Potential mechanisms underlying the therapeutic roles of Flavonoid from *Sedum lineare* Thunb in depression: Based on molecular docking study

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Abstract

Background: People all across the world suffer with depression, a chronic illness that can be fatal. The medications used to treat this illness have a number of negative effects and may interact with other drugs or foods. Furthermore, only thirty percent of patients take their current meds as prescribed, and the remaining patients do not fully heal. Finding a medication that is reasonably effective, inexpensive, and has few adverse effects appears to be required.*Sedum lineare* "Sedi linearis Herba" is the name given to the Thunb. plant as a medicinal substance. It was frequently employed as a treatment for a variety of illnesses, including hepatitis, dysentery, dermatitis rhus, burns, scalds, traumatic bleeding, and a host of other ailments. It has been demonstrated that this plant have the ability to alter thyroid hormone in addition to having antibacterial, anticancer, nephroprotective, analgesic, anti-inflammatory, and anti-diarrheal properties.

Aim and Objective: The current study's objective was to assess the anti-depressant efficacy of kaempferol*via in-silico* molecular docking.

Method: Studies using in-silico molecular modelling were conducted to evaluate the antidepressant potential of kaempferol by designing it to target both the ***5HT-1 and 5HT-2*** receptors. The Auto Dock software was utilized to determine the binding through a grid-based docking technique.

Result: The results of the lead molecule's molecular modelling with 5HT-1 and 5HT-2 receptors demonstrated that the chosen compounds had a high affinity for the chosen target protein. It was discovered that the binding energies of kaempferol to the 5HT-1 and 5HT-2 receptors were -6.67 and -6.92 Kcalmol-1, respectively.

Key words: Depression, kaempferol, *5HT-1 & 5HT-2* receptors.

Introduction

The primary cause of disability, depression is thought to impact 121 million people worldwide. Suicide is a tragic death that claims the lives of approximately 850,000 people annually and can be caused by depression. According to a World Health Organization poll, depression will move up to the second spot on the handicap table in 2020. For all ages and both sexes, the Adjusted Life Year (DALY) is determined. Currently, depression ranks second among all sexes in the 15-44 age range in terms of DALY [1].

One of the most frequently seen mental illnesses, depression has an impact on our thoughts, feelings, behaviours, and overall wellbeing. By 2030, the WHO predicts that it will be the primary cause of numerous additional diseases. Clinical depression is treated with a variety of synthetic medications, each with its own set of drawbacks, such as a delayed rate of action onset, poor remission, and varying physiological reaction rates to depression-related problems. Furthermore, patients cannot receive clinical therapy until it poses a risk to their ability to work or maintain a family. Furthermore, synthetic medications often have a single target. In contrast to synthetic drugs, many plants contain flavonoids, act on multiple molecular targets, and influence multiple neurotransmissions or pathways to exhibit antidepressant effects. Examples of these include: B. inhibition of monoamine oxidase and tropomyosin receptor kinase B; concurrent increase in nerve growth and brain-derived neurotrophic factors; and B. noradrenalinergic, serotonergic, GABAergic, and dopaminergic pathways.

As per Oxford Dictionary **(2008)** depression defined as ***"A mental condition characterized by harsh feelings of hopelessness and insufficiency, typically accompanied by a lack of energy and interest in life."***

Mental depression is a chronic disorder that affects a person's behaviour, thinking, mood, and physical health. There are biological and emotional components that contribute to depressive symptoms. Biological symptoms include delayed thoughts, behaviours, and appetite; emotional signs include mystery, indecision, guilt, pessimism, inadequacy, low self-esteem, feelings of ugliness, indecision, and lack of motivation [2].

Particularly, unipolar depression and bipolar depression are the two primary forms of depression. In almost 75% of instances, mood swings in unipolar depression are consistently on the same path, are not commonly related with traumatic life experiences, and are

supplemented by signs of worry and agitation. These include bipolar depression (which accounts for around 25% of instances), also known as endogenous depression, which typically manifests in early adulthood, shows recognisable patterns, and varies over a few weeks between periods of despair and mania[3].

The study and development of novel chemicals having specific effects on humans is known as drug designing. Consequently, it is imperative to develop novel bioactive substances with unique molecular structures and modes of action from the prescription medications that are presently available. The interdisciplinary, costly, and time-consuming process of finding new pharmaceuticals has changed as a result of recent developments in the production of novel medicinal substances. The cost of developing new drugs can be reduced by up to 50% with CADD technology. By using the molecular docking technique, one may comprehend the I drug-receptor interaction, II binding affinity, III orientation, and approach of drug molecules to the target site. Precise structural modelling and accurate activity forecasting are the two main objectives of docking research [4].

It has been stated that the Chinese herb *Sedum LinearеThunb. (SLT),* which is made from the whole grass of the Crassulaceae plant, has anti-inflammatory properties. The whole S. lineareThunb plant, known as Herba in Chinese, has long been used in Sedi Linear Traditional Chinese Medicine (TCM) to treat a variety of conditions, such as hepatitis, throat swelling, diarrhoea, dermatitis rhus, burns, scalds, and traumatic bleeding. Recently, there has been increased attention in the possibility that Sedi linearis Herba can inhibit the growth of malignant cells, especially in the Tujia minority region in western Hubei, China.*S. linеare*Thunb extracts contain various secondary metabolites, including phenolic compounds, flavonoids, and other bioactive compounds. [5].As per literature survey it had been found that plant hydro-alcoholic extract is rich source of quercetin and kaempferol. So kaempferol was chosen as lead molecule for *in-silico* docking study.

S.No.	Descriptionof Kaempferol [6]	
1.	Molecular formula	$C_{15}H_{10}O_6$
2.	Molecular weight	286.24 g/mol
3.	Source	Polyphenol antioxidant found in fruits and vegetables
4.	Category	Flavonol
5.	Pharmacology	It is essential components in cellular signal transduction pathways connected to apoptosis, angiogenesis, inflammation, and metastasis on a molecular level. Significantly, kaempferol suppresses cancer cell proliferation, angiogenesis, and death while also appearing to maintain normal cell viability and occasionally exhibiting a protective impact.

Experiment work

Molecular docking studies

Ligand Preparation:

Using ChemSketch [7], the two-dimensional structure of the produced ligand was transformed into their three-dimensional structures, which were optimized with three-dimensional geometry. For AutoDock compatibility, the optimized structure was saved in PDB format. Below are the fundamental structures of the prepared ligand:

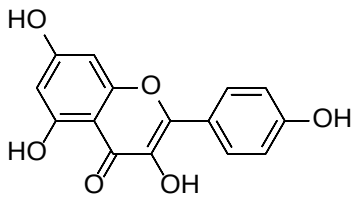


Figure 1: 2D structure of kaempferol

Preparation of the grid file

By creating a grid box around the active locations, Autodock was able to designate its regions of interest. Since the grid box is designed to cover every amino acid found in active sites that

is required for binding aside from those found in receptors, it serves a crucial function in the docking process. Three thumbwheel widgets on the grid box allow us to adjust the number of points in the x, y, and z dimensions. Table 1 provides the grid points and spacing for every receptor that was taken into consideration in this investigation [8–9].

Table 1. Grid parameters used in current docking analysis of 5-HT1 and 5-HT2 receptor

S. No.	Receptor	x-axis	y-axis	z-axis	Spacing	x center	y center	z center
1	5-HT1	40	40	40	0.375	102.141	115.111	108.423
2	5-HT2	40	40	40	0.403	152.197	154.556	131.732

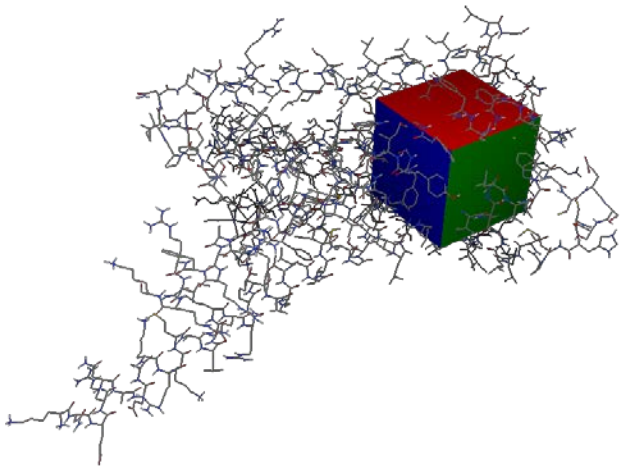


Figure 2: Grid box covering all active sites in 5-HT1 receptor

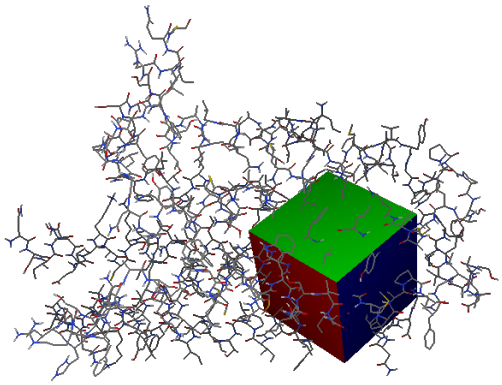


Figure 3: Grid box covering all active sites in 5-HT2 receptor

Preparation of the docking file

Every computation was performed using Autodock 4.2 as the docking tool. Pymol, Chimera, DS visualizer, MMP Plus, and other programmes were used to carry out the visualization and other tasks required for docking investigations [10–11].

Docking Study

Crystal structure

The protein's crystal structure, which includes the 5-HT1 and 5-HT2 receptors, from the Protein Data Bank website was obtained. The Protein Data Bank contains all of the primary data pertaining to the structure of the receptors [12–14]. Chimera software was used to separate the complex ligand for each of the target receptors.

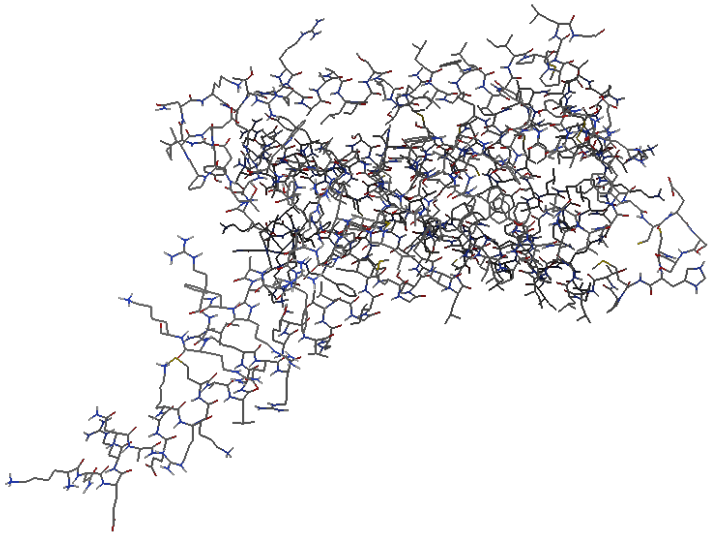


Figure 4: Crystal structure of 5-HT1 receptor (PDB ID-7e2y)

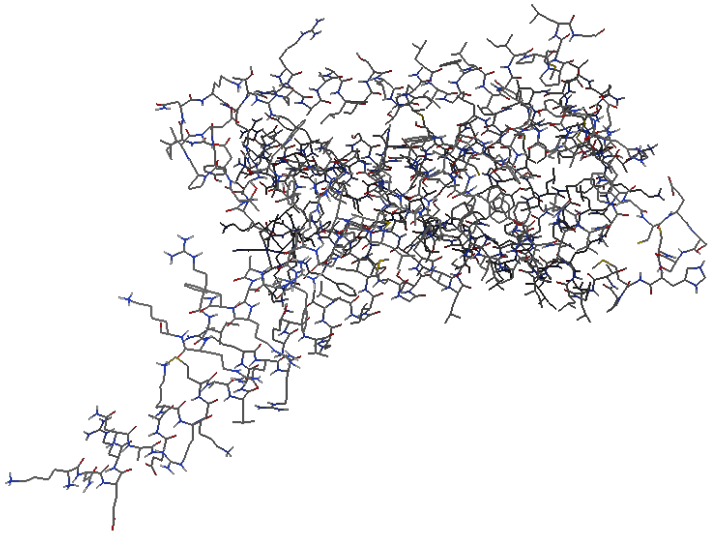


Figure 5: Crystal structure of 5-HT2 receptor (PDB ID-7ran)

Processing of Protein

The single chain present in all downloaded receptor proteins is chain A, which was chosen for experimental purposes and had its complex ligand removed. Chimera software was used to extract the bound ligand from the macromolecular complex [15–16].

Molecular Docking Simulation Studies

Autodock was used to dock the ligand kaempferol against the 5-HT1 and 5-HT2 receptors. Every ligand's link was maintained in its flexible state, while the receptor's residues were not [17–18].

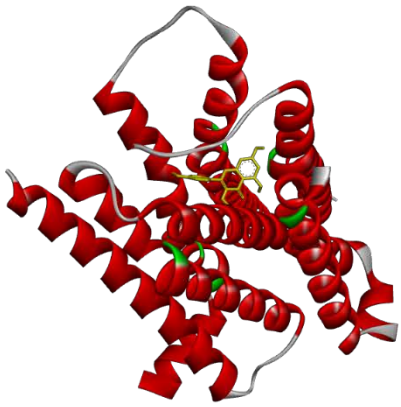


Figure 6: Binding mode of kaempferol within the active site of 5-HT1 receptor

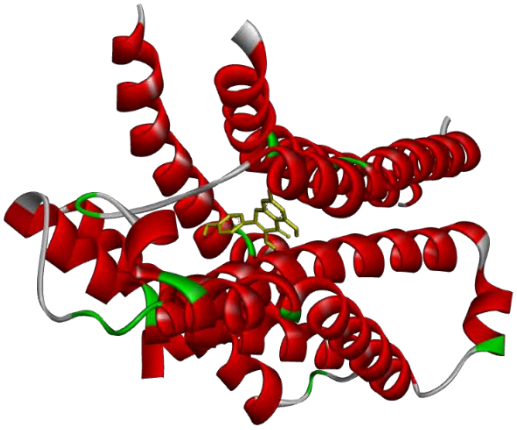


Figure 7: Binding mode of kaempferol within the active site of 5-HT2 receptor

Toxicity & ADME-T Studies

The online programme OSIRIS examined the ligand molecules, namely kaempferol, in order to anticipate the presence of any hazardous groups as well as the ADME-T characteristics and their presence [19].

RESULTS AND DISCUSSION

Both depression and its public health are grave concerns. Although a variety of variables can contribute to the development of depression, it is crucial to comprehend the disorder's effects, potential triggers, and methods of therapy in order to support the wellbeing of individuals who are impacted. To establish the necessity and length of ongoing treatment, it is also necessary to research the global history of depression illnesses. In order to effectively treat depression in primary care, studies should also assess affordable treatment methods.

As unlike to synthetic medications, many plants contain flavonoids that act on several molecular targets and have an antidepressant effect by influencing different neuronal transmissions or pathways, including dopaminergic, GABANergic, serotonergic, and noradrenergic; they also inhibit monoamine oxidase and tropomyosin receptor kinase B; and they simultaneously promote nerve growth and brain-derived neurotrophic factors. These

herbal medications containing flavonoids are probably beneficial for people who suffer from subclinical depression.

One monoamine neurotransmitter that is crucial to physiological processes is serotonin (5-HT). 5-HT has been linked to pathological states, such as problems involving mood, anxiety, psychosis, and pain, as well as sleep, feeding, sexual behaviour, temperature regulation, pain, and cognition. 5-HT1A receptors have long been thought to be a promising target for the antidepressant effects of pharmaceuticals. A novel class of antidepressant medications was suggested by the possibility of postsynaptic 5-HT1A agonists, and combination 5-HT1A receptor ligands/serotonin transporter (SERT) inhibitors appear to have an intriguing pharmacological profile. However, it should be mentioned that 5-HT1A receptors can communicate through both G protein-dependent and G protein-independent routes and trigger a variety of distinct metabolic pathways. Thus, the factors influencing the multiplicity of 5-HT1A receptor signalling pathways would be the culmination of effects exclusive to the host cell environment. Furthermore, in pre- and postsynaptic locations, receptor trafficking manifests itself differently. It should be mentioned that the 5-HT1A receptor's anxiolytic and/or antidepressant properties are also influenced by its interactions with other signal transduction systems, such as the glutaminergic and GABAergic systems, the 5-HT1B or 5-HT2A/2B/2C receptors, and others. Therefore, determining the molecular targets particular to the brain for 5-HT1A receptor ligands may lead to improved targeting, providing promise for more potent medications for a range of diseases.

When kaempferol was molecularly docked with the 5-HT1 and 5-HT2 receptors, Table 5 showed that it had demonstrated a chemical interaction with the amino acids in the active pockets, as seen in Figures7. The grid base parameter used for docking analysis was tabulated in table 1. The ligand molecules have demonstrated an encouraging docking score, theoretically. According to table 2, kaempferol's docking results with 5-HT1 and 5-HT2 were -6.67 and -6.97 kcal mol–1, respectively. This suggests that kaempferol is a powerful inhibitor of the 5-HT1 and 5-HT2 receptor. Figures 8–11 depicted the interplay between 2D and 3D binding. The binding interaction of kaempferol with 5-HT1 and 5-HT2 receptors showed C-H, covalent & pi-alkyl bounding(table 3).

The pharmacokinetic profile of kaempferol shows that while they have a decent pharmacokinetic profile, they also have significant hazardous effects, such as impacts on the

reproductive system and mutagenicity. Figures 12, display the pharmacokinetic and toxicity profiling data of ligands such as kaempferol.

Table 2: Results of docking of ligands like kaempferol against 5-HT1 and 5-HT2 receptor

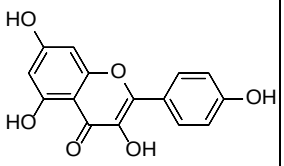
S. No	Compound Name	Structure	5-HT1	5-HT2
1	Kaempferol		-6.67 (ki: 12.97µM)	-6.92 (ki: 8.5µM)

Table 3: Binding interaction of lead molecule with receptor

Lead molecule & Receptor	CH-bonding	Covalent bonding	Vander Waals interaction	Pi-Alkylbonding	Pi-Sigma bonding
Kaempferol with 5HT1 receptor	Asp 116 Asn 386 Thr 121	-------	Ile 167 Phe 362 Ser 199 Thr 196 Tyr 390 Ile 189 Thr 200	Val 117 Cys 120 Ala 203	--------
Kaempferol	Ser 242	Phe 340	Ile 206	----------	---------

with 5HT2 receptor	Val 156 Thr 160 Asp 155 Val 235		Gly 238 Ser 239 Leu 236 Phe 339 Trp 336 Val 366 Tyr 370 Ser 159		

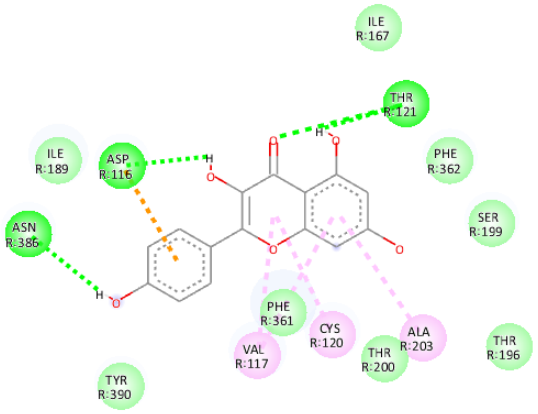


Figure 8: Two-dimensional binding mode of kaempferol within the active site of 5-HT1 receptor

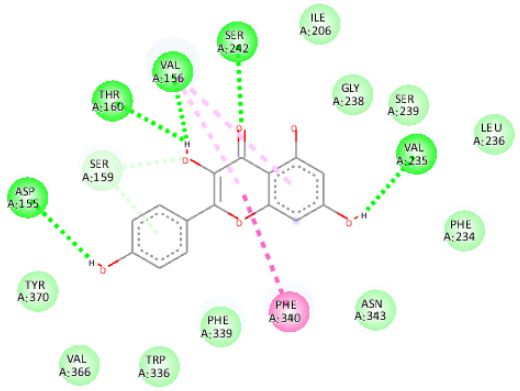


Figure 9: Two-dimensional binding mode of kaempferol within the active site of 5-HT2 receptor

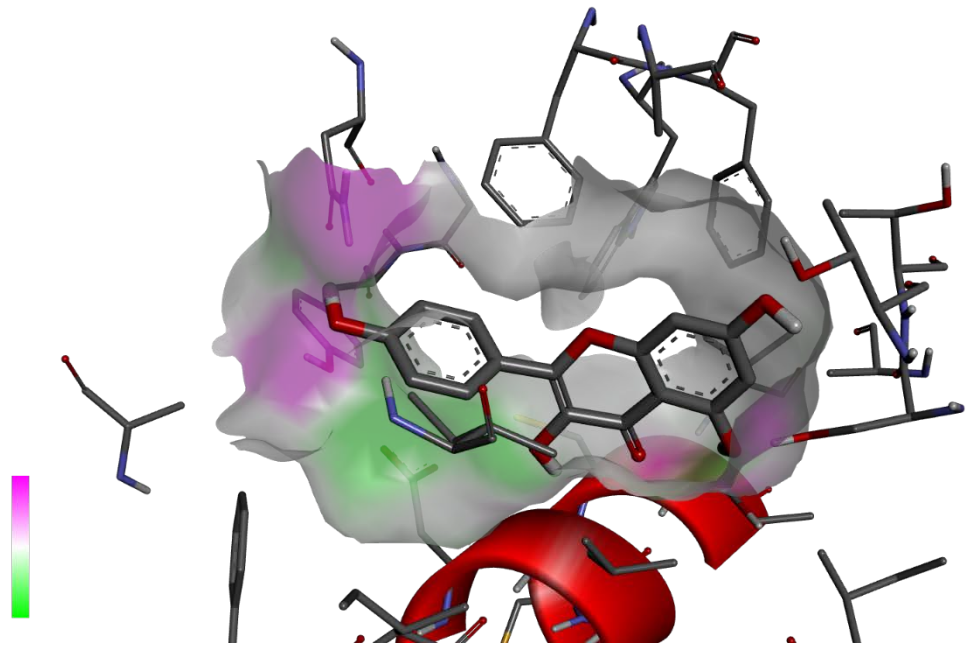


Figure 10: Three-dimensional binding mode of kaempferol within the active site of 5-HT1 receptor

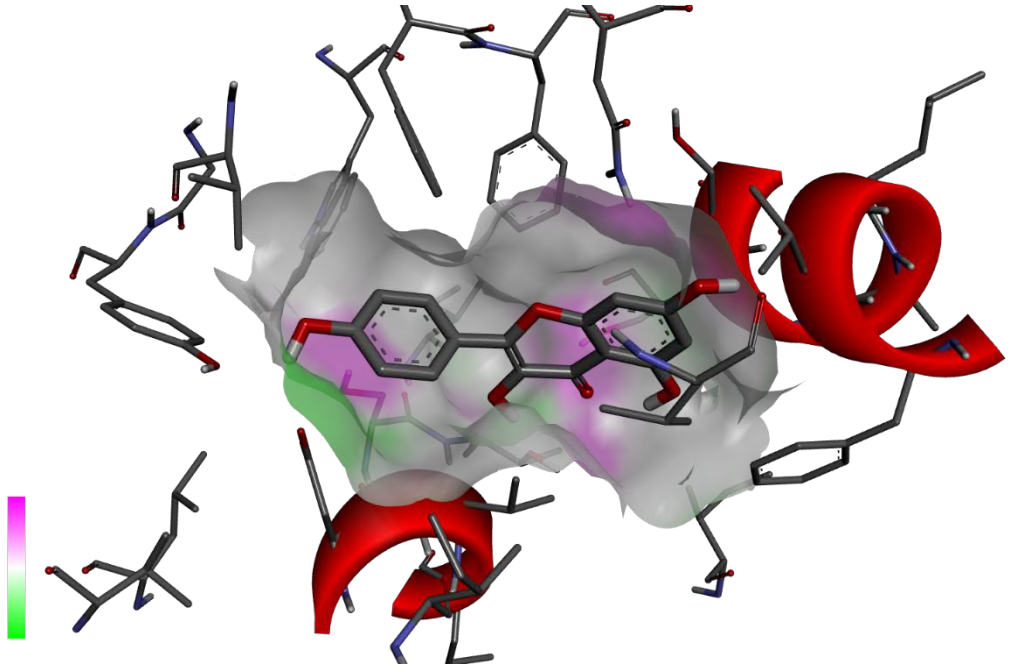


Figure 11: Three-dimensional binding mode of kaempferol within the active site of 5-HT1 receptor

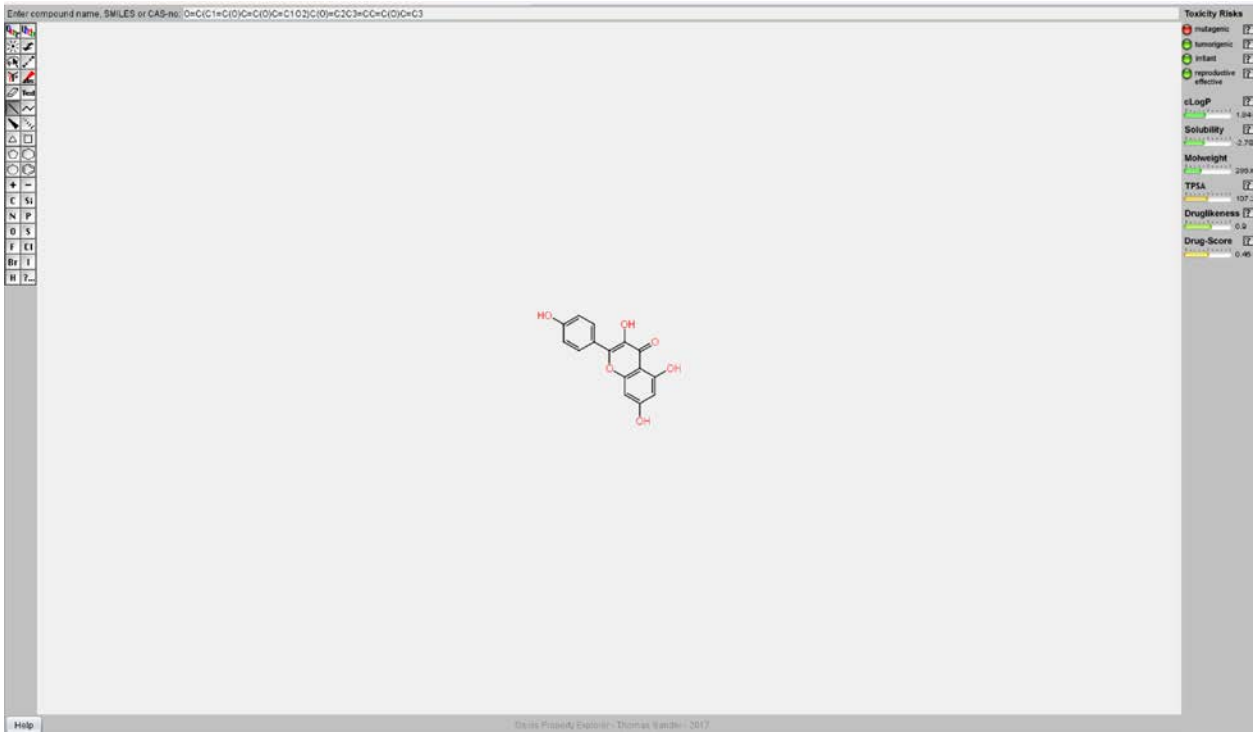
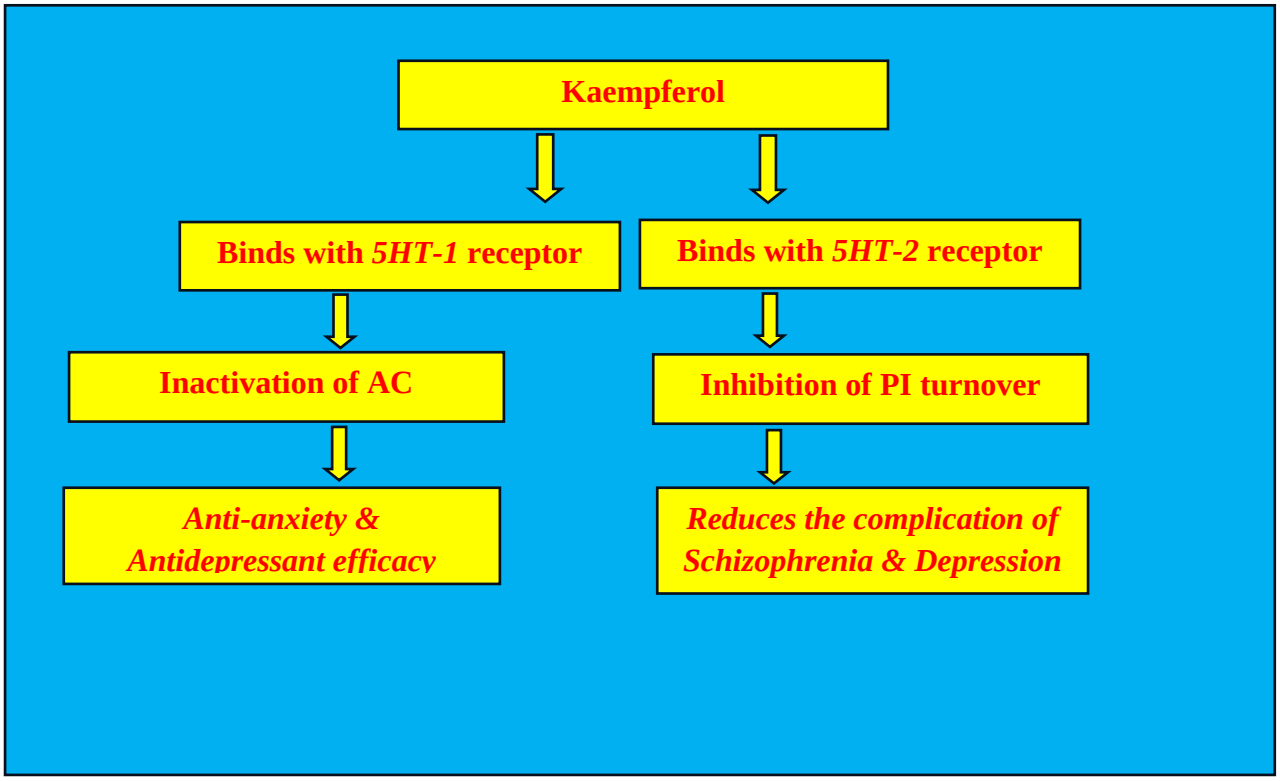


Figure 12: Pharmacokinetic and toxicity profiling of kaempferol

Divulgence of Investigation

*S. linearе*Thunb as a potential source of bioactive compounds was demonstrated since past era. The presence of diverse phytoconstituents, significant levels of phenols and flavonoids, and the identification of quercetin offer promising avenues for future research and the development of natural therapeutics.Over the past fifteen years, flavonoids have come to be known for much more, and some positive effects have been reported. It seems that flavonoids and mental health interact in a very important way. One flavonoid antioxidant that can be found in fruits and vegetables is kaempferol. The protective benefits of dietary kaempferol in lowering the risk of chronic diseases, including cancer, have been documented in numerous research. However, very little is understood about the cellular and molecular processes that underlie the effects of kaempferol on the central nervous system (CNS). Furthermore, it is unknown how the structural characteristics of kaempferol, their glycosylation, and the biological advantages of these molecules relate to one another. This study used molecular docking to assess kaempferol's potential as an antidepressant. The outcome demonstrated that kaempferol, the chosen lead chemical, had a strong inhibitory effect on both 5-HT1 and 5-HT2 receptors. The finding of current investigation showed that the lead compound (kaempferol) found in *S.linearе*justify the anti-anxiety & anti-depressant potential.The visual representation of kaempferol's putative antidepressant mechanism is as follows:



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