOR ANTAGONISM WITH SEROTONIN REUPTAKE BLOCKADE

Trazodone blocks 5-HT_2 receptors and blocks the reuptake of serotonin by inhibiting serotonin transporters. It also has some alpha and H_1 blocking action[35]. Some of the side effects of SSRIs, such as the short-term increase in anxiety or insomnia, and sexual dysfunction will not occur with this group of drugs.

Noradrenergic and Specific Serotonergic Antidepressant (NaSSA) mirtazapine, blocks alpha $_2$ auto receptors on noradrenaline neurons and hetero receptors on serotonin neurons, thereby enhancing both noradrenaline and serotonin release. Sexual side effect is less but it has weight gain and sedation as adverse effects. Lipid profile and blood count has to be monitored. Agranulocytosis and neutropenia may occur.

SPECIFIC NORADRENALINE REUPTAKE INHIBITOR

Reboxetine is the truly selective noradrenaline reuptake inhibitor[36] with only very little affinity for other neuroreceptors including serotonin, dopamine, histamine, muscarinic and alpha adrenergic sites. Muscarinic side effects, weight gain and sedation are less.

SEROTONIN REUPTAKE ENHANCER

Tianeptine increases the presynaptic uptake of serotonin after single as well as repeated administration, but this action is not linked to any effects on the 5-HT postsynaptic systems. Tianeptine has no affinity for alpha-1 adrenergic, H1 histaminic and muscarinic receptors. Long term treatment with tianeptine prevents chronic stress induced changes in amygdala and hippocampus[37].

FUTURE ANTIDEPRESSANTS[38]

1. Drugs affecting monoamine transmission

Drugs with one or more of the following properties like β3-adrenoreceptor agonism, D_2 dopamine receptor agonism or antagonism, 5-HT_{1A} receptor agonism or partial agonism and 5-HT_{2A} receptor antagonism as well as dopamine, noradrenaline and 5-HT uptake inhibition.

2. Drugs acting on ion channels Agonists, partial agonists and antagonists of nicotinic receptors have antidepressant properties. Agonists induce receptor

desensitization and partial agonists inhibit endogenous acetylcholine, ultimately resulting in reduced receptor activation.

3. Drugs acting at the NMDA receptor

Single dose of ketamine has been reported to alleviate depression quickly and the effect lasts for many days.

4. Drugs acting at the AMPA receptor

AMPA kines, drugs that potentiate responses at the AMPA receptor show efficacy in animal models.

5. Other novel drug targets

P2X receptors

5. Novel receptor targets-

- GRII cortisol receptor antagonists
- Melanocyte inhibiting factor (MIF-1) analogues
- Melatonin M_1/M_2 receptor agonists
- NK_1 and NK_2 receptor antagonists

Time has to answer whether these novel targets will yield an effective antidepressant response which will increase the efficacy in treatment of depression with negligible side effects.

CHOOSING OF ANTIDEPRESSANTS

Most of the depressive episodes are self-limiting cyclical disorder and pharmacotherapy aims at decreasing the cycle duration, widening the intercycle period and stabilizing the mood entirely.

The success depends on the right diagnosis, right drug, right dosage and right duration of treatment all these four factors lead to the right outcome. About 70 to 85 % of depressed patients can lead a normal life.

The response to antidepressants is reported to be as high as 60 – 70 %, some may show improvement in the first week, but may not fully respond before 4 – 6 weeks. The therapeutic lag for the initial period of the treatment is mainly due to presynaptic α_2 and $5HT_1$autoreceptor activation.

As a result there is accumulation of noradrenaline and serotonin in the synaptic cleft which results in decreased firing of locus coeruleus and raphe neurons. After a long term administration of antidepressants there is desensitization of the presynaptic $alpha_2$ receptors, $5HT_{1A}$ and $5HT_{1D}$autoreceptors and also they induce other adaptive changes in number and sensitivity of pre and post synaptic noradrenaline and serotonin receptors. The rate of response is equal for all the antidepressants the choice is based on side effects profile, patient's psychiatric history, medical history and family history. The antidepressants are usually started in low doses and then slowly increased to the desired level[39].

COURSE OF ANTIDEPRESSIVE MEDICATION

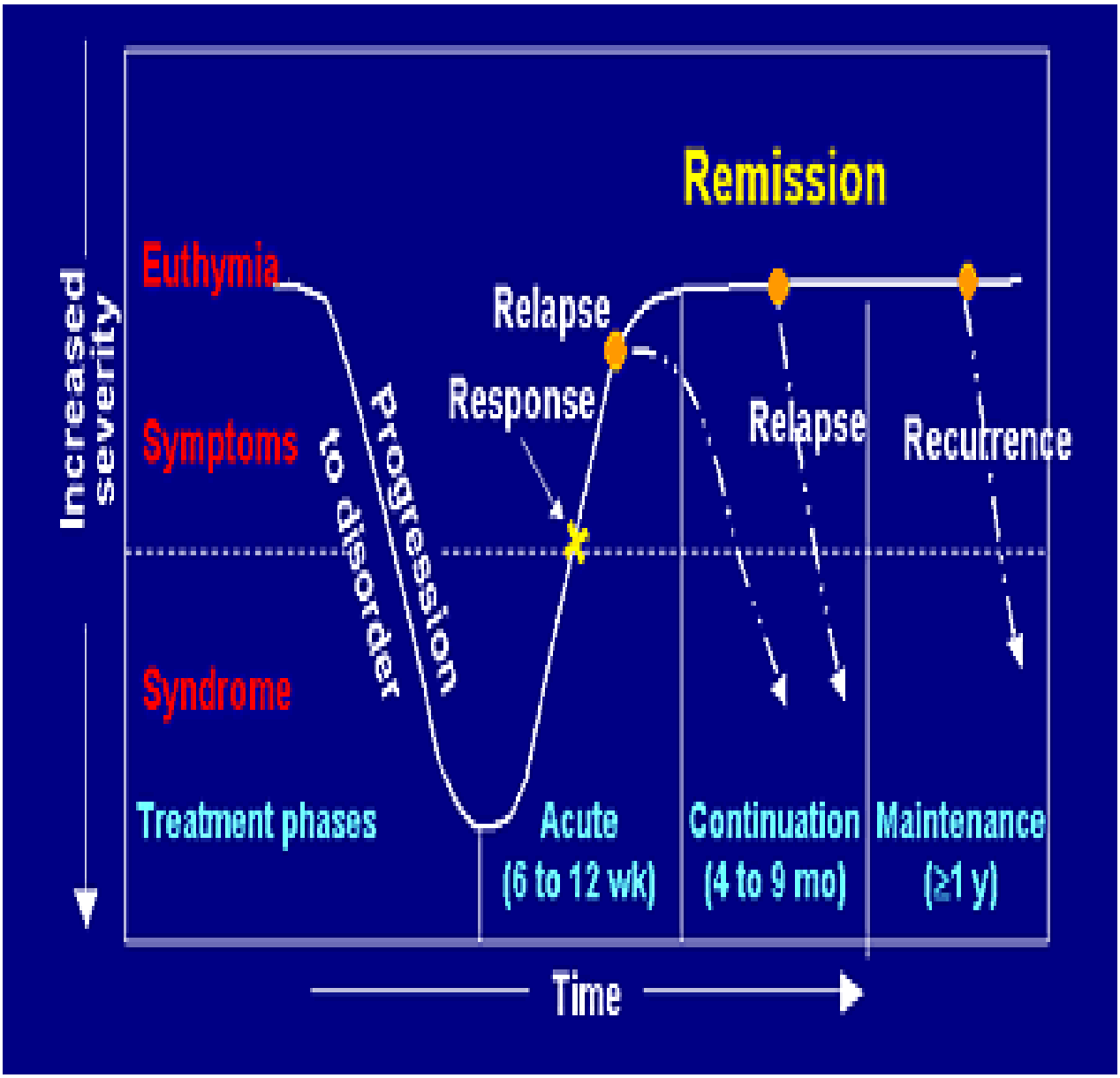
About 50 % of patients are diagnosed when they develop significant depression. Therefore early identification and treatment of symptoms may prevent the full depressive episode. The first depressive episode occurs before the age of 40 in 50 % of the patients. If untreated the depressive episodes lasts for six to 13 months, whereas for most patients on treatment, episodes lasts for about 3 months. Shorter courses of antidepressants for 1-6 months fail to show benefit rather a long course of treatment benefits the patient[40]. Major depressive disorder is not a benign disorder as it tends to be chronic and also relapse. Mild episodes of depression with absence of psychotic symptoms has good prognosis while depression associated with alcohol abuse, substance abuse, anxiety disorders have poor prognosis. Treatment outcome after long term use of antidepressants can be categorized as 5 R's i.e Response Remission, Relapse, Recovery and Recurrence[41]

REMISSION - a period in which the individual is asymptomatic, remission extends From 2 to 9 months.

RECOVERY - a remission that last for more than 9 months without relapse after appropriate treatment.

RELAPSE - a return of symptom, meeting criteria for full syndrome of major depressive disorder that occurs during a period of remission but before recovery.

RECURRENCE - a manifestation of new episode of major depressive disorder that occur during recovery.



The first positive step towards recovery is a significant response to treatment and then the phase of continuation of pharmacotherapy begins. The goals are to prevent relapse and remission. Maintenance pharmacotherapy is recommended to follow continuation therapy for most of patients with a history of recurrent episodes of depression.

COMPLEMENTARY AND ALTERNATIVE THERAPY IN DEPRESSION

NATURAL PRODUCTS IN DEPRESSION

OMEGA-3 FATTY ACIDS

Omega-3 fatty acids influence serotonin functioning and improve functions of hippocampus. Thus they help in depression and also improve overall mental functioning[42].

S-ADENOSYL METHIONINE (SAM-E)

SAM-E is present in our body, taking part in various cellular metabolism. It helps to improve the mood significantly. Meta analysis confirms its superiority over a placebo for depressive syndromes. Many study results suggest, SAM-E is effective in depression. It has good tolerability, quick onset of action with minimal side effects[43]. Hence, it will be a better option for patients who cannot tolerate drugs.

FOLIC ACID / FOLATE (VITAMIN B9)

Folate has a role as an adjunct, along with antidepressants. There is also evidence that low folate diet contributes to depression[44].

MAGNESIUM

Magnesium deficiency, leads to opening of calcium channels which result in neuronal damage and ultimately ending up with dysfunction of neurons. There is also

proof with low magnesium levels in CSF among persons who commit suicide and low brain magnesium in patients with resistant depression.

Other natural products like 5-Hydroxytryptophan, inositol, creatine are also having antidepressant property.

PLANT DERIVED DRUGS

Medicinal substances of plant origin are known to mankind for several centuries. Majority of medicines were formulated from herbs until the previous century. Current day pharmaceutical industry still depends on plant sources for the synthesis of many lead compounds as a vital part in new drug development. Plant derived drugs are called phytomedicines.

According to World Health Organization (WHO), more than 10,000 plant species are in use worldwide. 20 -50% of patients consume herbal medicines. They are taken either as a whole or its components, extracts, tablets, capsules, syrups.

India is known for its medicinal plants. Thousands of medicinal plants have been identified. These include herbal, medicinal and aromatic plants. India has indigenous medical systems like ayurveda, siddha and unani medicine where herbal medicines are mainly used.

Traditional medicine is being integrated worldwide along with the primary health care because of its easy accessibility to all and acceptability and cultural significance[45].

Medicines used in alternative therapies are not approved drugs with proven safety, efficacy and quality. These medicines are having traditional values and are used empirically.

In the recent times, there is an increase in consumption of herbal medicines because of their traditional values. Mostly female patients with chronic medical problems seem to choose herbal medicines. Depression, sleep disorders, obesity, benign prostatic hypertrophy, chronic pain, erectile dysfunction, common cold, cough, cancer, HIV, arthritis are some of the diseases where herbal medicines are commonly used.

Herbal medicines are widely used because of easy availability and the belief that 'naturalness' of the phytomedicines always protects rather than cause adverse effects. Due to lack of regulation regarding their production, they are more prone to cause adverse effects due to contaminants and adulterants. Since no medicine is free from side effects, herbal medicines must also be subjected to proper clinical trial.

Stress changes the normal physiological activities in the brain resulting in pathological conditions like depression. It is attributed that decreased neurotransmitters in the brain, nootropic as well as neuromodulatory factors, and increased generation of free radicals lead to oxidative damage. Phytochemical extracts obtained from natural products and botanicals serve as stress adaptogens and protect against neuronal damage[46]. Various studies have proved that phytomedicines help in diseases of central nervous system like epilepsy, Alzheimer's disease, especially depression.

REGULATIONS[47]

In 1962, FDA delineated drugs from botanicals. Botanicals are categorized as food supplements. Dietary Supplement Health and Education Act (DSHEA; 1994) further regulated the herbal medicines.

FDA has now imposed new regulations regarding the production and labeling specifications for herbal medicines. The research with herbal medications is under the control of National Center for Complementary and Alternative Medicine (NCCAM) overseas. American Herbal Products Association (AHPA) is the regulatory body established by the botanical industry. In the European Union, regulations are more stringent. Similar to conventional drugs, herbal products must also get license which is issued by the Committee on Herbal Medicinal Products (HMPC).

Some of the herbs possess antidepressant property. Few common herbs used for depression are as follows.

ST. JOHN'S WORT

Herbal remedy like St. John's wort is often used for depression. St John's wort, a plant based natural medicine, has become a preferred remedy to alleviate depression in people experiencing mild to moderate depression. It is derived from the herb *Hypericum perforatum*. It is mentioned commonly as goat weed, God's wonder plant, witches' herb. Flowering parts of this plant are used for the preparation of medicine. The active principle responsible for its action is found to be hyperforin & hypericin. It acts by

inhibition of MAO enzyme and monoamine reuptake. It facilitates GABAergic transmission and also decreases cortisol by inhibiting ACTH release, thereby helps in depression.

Clinical trials and meta analysis have proved that St John's wort has similar effects like TCAs, but we cannot conclude about its efficacy with those trials alone due to lack of standardization of the of herbal preparation and also large multicentric trials fail to show any evidence regarding its efficacy over placebo in patients with depression.

It is worthwhile to note that this drug has also adverse effects like gastrointestinal disturbances, dry mouth, mental confusion, sleepiness, increased somnolence. This drug has drug interactions because of its enzyme induction property. CYP1A2 and CYP3A4 induction occurs leading to decreased drug levels of warfarin, antipsychotics, oral contraceptives, some anticonvulsants and HIV protease reverse transcriptase inhibitors[48]. In patients with bipolar disorder it precipitates mania. When the patient takes this medicine along with antidepressants like TCAs, SSRIs, patient will develop serotonin syndrome due to high levels of serotonin.

In spite of its controversies regarding efficacy and potential drug interaction this drug is still preferred by some patients with mild to moderate depression. It is available over the counter, cheap, and easy to take the medicine. It is claimed that it is also helpful in anxiety and sleep disorders. It is available as capsules, tablets, extracts and tinctures.

KAVA

Another herb Kava, called commonly as toxic pepper, kava or tonga. It's botanical name is *Piper methysticum.* The active principle is found in its root which causes GABA receptor modulation function and it helps in anxiety, depression and sleep disorders. It is potentially toxic causing severe hepatotoxicity[49]. It inhibits CYP-450 enzymes and increases the sedation by benzodiazepines (BZDs). It also helps in schizophrenia by decreasing the effects of dopamine. It is available as capsules, tablets, extracts and tinctures.

GLYCYRRHIZA GLABRA

Commonly it is known as liquorice. It shows antidepressant activity which is due to increase in dopamine and noradrenaline mediated neurotransmission in brain but not by serotonin. It has monoamine oxidase inhibiting property too[50].

OCIMUM SANCTUM

Antidepressant activity of Ocimum sanctum roots is due to its antioxidant effect. Hence it can be used as an adjuvant in the treatment of depression[51].

GINKGO BILOBA

Lipophilic extract of the leaves has antidepressant effect and the active principle is also identified.

ROSMARINUS OFFICINALIS L.

Hydro alcoholic extract of leaves and stem has antidepressant effect by interfering with the monoaminergic neurotransmission.

CURCUMA LONG:

The aqueous extract of the roots has antidepressant activity which is more effective than fluoxetine and it is partly mediated through MAO A inhibition.

ASPARAGUS RACEMOSUS LINN

Methanolic extract from roots of asparagus augments the serotonergic and the noradrenergic neuronal activity and also possess antioxidant activity. Thus helps in depression.

EMBLICA OFFICINALIS

The antidepressant activity was comparable to that of standard drug imipramine.

LEMON GRASS

Cymbogon citratus is used all over the world for various purposes. It is used in several food preparations like salad, meat. The plant has a citrus flavor which is used to

add flavor in preparations like teas, soups, curries[52]. *C. citratus* is also used in folk medicine and in aroma therapy.

The genus *Cymbogon* comprises of about 55 species. It grows well in tropical areas of Asia, Africa, South and Central Americas. It is a perennial grass which has a stiff stem and short rhizomatous root.

ETHNOPHARMACOLOGY

Lemon grass belongs to the family: Poaceae, genus: *Cymbogon*, species: *citratus*. It has various common names: in English it is called as lemon grass or citronella; in Hindi it is called as sera or verveine; in Tamil it is called Vilaamichhan.

MORPHOLOGAL DESCRIPTION

The leaves of lemon grass are bluish green in colour, arranged like a flower spike, mostly arising from the ground, and they don't have a stem. The leaves smell like lemon when crushed. *Cymbogon* is a greek word; 'cymbe' means boat and 'pogon' means beard. It is named so because of the flower spike arrangement of its leaves.

TRADITIONAL USAGE OF LEMON GRASS

In South America, this plant is used to prepare tea and used as antipyretic, analgesic, anti-inflammatory, sedative and diuretic. In India, the whole plant is used to keep away snakes. Essential oil prepared from lemon grass helps to relieve gastric problems. Tea prepared using the leafy portion has a sedative action. In USA, extract prepared from the whole plant is used externally for bone injuries and to heal skin wounds. Some of its activities include anti-amoebic, anti-bacterial, anti-diarrheal, anti-filarial, anti-fungal, anti-malarial, anti-mutagenicity, anti-mycobacterial, anti-oxidants, hypoglycemic and neurobehavioral effects[53].

Essential oil is used as vapouriser against cold, flu, as a diuretic, to cool the body with external application, to facilitate digestion and to relieve spasms.

PHYTOCHEMISTRY

The chemical composition of the essential oil obtained from lemon grass includes terpenes, alcohols, esters, ketones and aldehydes. Its main constituents are citral, nerol, gerenol, triterpenoids, flavanoids and phenolic compounds[54]. Aqueous extract of lemon grass has no lipids.

PHARMACOLOGICAL ACTIONS[55]

The essential oil exhibits antifungal, antimalarial, larvicidal, antibacterial, antiamoebic, antinociceptive and ascaricidal effects. Aqueous extract prepared from fresh leaves shows anticancer, antioxidant, hypoglycemic, hypocholesterolemic, antifilarial, antidiarrhoeal and anti-inflammatory effects.

ANTI-INFLAMMATORY PROPERTY OF LEMON GRASS

Polyphenol and citral in the lemon grass is responsible for the anti inflammatory effect. Tannins, flavanoids, phenol present in the aqueous extract inhibit PGE_2, NO and iNOS expression in the LPS induced macrophage cell lines. COX-2 inhibition is due to citral present in essential oil. Citral reduces COX-2 mRNA, and its formation in a dose dependant manner.

ANALGESIC ACTIVITY OF LEMON GRASS

Terpene (especially myrcene) is responsible for its analgesic effect.

ANTI-OXIDANT EFFECT OF LEMONGRASS

Lemongrass decoction consumption and infusion has antioxidant effect which is proved by 2,2-diphenyl-1-picrylhydrazyl (DPPH) decolouration and inhibition of lipid peroxidation. Flavanoid and tannin extract is more potent than phenolic acid fraction. Aqueous extract inhibits reactive oxygen species production, increases glutathione production and action of superoxide dismutase and thus inhibits lipid peroxidation.

ANXIOLYTIC PROPERTIES OF LEMONGRASS

Lemon grass tea shows anxiolytic effects by acting via GABAergic system.

ANTI-BACTERIAL EFFECT OF LEMON GRASS

α-citral and β-citral present in lemon grass tea inhibit the bacterial growth which is enhanced by myrcene. Active against both gram positive and gram negative organisms.

ANTI-OBESITY AND ANTI-HYPOGLYCEMIC EFFECT OF LEMON GRASS

Flavanoids and alkaloids present in the lemon grass by increasing the insulin secretion and by improving the glucose utilization in the periphery, produces hypoglycemic effect.

ANTI-FUNGAL EFFECT OF LEMON GRASS

Essential oil inhibits filamentous fungi showing a broad spectrum of activity against both pathogenic and nonpathogenic fungi.

ANTICANCER ACTIVITY

In Salmonella mutation test, ethanolic extract of lemongrass does not show any mutagenic effect and it has got cytoprotective effect which is proved by maintaining the membrane integrity of mitochondria in stressed murine alveolar macrophages.

LEMON GRASS IN THE PRESENT STUDY

Aqueous extract of lemon grass is used to study the anti depressant effect in mice. Free radical injury is one of the etiological factors in depression. It is proved that lemon

grass extract shows free radical scavenging and antioxidant effect. In addition it has anti-inflammatory effect too. So lemon grass extract, a folk medicine was administered to mice and its antidepressant property was tested using tail suspension test.

EVALUATION OF NEW ANTIDEPRESSANTS[56]

The observations made after using reserpine and imipramine enlightened us about the understanding of pathophysiology of depression. The first generation drugs like nonselective MAOIs are associated with dangerous food and drug interactions and TCAs are having additional anticholinergic and alpha blocking activity and have adverse effects including fatal arrhythmias and reduce seizure threshold also. Then came SSRIs which are the safe and better tolerated but they too have insomnia and sexual dysfunction. If a person in depression develops sexual dysfunction, this may further depress him. Even though many drugs are available now for the management of depression like SSRIs, SNRIs, TCAs, MAOIs, NaSSA, no drug is efficient to cure the disease completely. Hence the discovery of an ideal antidepressant which is safe and efficacious with lesser adverse effects is a topic of current interest.

Due to lack of animal models mimicking depressive illness in humans, screening methodology using animals for the new drug development is difficult. Screening methods currently in use are based on observations comparing the efficacy of known antidepressants and their effects on multiple

test models. When tests for motor activity are done along with the antidepressant activity, the specificity of drug action as an antidepressant alone can be established.

It is always important to choose the right procedure with acceptable standards and specificity. Discovery of new targets for drug action has modified the existing animal paradigms to identify drug molecules acting on them.

In vivo methods:

1. Water wheel model
2. Learned helplessness test
3. Isolation induced hyperactivity
4. Tail suspension test
5. Reserpine induced hypothermia
6. Amphetamine potentiation
7. Apomorphine antagonism
8. Resident intruder paradigms in rat
9. Muricidal behavior in rats
10. Olfactory bulbectomy

WATER WHEEL MODEL

Principle behind this test is ‘behavioral despair activity’ in animals. Animal is forced to swim in a water tank, without any escape. A rotating

wheel which is present inside the water tank adds on to the despair. In this test, the typical number of rotations of water wheel prior to the onset of behavioral despair is noted. A potential antidepressant will increase the escape behavior that is counted as number of water wheel turns.

FORCED SWIM TEST

In a restricted space, mice or rats are forced to swim, with no chance to escape. After an initial phase of struggling, the animal develops a characteristic behavior of immobility. Duration of immobility gets reduced with antidepressants.

LEARNED HELPLESSNESS TEST

By inducing chronic stress in the animals, with repeated foot shock the animals fail to show ‘escape response’. Antidepressants increase the escape response.

RESERPINE INDUCED HYPOTHERMIA

Principle of this study is any drug which will reverse the effect of reserpine induced hypothermia, can have antidepressant activity.

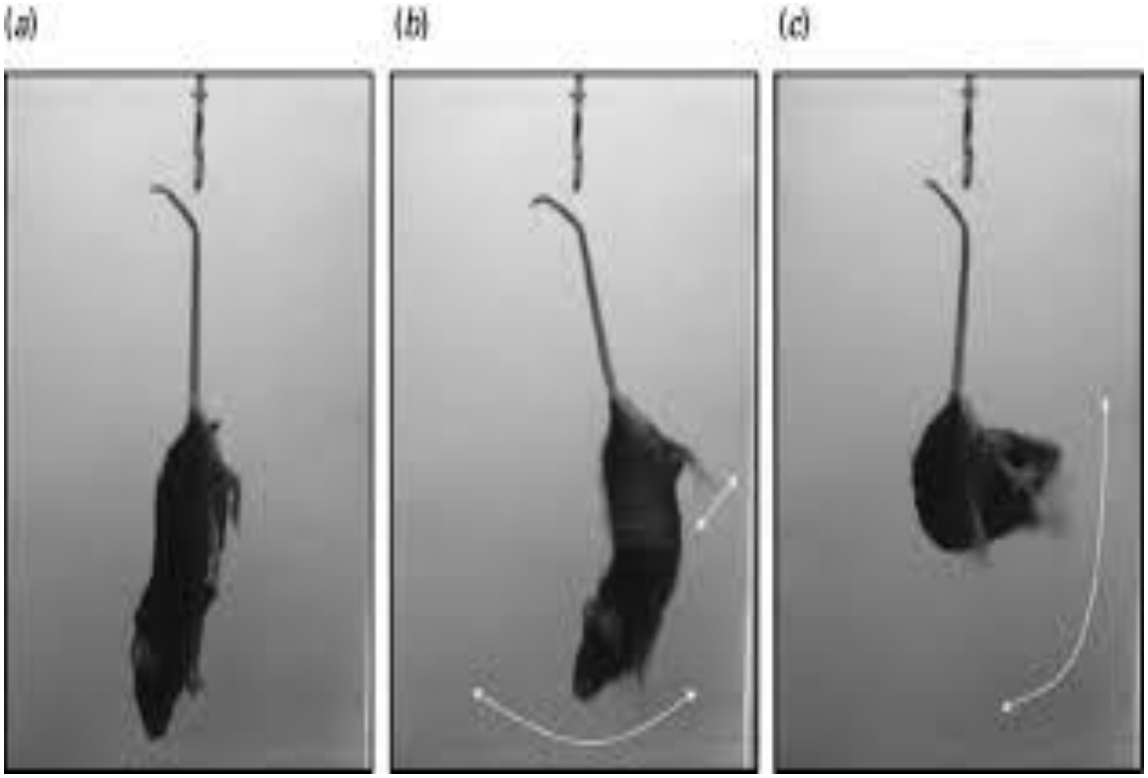
OLFACTORY BULBECTOMY

Sham operated animal exhibit loss of passive avoidance with decreased grooming, suppressed feeding and locomotor stimulation. The behavioral

changes are visible 2 to 5 weeks after surgery and attenuated by drugs with antidepressant property.

TAIL SUSPENSION TEST

Tail suspension test is a behavioral test done in mice to screen the anti depressant effect of drugs and was first done by Steru et al. Mice are better suited for this experiment than rats because of reproducibility. It is a rapid and reliable method used in new drug development to screen their antidepressant effect via high throughput screening.



While suspending mouse upside down on its tail with a tape, in a position which is cumbersome that it cannot escape or hold on to nearby surfaces, 'behavioral despair' is induced. The test is done for a period of 6 minutes. The mice will have escape-oriented behaviors. Due to mood changes, mice will be immobile for a while. The duration of

immobility in the last 4 minutes is noted. The immobility period indicates the degree of depression. This test is not useful for MAO inhibitors. Drugs acting through serotonergic mechanism can be screened by this animal model. This test can also be done using a computer assisted system which calculates the immobility, and activity of the mice in real time.

MATERIALS

AND

METHODS

In the present study, the antidepressant activity of lemon grass was evaluated in Swiss albino mice by tail suspension test. Approval was obtained from Institutional Animal Ethical Committee of Madurai Medical College, Madurai, before commencing the experiment. (R.No:5953/E1/5/2015)

STUDY CENTER

Institute of Pharmacology

Madurai Medical College, Madurai

DURATION OF THE STUDY:

This study was done for a period of 6 months since December 2015.

NUMBER OF ANIMALS USED:

30 adult male albino mice weighing about 25 – 30 grams

DRUGS AND CHEMICALS REQUIRED:

Standard drug - Tab. Imipramine 5 mg/kg (oral)

Test drug - Aqueous extract of *Cymbogon citratus* (Lemon Grass) (oral)

Distilled water

INSTRUMENTS REQUIRED

Adhesive tapes

Table with a bar for suspension

Plastic tubes 4cms x 1.5cms

Digital camera with tripod stand

Stirrers and beakers

Electronic balance

Syringes

Oral feeding tube

Stop watch

LEMON GRASS EXTRACT PREPARATION

The plant was obtained from hilly areas of Courtallam and the species was identified and authenticated by Dr. Stephen, taxonomist, of American College, Madurai and further processing was done by Pharmacognosy department, of Madurai Medical College.

100 g of fresh leaves was extracted with distilled water for 24 hours and filtered with sterile Whatman's number 1 filter paper. The filtrate obtained was concentrated under reduced pressure using rotavapour (Buchi model). The green colour residue obtained was stored in refrigerator at 4°C until required. The extract was weighed and reconstituted daily, with distilled water according to the dosage needed (5mg/kg & 10mg/kg) and administered orally, for a period of 15 days.

IMIPRAMINE HYDROCHLORIDE

Imipramine hydrochloride tablets purchased from Sun Pharma were used as a standard drug in the dose of 10 mg/kg & 20 mg/kg, dissolved with distilled water and administered by the oral route.

PROCEDURE

The animals were selected from Central Animal House, Madurai Medical College, Madurai. The animals were housed in polypropylene cage at room temperature with a 12 hours: 12 hours light / dark cycle. They had free access to food and water ad libitum. They were acclimatized to laboratory conditions for at least 1 week before starting the study.

The study followed the principles of CPCSEA and utmost care was taken while handling the animals and adequate care was provided to them during and after experimentation.

The animals were divided into 5 groups of 6 animals in each group. Group I served as control, group II as standard and groups III, IV and V served as test groups respectively. Drugs were administered orally, using oral feeding tube fit on a 1 ml syringe, once daily in the morning, for a period of 15 days, according to the groups as per the table below.

Table 1

Details of drug administration

GROUP	STUDY	TREATMENT
I	CONTROL	Normal feed + Water
II	STANDARD	Normal feed + Water + Tab. Imipramine 20 mg/kg orally
III	TEST - 1	Normal feed + Water + Aqueous extract of C. citratus 5mg/kg orally
IV	TEST - 2	Normal feed + Water +Aqueous extract of C. citratus 10mg/kg orally
V	TEST - 3	Normal feed + Water + Aqueous extract of C. citratus 10mg/kg + Tab. Imipramine 10mg/kg orally

ORAL FEEDING TECHNIQUE

18 – 20 G hypodermic needle which is blunted at the tip with a small ball soldered around the tip was used. The needle was attached to 1 ml syringe containing the drug to be administered. Each mouse was grasped gently and secured by the nape of the neck, holding the whole animal with the left hand. After introducing the oral feeding tube laterally through the interdental space, it was advanced into the oesophagus with a gentle rotatory movement. Once it reached the desired level the drug was gently pushed inside.

TAIL SUSPENSION TEST

On day 1, day 8 and day 15, tail suspension test was carried out for the control, standard, test-1, test-2 and test-3 groups, one by one, group wise. After one hour of drug administration, tail suspension test was done, for all the 6 animals belonging to each group, at a time.

Prior to tail suspension, the camera was set in position in order to obtain the highest possible resolution of the animals. Plastic tubes, which act as climb stoppers, were introduced to the tail, prior to application of adhesive tapes. Tapes were applied to

the tail tips leaving the last 2-3 mm free. Application of tapes was done gently to prevent apprehension and stress in animals even before suspension itself.

Then the mice were suspended one by one, by attaching the free end of the tape to the wooden bar, of a table with three walled rectangular chamber of dimensions, height 55 cm x breath 20 cm x depth 12 cm. The compartments were adequately sized to prevent mouse getting in contact with the walls and the distance between the tips of the nose to the table was approximately 20 cm.

The camera was set in such a way that the view was not obscured. The entire tail suspension time (TST) for the first 6 minutes was recorded without any interruption. At the end of 6 minutes the mice were placed back into the home cage after removing the tapes gently from their tails.

TAIL SUSPENSION TEST – BEHAVIORAL ANALYSIS

Tail suspension time was the first 6 minutes of hanging. The behavioral assessment of all the animals was done from the video recordings. Each animal was totally observed for 6 minutes. In this, the first 2 minutes were left for the animal to acclimatize and the behavioral assessment was done for the next 4 minutes.

During the behavioral assessment, the time that each mouse spent as immobile was noted, using a stop watch. The most important aspect of TST is the consistent differentiation between mobility and immobility. Escape related behaviors like movement of four limbs, shaking the body, running like limb movements, trying to touch the side walls were considered as mobility whereas small movements of the forelimbs alone

TAIL SUSPENSION TEST

without involvement of hind limbs and pendulum like movement of the tape due to momentum gained by the previous mobility were taken as immobility only. For each mouse, the immobility period was noted and the findings were tabulated. The immobility period recorded by doing tail suspension test in mice, after administration of test and standard drugs were subjected to statistical analysis.

RESULTS

STASTICAL METHODS APPLIED

ONE - WAY ANOVA - ANALYSIS OF VARIANCE

The One - way ANOVA procedure produces a one - way analysis of variance for a quantitative dependent variable by a single factor (independent) variable. Analysis of variance is used to test the hypothesis that several means are equal. This technique is an extension of the two-sample t test.

POST HOC TEST - TUKEY'S POST HOC TEST

Once it is determined that differences exist among the means, post hoc range test and pair wise multiple comparisons can determine which means differ. Range tests identify homogenous subsets of means that are not different from each other. Pair wise multiple comparisons test the difference between each pair of means, and yield a matrix.

TAIL SUSPENSION TEST DONE IN CONTROL GROUP

Control group mice were treated with normal feed and water and tail suspension test was done one hour after oral treatment on day 1, day 8, day 15 and the immobility time was observed. The mean immobility period was calculated for the control group and expressed as mean ± S.D. The mean immobility period for day 1 was 196 ± 5.71, on day 8 mean immobility period was 193 ± 12 and on day 15 mean immobility period was 199.5 ± 2.4.

Table 1:

TST results for control group

Day	Immobility period (seconds) in control group mice						Mean ± S.D.
	1	2	3	4	5	6	
Day 1	198	201	189	203	197	190	196 ± 5.71
Day 8	196	198	197	200	201	195	193 ± 12
Day 15	196	198	201	203	199	200	199.5 ± 2.4

TAIL SUSPENSION TEST DONE IN STANDARD GROUP

Standard group mice were treated with Tab. Imipramine 20 mg/kg orally and 1 hour after that tail suspension test was done on day 1, day 8, day 15 and the period of immobility was observed. The mean immobility period for the standard group was calculated and expressed as Mean ± S.D. The mean immobility period for day 1 was found to be 207 ± 2.88, on day 8 mean immobility period was 167 ± 3.1 and on day 15 mean immobility period was 149.3 ± 2.4.

Table 2:

TST results for standard group

Day	Immobility period (seconds) in standard group mice						Mean ± S.D.
	1	2	3	4	5	6	
Day 1	210	206	208	211	203	207	207 ± 2.88
Day 8	171	168	167	172	165	164	167 ± 3.1
Day 15	150	147	151	153	148	147	149.3 ± 2.4

TAIL SUSPENSION TEST DONE IN TEST GROUP I

The mice in test group – I were treated with aqueous extract of lemon grass 5mg/kg orally and 1 hour after that the tail suspension test was performed on day 1, day 8, day 15 and the period of immobility was observed. The mean immobility period for the test group I was calculated and expressed as Mean ± S.D. The mean immobility period for day 1 was found to be 210 ± 2.31, and on day 8 the mean immobility period was 182 ± 3.2 while on day 15 the mean immobility period was found to be 162 ± 3.0.

Table 3:

TST results for test group I

Day	Immobility period (seconds) in test group I mice						Mean ± S.D.
	1	2	3	4	5	6	
Day 1	215	210	211	210	208	211	210 ± 2.31
Day 8	184	183	188	179	180	181	182 ± 3.2
Day 15	165	163	159	167	161	160	162 ± 3.0

TAIL SUSPENSION TEST DONE IN TEST GROUP II

The mice in test group – II were treated with aqueous extract of lemon grass 10mg/kg orally and 1 hour after that tail suspension test was done on day 1, day 8, day 15 and the period of immobility was observed. The mean immobility period for the test group II was calculated and expressed as Mean ± S.D. The mean immobility period for day 1 was found to be 199 ± 2.31, on day 8 mean immobility period was 171 ± 2.6 and on day 15 mean immobility period was 147.5 ± 3.2.

Table 4:

TST results for test group II

Day	Immobility period (seconds) in test group II mice						Mean ± S.D.
	1	2	3	4	5	6	
Day 1	201	199	200	197	202	196	199 ± 2.31
Day 8	170	172	175	169	168	173	171 ± 2.6
Day 15	149	151	147	142	150	146	147.5 ± 3.2

TAIL SUSPENSION TEST DONE IN TEST GROUP III

The Mice in test group – III were treated with aqueous extract of lemon grass 10mg/kg and Tab. Imipramine 10 mg/kg and 1 hour after that tail suspension test was done on day 1, day 8, day 15 and the period of immobility was observed. The mean immobility period for the Test group III was calculated and expressed as Mean ± S.D. The results were mean immobility period for day 1 was 202 ± 2.16, on day 8 mean immobility period was 181 ± 3.8 and on day 15 mean immobility period was 160 ± 2.3

Table 5:

TST results for test group III

Day	Immobility period (seconds) in test group III mice						Mean ± S.D.
	1	2	3	4	5	6	
Day 1	203	201	200	204	206	202	202 ± 2.16
Day 8	181	179	176	183	184	187	181 ± 3.8
Day 15	161	163	162	160	157	158	160 ± 2.3

The calculated mean immobility period was subjected to one way analysis of variance. Analysis of variance is used to treat the test hypothesis that several means are equal. This technique is an extension of the two-sample t test.

Table 5.5:

Mean immobility period in seconds on day 1, day 8, day 15 in all groups

DAY	CONTROL	STANDARD	TESTI	TEST II	TEST III
Day 1	196 ± 5.71	207 ± 2.88**	210 ± 2.31**	199 ± 2.31*	202 ± 2.16*
Day 8	193 ± 1.2	167 ± 3.1**	182 ± 3.2*	171 ± 2.6**	181 ± 3.8*
Day 15	199.5 ± 2.4	149.3 ± 2.4**	162 ± 3.0**	147.5 ± 3.2**	160 ± 2.3**

Not significant: $p > 0.05$; Significant: $p \leq 0.05^{*}$; Highly significant: $p \leq 0.01^{**}$

RESULTS OF ONE WAY ANOVA

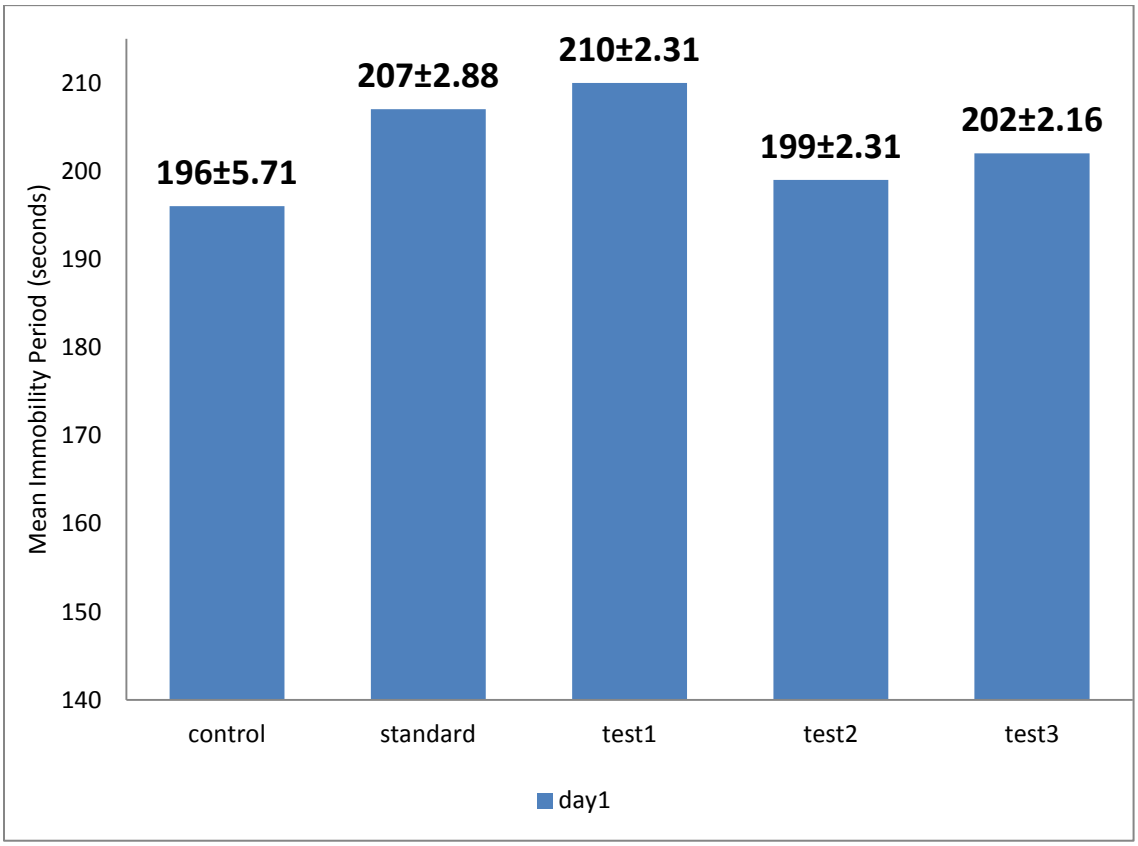
When the results of day 1, 8 and 15 were analyzed by one way anova, F value is significant (< 0.05) for the degree of freedom. So the difference in the mean immobility time between groups is significant.

POST HOC ANALYSIS

The results were analyzed with post hoc – Tukey's test which showed a significant difference in the immobility period in the standard, test-1, test-2, test-3 groups on day-1, day-8 and day-15 when compared to control. In some instances it was highly significant.

Figure 1:

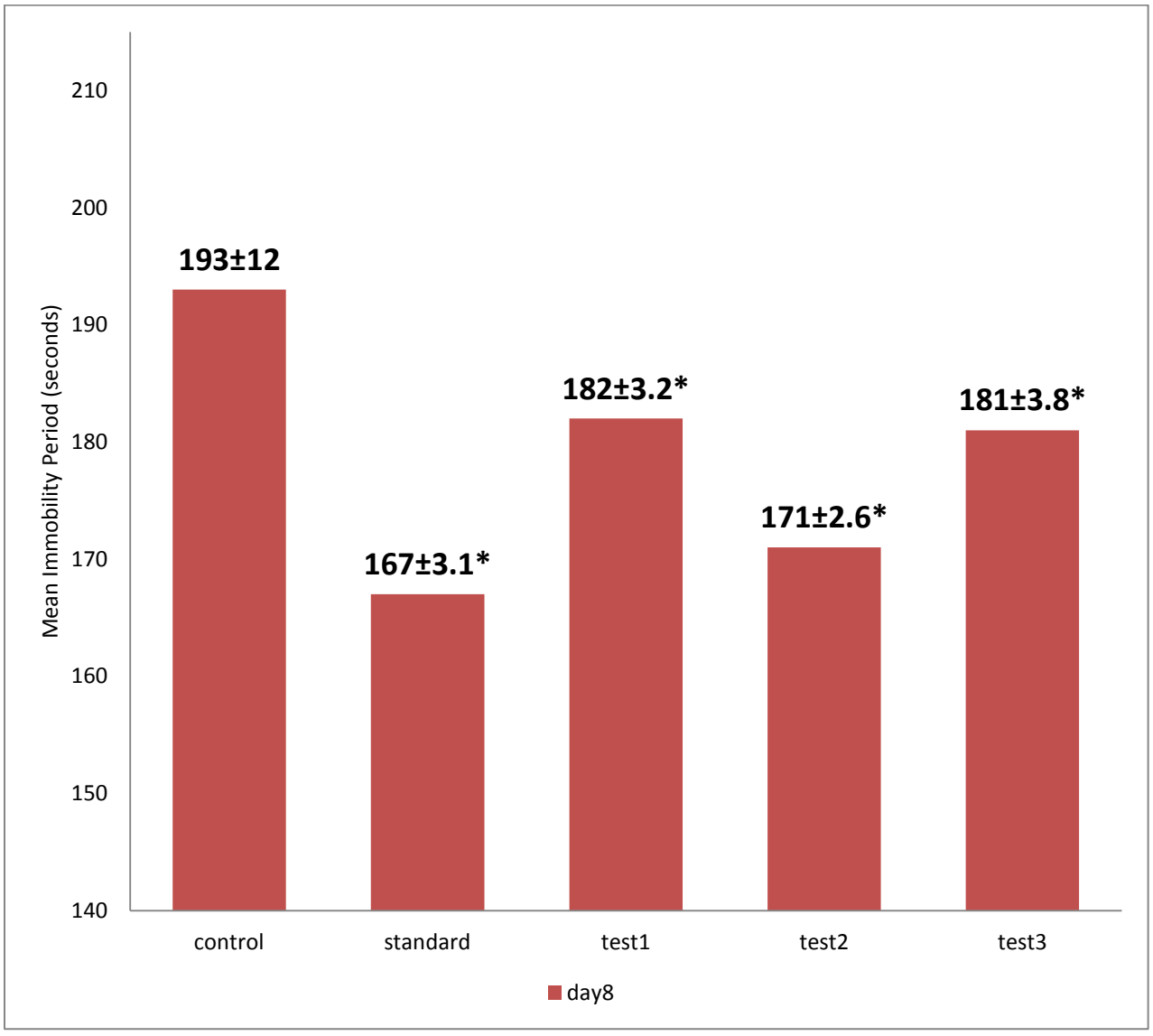
The mean immobility period in seconds on day 1 in all groups



On day 1, the mean immobility time showed that there was a significant increase in the immobility period in the standard and the three test groups when compared to the control group.

Figure 2:

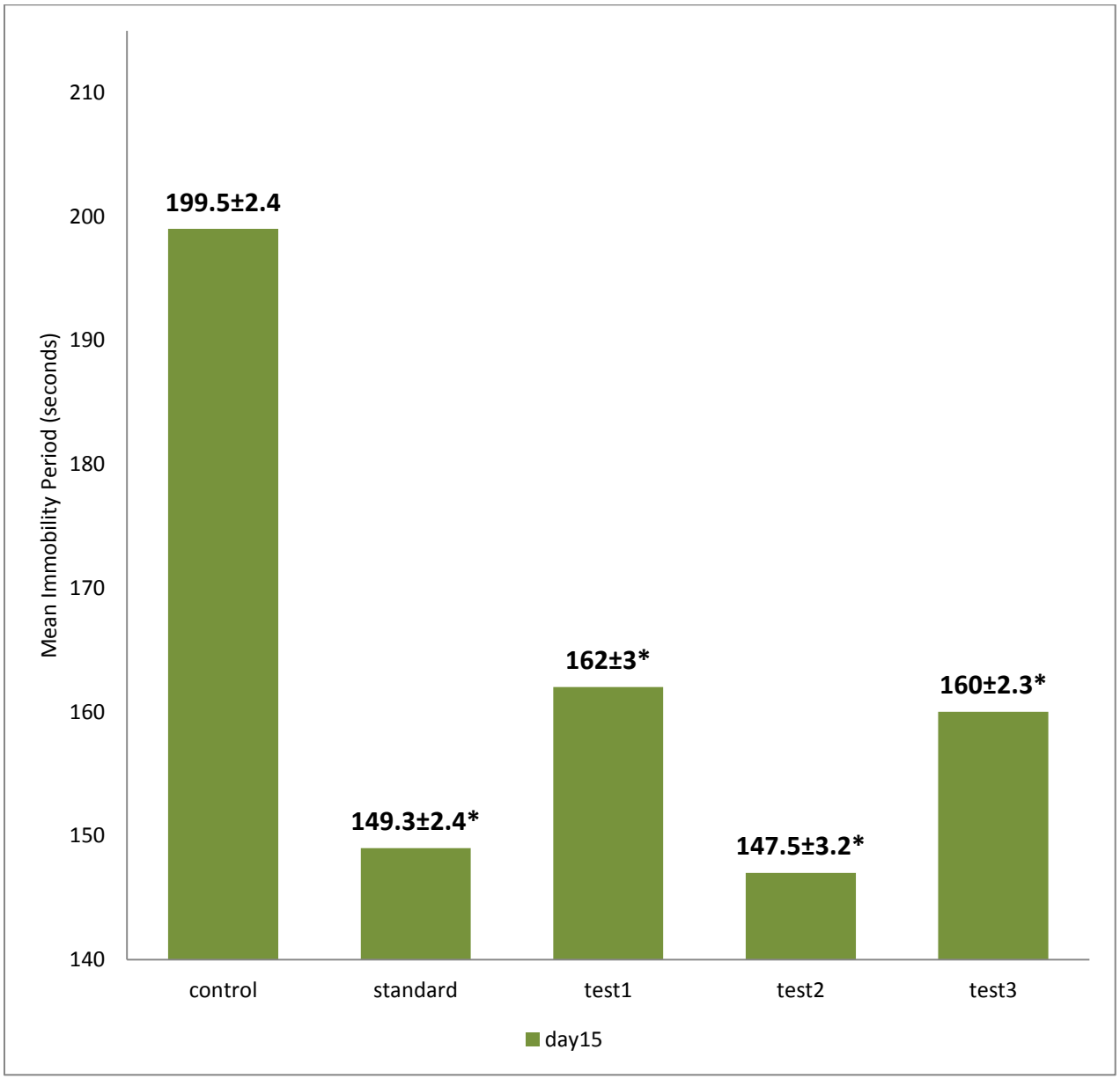
The mean immobility period in seconds on day 8 in all groups



On day 8, the mean immobility period showed that there was a significant decrease all the three test groups ($p \leq 0.05^{*}$), while it was highly significant decrease in standard group ($p \leq 0.01^{**}$) when compared to the control group.

Figure 3:

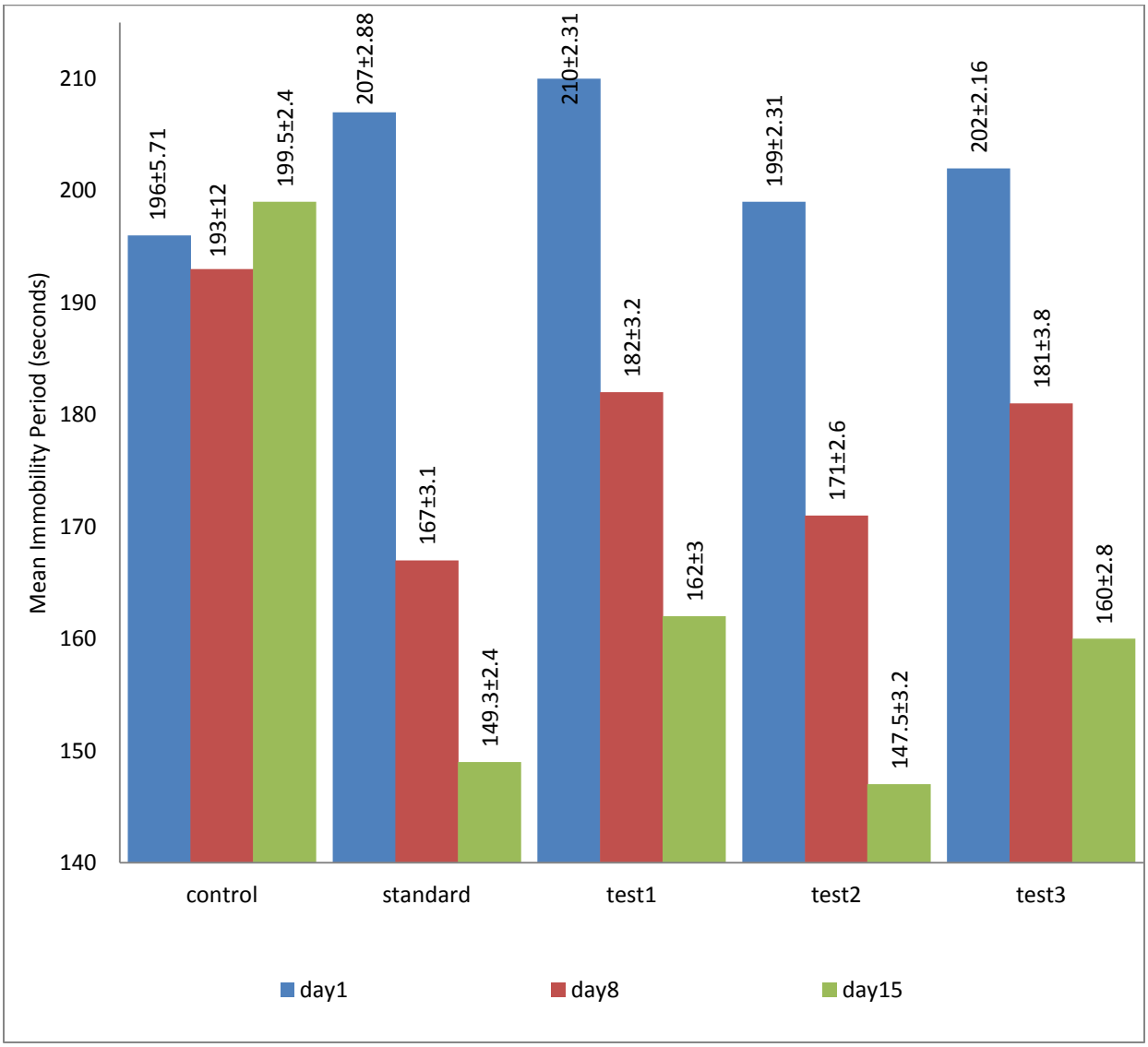
The mean immobility period in seconds on day 15 in all groups



On day 15, the mean immobility time showed that there was a highly significant difference in the standard and the three test groups ($p \leq 0.01^{**}$) when compared to the control group.

Figure 4:

The mean immobility period in seconds on day 1, day 8, day 15 in all groups



On day 1, the mean immobility time showed that there was a significant increase in the immobility period in the standard and the three test groups when compared to the control group.

On day 8, the mean immobility period showed that there was a significant decrease all the three test groups ($p \leq 0.05^{*}$), while it was highly significant decrease in standard group ($p \leq 0.01^{**}$) when compared to the control group.

On day 15, the mean immobility time showed that there was a highly significant difference in the standard and the three test groups ($p \leq 0.01^{**}$) when compared to the control group.

DISCUSSION

Depression is a common psychiatric disorder, leading to major cause of disease burden all over the world. It is steadily increasing in the recent past. It is a mood disorder presenting with sadness, loss of pleasure, lack of interest almost all days for at least two weeks. It aggravates the underlying medical problems and worsens their prognosis. It is potentially fatal since 50 % patients try to commit and 15 % die due to suicide.

With the introduction of pharmacotherapy depression is treatable now. All the available antidepressants have an array of adverse effects. With appropriate drug therapy only 60 % patients get remission. A most efficient drug, free from side effects is desired at this moment to check the increasing incidence of this disabling disease.

In our tradition various herbs are used since ancient times as a remedy for depression. They are found to have less adverse effects with good efficacy. St. John's wort, Kava, Saffron and Turmeric are proved to be effective antidepressants acting through various mechanisms. Till now, an ideal antidepressant is a hot topic of research. In this study, one such herbal remedy, *Cymbogon citratus* (Lemon grass) was evaluated for its antidepressant effect in albino mice using tail suspension test.

Among the behavioral models available, Forced Swim Test (FST) and Tail Suspension Test (TST) are widely adopted animal models for evaluating the antidepressant effect. Tail suspension test is based on the innate behavior, with a greater

sensitivity and high predictability. TST is easy to perform and well suited for high throughput screening of new compounds.

In this study, antidepressant effect of aqueous extract of lemon grass was assessed using tail suspension test by observing the immobility period. On day 1, it showed a significant increase in immobility period, in the standard and test groups when compared to control group. So it is evident that both drugs, imipramine and lemon grass extract, did not produce the desirable effect and the mechanism of which could not be explained.

On day 8, there was a highly significant decrease in immobility period in the standard group ($p \leq 0.01$) and a significant decrease in immobility period in the test groups ($p \leq 0.05$) when compared to control group. Additionally there was also a dose related effect in response with the test group. Lemon grass extract at a dose of 10 mg/kg administered, showed a greater decrease in immobility period compared to a dose of 5 mg/kg.

On day 15, there was a highly significant decrease in immobility period in the standard group and all the test groups ($p \leq 0.01$). It could be inferred that after continuous administration for 15 days, lemon grass extract at a dose of 10 mg/kg was more effective than the standard drug imipramine.

But when we analyze the results produced by co-administration of lemon grass extract at a dose of 10 mg/kg and imipramine at a dose of 10 mg/kg, it was found that lemon grass didn't produce synergistic effect with imipramine. It could be possible that

imipramine might have saturated the site of action of lemon grass. So there is a reduction in the response of lemon grass extract at a dose of 10 mg/kg when combined with imipramine. This was almost similar to that of lemon grass extract at a dose of 5 mg/kg.

It could be possible that lemon grass would have been acting as a partial agonist at the site of action of imipramine when given alone and oppose the effect of full agonist or it may be concluded that both lemon grass and imipramine compete for the same uptake mechanisms.

In a previous study done by Dudhgaonkar[57]*et al*, both tail suspension test and forced swim test were done after administration of lemon grass in graded doses and also along with imipramine. Imipramine was the standard drug. In their study also aqueous extract of lemon grass at a dose of 10 mg/kg showed maximum response (maximum reduction in immobility time). The effect of imipramine with lemon grass was less compared to lemon grass at a dose of 10 mg/kg.

It is postulated that depression is caused by impairment of monoaminergic transmission. Imipramine, which is a tricyclic antidepressant is an effective antidepressant by blocking the reuptake of noradrenaline and serotonin thereby enhancing monoaminergic neurotransmission in both adrenergic and serotonergic neurons. So, decrease in immobility time observed after administering lemon grass would have been due to an enhancement of monoaminergic transmission similar to that of imipramine.

Depression is a result of multiple etiological factors which include free radical injury of neurons also. Phytochemicals like phenols, flavanoids found in the aqueous

extract of lemon grass could be the cause for its free radical scavenging effect. Antioxidant property of lemon grass is already proved by various studies. So by preventing free radical injury it could have helped in depression.

In a study done by Rao[58] et al 2009, free radical scavenging ability of lemon grass was proved by different methods. Significant free radical scavenging effect was observed in in-vitro tests with 2, 2-diphenyl-2-picryl hydrazyl (DPPH) and nitric oxide free radicals. Naiyana*et al* 2010, also confirmed the antioxidant capacity of lemon grass by various methods like DPPH, Ferric Reducing Antioxidant Potential (FRAP). In another study by Patel *et al* 2006, extract from dried lemon grass had more antioxidants than from fresh lemon grass. Oboh[59]*et al.,* 2010, concluded hard water extract of lemon grass shows a significantly high antioxidant effect compared to cold water extract.

Myeloperoxidase enzyme has a dominant role on inflammation. Flavanoids present in lemon grass react with myeloperoxidase in the presence of H_2O_2 and prevents lipid peroxidation and tyrosine nitration of apoproteins. In a previous study done by Figueirinha*et al.,* 2010, aqueous extract of lemon grass inhibited the expression of iNOS, production of NO, PGE_2 in the LPS induced macrophage cell lines and in dendritic cell lines[60]. In another study done by Francisco *et al.,* 2011, aqueous extract of lemon grass inhibited the activation of NF- kB. Above studies prove the anti-inflammatory effect of aqueous extract of lemon grass.

So it could be possible that by various mechanisms, like monoamine transmission enhancement similar to imipramine, antioxidant effect and anti-inflammatory effect due

to the presence of flavanoids and phenols, aqueous extract of lemon grasshas antidepressant property which is showed by reduction in the immobility time in tail suspension test.

CONCLUSION

AND

SUMMARY

Depression is a major public health problem. It contributes to 4th leading cause of global disease burden ultimately resulting in a great economic loss to the society due to loss of productivity and utility of medical resources. There is also an increased suicidal tendency which results in loss of lives at productive age group.

Early identification and treatment at grass root level can bring down the morbidity and mortality. Now with a better understanding of etiopathogenesis of the disease, with advent of new drugs and other treatment modalities for depression, it is possible to treat the disease. Pharmacotherapy is the corner stone in the management of major depressive disorder. By combining non pharmacological therapy like Cognitive Behavioral Therapy (CBT) and interpersonal therapy, depression is almost curable now.

Even though various groups of medicine are currently available, no drug is free from adverse effects. Tolerability, safety at over doses is also a problem with many antidepressants. Noncompliance is approximately 40% in patients taking drug therapy alone because of delayed onset of therapeutic effects. In a good compliant patient too, who is adequately managed with drugs alone, remission is only in 60-70%.Hence an ideal drug exhibiting better safety and tolerability, which cures depression most efficiently is sought for.

Many effective drugs have come from a wide armamentarium of herbs. Already natural products like St. John's wort, saffron are effective for treating mild to moderate depression. In this present study, *Cymbogon citratus* which is used for various illnesses

as traditional medicine was evaluated for it's the antidepressant effect in albino mice, using tail suspension test.

It was observed that *Cymbogon citratus* (lemon grass) at low dose (5mg/kg) showed significant antidepressant effect on 15^{th} day and at high dose (10 mg/kg), showed significant antidepressant effect on 8^{th} day itself which was comparable to that of imipramine. Combined effect of lemon grass at 10 mg/kg and imipramine 10 mg/kg is not synergistic.

The study objective has been achieved. Further studies need to be done with more number of animals and different experimental models, to know the exact molecular and biological mechanism behind *Cymbogon citratus* as an anti depressant. Identification and separation of the active principle that is responsible for the antidepressant effect has to be further evaluated. That will help the society in the near future.

TAIL SUSPENSION TEST

ANNEXURES

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MASTER CHART

TST on day 1 showing immobility period in seconds

S. No.	CONTROL	STANDARD	TEST 1	TEST 2	TEST 3
Mouse-1	198	210	215	201	203
Mouse-2	201	206	210	199	201
Mouse-3	189	208	211	200	200
Mouse-4	203	211	210	197	204
Mouse-5	197	203	208	202	206
Mouse-6	190	207	211	196	202

TST on day 8 showing immobility period in seconds

S. NO.	CONTROL	STANDARD	TEST1	TEST2	TEST3
Mouse-1	196	171	184	170	181
Mouse-2	198	168	183	172	179
Mouse-3	197	167	188	175	176
Mouse-4	200	172	179	169	183
Mouse-5	201	165	180	168	184
Mouse-6	195	164	181	173	187

TST on day 15 showing immobility period in seconds

S. NO.	CONTROL	STANDARD	TEST1	TEST2	TEST3
Mouse-1	196	150	165	149	161
Mouse-2	198	147	163	151	163
Mouse-3	201	151	159	147	162
Mouse-4	203	153	167	142	160
Mouse-5	199	148	161	150	157
Mouse-6	200	147	160	146	158

ABBREVIATION

CBT	Cognitive Behavioral Therapy
MDD	Major Depressive Disorder
SSRIs	Selective Serotonin Reuptake Inhibitors
SNRIs	Serotonin Noradrenalin Reuptake Inhibitors
TCAs	Tricyclic Antidepressants
MAOIs	Monoamine Oxidase Inhibitors
WHO	World Health Organization
CSF	Cerebro Spinal Fluid
MRI	Magnetic Resonance Imaging
PET	Proton Emission Tomography
5-HT	5-Hydroxy Tryptamine
NA	Noradrenaline
VMAT	Vesicular Monoamine Transporter
HPA	Hypothalamo Pituitary Adrenal axis
CRH	Corticotrophin Releasing Hormone

BDNF	Brain Derived Neurotropic Factor
DSM-IV-TR	Diagnostic and Statistical Manual of Mental disorders, Fourth edition, Text Revision
ICD-10	International Classification of Diseases
ECT	Electro- Convulsive Therapy
TMS	Transcranial Magnetic Stimulation
SERT	Serotonin Transporter
NET	Nor epinephrine Transporter
GMP	Guanylyl Mono Phosphate
CREB	cAMP Responsive Element Binding Protein
NaSSA	Noradrenergic and Specific Serotonergic Antidepressant
NMDA	N- Methyl D- Aspartate
AMPA	α- Amino- 3- hydroxyl 5- Methyl- 4-isoxazole Propionic Acid
SIADH	Syndrome of Inappropriate Anti Diuretic Hormone secretion
MI	Myocardial Infarction
SAM-E	S-Adenosyl Methionine

FDA	U. S. Food and Drugs Administration
GABA	Gamma- Amino butyric acid
ACTH	Adrenocorticotropic Hormone
PGE_2	Prostaglandin E_2
NO	Nitric Oxide
iNOS	Inducible Nitric Oxide Synthase
COX-2	Cyclo-oxygenase- 2
CPCSEA	Committee for the Purpose of Control and Supervision of Experiments on Animals
TST	Tail Suspension Test
NF- kB	Nuclear Factor- kB

ETHICAL CLEARANCE LETTER

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ETHICAL CLEARANCE CERTIFICATE

DR.S.REVWATHY M.D., D.G.O., D.N.B.,Dean & Chairman Animal Ethical committee, Madurai Medical College, Madurai, hereby endorse ethical clearance to the proposal.

EVALUATION OF ANTIDEPRESSANT EFFECT OF *CYMBOPOGON CITRATUS* (LEMON GRASS) IN ALBINO MICE

Submitted by
Dr.R.Mangaladevi,
Post graduate student,
Institute of pharmacology
Madurai Medical College,
Madurai.

The study did not violate the regulations and guidelines prescribed by ICMR and are within the permitted norms of animal experimentation in this country. The outcome of the study may be beneficial to the human and animals.

Dean & Chairman

Date :
Place: Madurai

Office seal:

PLANT IDENTIFICATION CERTIFICATE

Dr.D.Stephen MSc, Ph.D
Assistant Professor

Department of Botony
The American College
Madurai

CERTIFICATE

This is to certify that the specimen brought by **DR. R. MANGALA DEVI,** postgraduate in MD Pharmacology, Institute of Pharmacology, Madurai Medical College, Madurai is identified as ***Cymbopogon citratus*** (Lemongrass) belonging to the family of **Poaceae.**

Station : Madurai
Date : 2.12.2015

Dr.D.Stephen MSc, Ph.D

Dr. D. STEPHEN, Ph.D.,
ASST. PROFESSOR IN BOTANY
THE AMERICAN COLLEGE
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