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BRIDGING ETHNOMEDICINE AND MODERN SCIENCE: PHYTOCHEMICAL AND PHARMACOKINETIC STUDY OF THE COSTUS IGNEUS (INSULIN PLANT)

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Abstract:

Costus igneus, also referred to as the "Insulin Plant," is a tropical medicinal plant with proven antidiabetic activity and common use in traditional treatment of metabolic disorders.This research sought to investigate the phytochemical composition, total phenolic content, and pharmacokinetic characteristics of Costus igneus leaf extracts that were procured using methanolic, aqueous, and hydroalcoholic extraction procedures. Initial screening showed the occurrence of major bioactive compounds like flavonoids, alkaloids, terpenoids, saponins, phenolics, and glycosides, which have been reported to be responsible for the drug effects of the plant. Total phenolic content quantification showed the high occurrence of antioxidant components, indicating good therapeutic potential. Also, in silico analysis with SwissADME was utilized to assess pharmacokinetic properties of major phytoconstituents like quercetin, kaempferol, diosgenin, and corosolic acid. These compounds had desirable drug-likeness profiles, gastrointestinal absorption, and low cytochrome P450 inhibition, validating their potential as therapeutic compounds. The findings validate the historical claims of the antidiabetic, antioxidant, anti-inflammatory, hepatoprotective, and nephroprotective activities of Costus igneus. This research fills the gap between traditional use and scientific proof, providing information for further development of standardized herbal drugs and furthering the inclusion of Costus igneus in evidence-based phytomedicine**.**

Keywords**:** Costus igneus, Phytochemical screening, Antidiabetic activity, SwissADME, Bioactive compounds, Herbal medicine

I. INTRODUCTION

Naturally occurring plant bioactive substances called phytochemicals help shield plants from disease-causing organisms and environmental stressors. The potential therapeutic uses of phytochemicals have attracted a lot of scientific interest, especially in light of developments in analytical and screening technologies (Muthukumar et al., 2019). Approximately 14–18% of higher plant species are used for medicinal purposes worldwide, and ethnobotanical methods have been used to identify over 74% of pharmacologically active chemicals derived from plants (Rani, 2019). The "insulin plant," or Costus igneus, is a perennial herbaceous plant that belongs to the Costaceae family. It is native to Southeast Asia and has long been used as a natural treatment for diabetes in India. Patients with diabetes typically consume one leaf daily. The plant, also known as spiral flag or fiery costus, has spirally arranged leaves and beautiful orange flowers. Phytochemical studies of Costus igneus have revealed the presence of a

wide range of bioactive compounds, including flavonoids, alkaloids, tannins, saponins, terpenoids, steroids, cardiac glycosides, and phlobatannins. These compounds have been linked to a variety of pharmacological activities, including anti-diabetic, anti-inflammatory, antimicrobial, antioxidant, and hypolipidemic activities. Flavonoids in general have been thoroughly studied for their anti-diabetic activity, and substances like quercetin found in Costus igneus leaves are effective antioxidants that help improve insulin sensitivity and glucose consumption in peripheral tissues. Chromatographic procedures, including TLC and HPLC, have been used to identify and quantify flavonoids, including quercetin, in Costus igneus methanolic extracts and validate their potential therapeutic value (Peasari et al., 2018). Quercetin's capacity to bind to significant sites in the insulin signaling cascade has also been demonstrated by computational modeling, making it a promising medication option for the treatment of diabetes. Given the rise in diabetes cases worldwide and the shortcomings of conventional synthetic medications, Costus igneus is a rich source of natural compounds that provide affordable and effective antidiabetic treatments.

Morphology

It is a perennial, honest, spreading plant, which is about two meters high, the highest stalk fell and lay on the ground. The leaves are simple, alternative, complete, rectangular, evergreen, with parallel honor with a length of 4-8 inches. Large, smooth, dark green leaves of this tropical evergreen have light purple undercords and are arranged for the spiral around the stems, which are generated from attractive, armpits bundle from underground root trunks. Beautiful, 1.5-inch diameter, orange flowers are produced in warm months, and appear on the head -like head of the proposals for branches. The fruits are inconsistent, not flashy, less than 0.5 inches and green in color(Gilman, 2012).

Fig No.1 Costus igneus leaf

Taxonomy:

Table no. 1: Taxonomy of Costus igneus (Hegde, Rao, & Rao, 2014)

Heirarchy	Costus igneus
Domain	Eukaryota
Kingdom	Plantae
Sub-kingdom	Viridaeplantae
Phylum	Tracheophyta
Sub-phylum	Euphylophitina
Infra-phylum	Radiotopses
Class	Liliopsida
Sub-class	Commelinidae
Superorder	Zingiberane
Order	Zingiberales
Family	Costaceae
Sub-family	Asteroidae
Tribe	Coriopsidae
Genus	Costus

Cultivation:

Costus igneus is a robust and resilient species that can readily be grown under a variety of climatic conditions, particularly in tropical and subtropical areas. It thrives optimally in conditions that simulate its natural environment, which includes warm temperature, moderate to high humidity, and well-drained soil (Yadav et al., 2024). This flexibility has made it more popular, not just in the conventional medicinal gardens, but also as an ornamental plant in urban areas (Mathew & Varghese, 2019). The plant grows most luxuriantly in loamy or sandy-loam soils that are rich in organic matter, conducive to strong development. A desirable soil pH ranges from 6.0 to 7.5, tending towards slightly acidic to neutral (Kharmate et al., 2024). Although Costus igneus is tolerant of periodic dry spells, regular watering during periods of active growth enhances better development. Yet, waterlogging should be avoided under all circumstances because it makes the plant susceptible to root rot and fungal infections (Yadav et al., 2024). Vegetative propagation is most commonly done using stem cuttings and division of rhizomes. Stem cuttings, containing at least two nodes, are planted in moist soil under partial shade, where they root readily (Mathew & Varghese, 2019). Once established, plants grow rapidly, reaching heights of 2 to 3 feet within a few months. Under light demands, Costus igneus thrives under partial to full sunlight; partial shade tends to trigger the development of darker, more fragile leaves and promotes phytochemical diversity, which is advantageous for its medicinal properties (Kharmate et al., 2024). Mulching or intercropping with taller plants may provide shade as well as maintain soil moisture in heavily planted plants. In its first stage of growth, organic compost or farmyard manure is advisable. Fertilization with balanced NPK fertilizer could enhance foliage growth but should be carried out carefully to not dilute the concentration of secondary metabolites essential to the plant's medicinal value (Yadav et al., 2024). Leaf harvesting can begin around 3 to 4 months post-planting. Dissection of mature and healthy leaves promotes new shoot development. Regular pruning keeps the plant in shape and allows for better circulation of air inside the canopy, which minimizes the risk of pests and fungal diseases. While generally resistant to pests, Costus igneus can sometimes be attacked by aphids or mealybugs, particularly with poor drainage or overcrowding. As needed, use organic control measures or light chemical applications to adequately treat these problems (Mathew & Varghese, 2019). Thanks to its easy cultivation needs, fast growth rate, and ongoing leaf generation throughout the year, Costus igneus is suitable for small-scale home plots and large-scale medicinal farms alike. Growing this species upholds traditional health practices as well as opening up prospects for commercial applications in herbal medicine and wellness industries (Kharmate et al., 2024).

Traditional Uses:

In Ayurvedic practices, Costus Speciosus is used to treat Vata and Kapha doshas and improve complexion. It is used traditionally for managing cough, fever, dyspepsia, respiratory issues, and it also features prominently in the ‘‘Amber Mezhugu,’’ a formulation for rheumatic conditions (Kharmate et al., 2024;). The rhizome possesses anthelmintic, anticholinosterase, antifertility, anti-inflammatory, and antimicrobial actions due to its essential oils and steroidal saponins (Yadav et al., 2024). Costus pictus or the “insulin plant” possess stimulent, carminative, diuretic and antibacterial properties. From the leaves and rhizomes, diosgenin which is an anti diabetic steroid is present. Researches have proven that the aqueous leaf extract reduces blood glucose level in diabetic rats (Mathew & Varghese, 2019;). As for bronchitis, C. igneus root in Siddha medicine is used in powder, decoction, or oil forms for treatment. It is also used for intestinal worms, asthma, skin ailments, and even edema, hemorrhoids, and wheezing. Its therapeutic potential comes from phytochemicals like essential oils, inulin, and saussurine (Kharmate et al., 2024).

Regional Practices

In Ayurvedic medicine, Costus igneus is regarded as a therapeutic herb particularly effective in balancing the Vata and Kapha doshas. It is traditionally administered in formulations aimed at boosting metabolism, relieving inflammation, and increasing energy levels (Sharma & Dash, 2007). Due to its cooling nature, the plant is commonly used in Ayurvedic remedies for managing heat-induced conditions such as skin rashes, fever, and inflammatory discomfort. Its role in improving systemic balance has led to its inclusion in various polyherbal formulations.

In folk medicine across South and Southeast Asia, Costus igneus is widely used to treat digestive disorders, joint and muscular pain, and inflammatory ailments (Ravishankar et al., 2013). It is often administered in crude or semi-processed forms such as dried powder, fresh juice, or decoction. The plant's healing reputation makes it a preferred natural remedy for chronic issues such as gastritis, constipation, and musculoskeletal inflammation.

Documented Ethnopharmacological Studies

Ethnopharmacological research in recent years has validated several traditional uses of Costus igneus, particularly its antidiabetic effects. One significant study highlighted its ability to reduce blood glucose levels in diabetic models, supporting its ethnomedical use for managing diabetes (Pazhanichamy & Kallailingam, 2011). Additionally, anti-inflammatory and analgesic properties have been substantiated in experimental studies, linking traditional claims with modern pharmacological evidence (Ravindran et al., 2014). Further investigations have confirmed the antioxidant capabilities of Costus igneus, which are likely due to the presence of flavonoids and phenolic compounds. These antioxidants are believed to mitigate oxidative stress, a known contributor to diseases like atherosclerosis, Alzheimer's, and other neurodegenerative conditions (Ali et al., 2015). Moreover, studies have shown that extracts of the plant possess antibacterial and antifungal properties, supporting traditional use against infections (Baskar et al., 2012). However, despite these promising results, comprehensive clinical trials are still needed to validate these effects in human populations and determine appropriate dosages and safety profiles for wider medicinal use (Kumar et al., 2016).

Pharmacological activities

Antidiabetic Activity:

Diabetes mellitus is a chronic illness that affects a significant number of people worldwide. It is a severe, long-term illness that contributes significantly to global illness. Due to a complete or partial deficiency of the hormone insulin, this metabolic illness is characterized by hyperglycemia and disruptions in the metabolism of fat, protein, and carbohydrates. The development of micro and macrovascular complications of diabetes, which are the sources of morbidity and mortality, is also influenced by dislipidemia or hyperlipidemia. Since many plant products include bioactive compounds with therapeutic potential, there is considerable interest in evaluating them for the treatment of diabetes mellitus. Many researchers have recently assessed and determined the antidiabetic potential of Indian medicinal plants that have been utilized traditionally, conducting experiments on test animals (Dhanasekaran et al., 2014).

Anti-Inflammatory:

The term "anti-inflammatory activity" describes a substance's capacity to decrease swelling or inflammation. Many plant species have this characteristic, and Costus igneus is one of them. Research has shown that extracts from the leaves of Costus igneus have strong anti-inflammatory properties, and tests of different solvent fractions of its methanolic extract have shown that some fractions are especially good at reducing inflammation. These plant-based extracts have shown promise in blocking important mediators and enzymes that contribute to inflammation, including nitric oxide synthase (NOS), myeloperoxidase (MPO), lipoxygenase (LOX), and cyclooxygenase (COX). In addition, specific phytocompounds like β-amyrin, isolated from Costus igneus, have shown promising anti-inflammatory effects in both in vivo and in vitro models. These findings support the traditional use of Costus igneus in treating inflammatory conditions and suggest its potential as a natural source for anti-inflammatory agents (Adiga et al., 2014).

Antioxidant:

Antioxidant-rich foods can help prevent inflammation, cancer, neurological illnesses, cardiovascular disease, and other issues brought on by aging of the skin and cells. Through scavenging free radicals, preventing lipid peroxidation, and other processes, antioxidants provide protection against oxidative stress. Natural plant substances called antioxidants protect the body from harmful substances called free radicals. They accomplish this by helping to stop oxidation, which can kill cells and perhaps hasten the aging process. The antioxidant activity of the leaves, stem, and rhizome of Costus igneus is compared. The leaf had the lowest antioxidant activity when compared to the stem and rhizome. Rhizome antioxidant activity reached a maximum of 89%, compared to 72% for leaves and approximately 55% for stems that showed antioxidant activity. The costus igneus acetone extract had the most potent cytotoxic effect on the cancer cell line (MCF-7) when compared to the positive control doxorubicin. Costus igneus can be utilized in biological applications in a safe and efficient manner. Additionally, it possesses significant cytotoxic properties (Chacko et al., 2018; Kaloori & Margaret, 2022).

Hypolipidemic Activity:
Costus igneus has demonstrated encouraging hypolipidemic effects, especially in diabetic experimental animals. Because they lower high levels of triglycerides, total cholesterol, low-density lipoproteins (LDL), and very low-density lipoproteins (VLDL), extracts from its rhizome have been shown to assist control lipid metabolism. Additionally, the extract promotes elevated levels of high-density lipoprotein (HDL), which is advantageous for cardiovascular health. These outcomes imply that the costus igneus may aid in the treatment of lipid problems that are frequently linked to metabolic diseases such as diabetes. It may be used as a supportive medication to reduce hyperlipidemia and lower the risk of cardiovascular problems because its natural components seem to enhance lipid profiles (Kalailingam et al., 2011).

Hepatoprotective Activity:
Significant hepatoprotective activity has been demonstrated by costus igneus, especially in diabetic circumstances. By lowering high levels of hepatic enzymes, which are often indicators of liver damage, extracts from the plant's rhizome have been shown to aid in the restoration of normal liver function. Apart from controlling liver enzymes, Costus igneus also seems to have a beneficial effect on the metabolism of carbohydrates and increase the activity of antioxidant enzymes, which helps to protect the liver as a whole. According to these results, the plant may enhance liver health and lessen liver damage, particularly in metabolic diseases like diabetes (Kalailingam et al., 2011).

Antimicrobial activity:
Significant antibacterial activity against a wide range of microorganisms has been shown by Costus igneus. The growth of gram-positive and gram-negative bacterial strains as well as several fungal species has been effectively inhibited by extracts from different portions of the plant, especially the methanolic and ethanolic extracts. The plant's bioactive chemicals, which have the ability to alter microbial cell structures and prevent their metabolic processes, are responsible for these antibacterial qualities. As a natural source of antimicrobial compounds for treating infections and promoting general health, the plant's extracts have demonstrated promise in the fight against common pathogens (Gothandam et al., 2010; Saraswathi et al., 2010).

Nephroprotective activity:
Costus igneus leaves have been shown to have nephroprotective activity, which protects the kidneys from injury induced by toxins, oxidative stress, and inflammation. The antioxidant constituents of the plant, flavonoids, and phenolics scavenge ROS, limiting oxidative damage to kidney tissues. In animal experiments, Costus igneus has been found to decrease serum creatinine and blood urea nitrogen (BUN) levels, markers of kidney injury. It also decreases inflammatory cytokines such as TNF-α and IL-6, suppressing renal inflammation. Costus igneus also aids in regeneration of injured kidney cells, promoting recovery from nephrotoxic damages (Kalailingam et al., 2011).

Background and context:
The employment of plants in medicine dates back to thousands of years as the base of ancient systems of healing like Ayurveda, Siddha, Unani, and Chinese medicine. In this extensive woven history of plant medicine, Costus igneus, commonly referred to as the Insulin Plant, has earned a prominent position, especially for its renowned use in controlling diabetes mellitus—a metabolic disorder of long-standing chronic nature which still poses significant global health issues. The ancient use of Costus igneus in India and surrounding areas has been extensively documented. Practitioners of folk medicine have traditionally suggested chewing fresh leaves daily to assist in lowering blood glucose levels. In certain rural areas, the leaves are also made into teas or added to dietary preparations for the purpose of ensuring healthy metabolic function. Outside of diabetes, common uses are for bronchitis, asthma, cutaneous infections, and kidney stones, and demonstrate its wider range of therapeutic benefits.

Nonetheless, although universally used traditionally, science only relatively recently started to validate the medicinal activity of Costus igneus. Initial phytochemical screening has proved the occurrence of bioactive molecules like flavonoids, alkaloids, steroids, glycosides, terpenoids, and saponins—compounds that have been proven to possess a variety of biological activities. Initial pharmacological screening has also shown promising antidiabetic, antioxidant, anti-inflammatory, and antimicrobial activities. However, current gaps in research exist. The majority of studies have been confined to primary pharmacological screening without venturing further into the chemical characterization, standardization,

or pharmacokinetic activity of the active constituents. Inadequate information on how these phytochemicals are absorbed, distributed, metabolized, and excreted in the human body hinders their integration into mainstream medicine. Further, various methods of extraction like aqueous, methanolic, and hydroalcoholic extractions can considerably affect the yield and bioactive composition. Solvent polarity is a key factor in selectively isolating certain types of phytochemicals, which in turn can influence both the therapeutic efficacy and safety profile of the plant extract. Thus, comparative studies of extracts made with various solvents are important for maximizing the medicinal application of Costus igneus.

Over the last few years, the progress of computational software like SwissADME has revolutionized herbal research and early drug discovery. SwissADME allows for the prediction of essential pharmacokinetic attributes (ADME: Absorption, Distribution, Metabolism, and Excretion) and drug-likeness of phytochemicals in silico, giving an idea of their possible behavior in the human body. Such tools fill the gap between rudimentary phytochemical screening and sophisticated pharmacological applications by assisting in the prioritization of promising compounds for further research. In this changing scientific scenario, the present research project assumes its place. Through phytochemical screening, determination of total phenolic content, and the use of SwissADME computer software for pharmacokinetic profiling, the present research systematically profiles the chemical and pharmacokinetic characteristics of aqueous, methanolic, and hydroalcoholic Costus igneus leaf extracts. In this way, it adds useful information to the increasing database confirming the therapeutic value of this ancient medicinal plant. This study stands at the crossroads of ancient wisdom and contemporary science, providing a route towards evidence-based application of Costus igneus in modern herbal medicine and potentially stimulating the creation of new plant-derived therapeutic agents for the treatment of metabolic and inflammatory disorders.

Research objectives:

- To evaluate the chemical parameters and perform qualitative phytochemical screening of Costus igneus leaf extracts prepared with different solvents (aqueous, methanolic, hydroalcoholic).
- To estimate the total phenolic content (TPC) of these extracts.
- To predict the pharmacokinetic properties (such as absorption, distribution, metabolism, and excretion profiles) of major phytoconstituents using SwissADME software.
- To compare the influence of extraction solvents on the phytochemical profile and pharmacokinetic behavior.

Significance of research:

The importance of this study is that it has the potential to fill the gap between conventional knowledge and contemporary science, especially in relation to the therapeutic uses of Costus igneus. Through the systematic examination of the chemical, pharmacological, and pharmacokinetic characteristics of the plant, the study seeks to legitimize its age-old use in traditional medicine as well as establish a scientific basis for its integration into conventional healthcare practices.

The application of Costus igneus in traditional medicine, especially in India, where it has been commonly referred to as the "Insulin Plant," is well known to have the ability to control diabetes mellitus. Nevertheless, there has been a lack of scientific support for such claims, and most of the studies conducted so far have been conducted on small populations or pilot studies. The goal of this study is to prove the therapeutic properties of Costus igneus using contemporary scientific methods like phytochemical screening, total phenolic content quantification, and pharmacokinetic studies. Through a comprehensive breakdown of the plant's bioactive molecules, this work will contribute to the emerging body of knowledge of its utilization and provide a basis for judging its efficacy and safety.

Among the important contributions of this study is the identification and characterization of phytochemicals found in various leaf extracts of Costus igneus (aqueous, methanolic, and hydroalcoholic). While previous studies identified bioactive compounds like flavonoids, terpenoids, and saponins, there has not yet been a study on the role and concentration of these compounds toward the therapeutic applications of the plant. This study will measure and compare the concentration of these compounds in various extracts, providing a better insight into which compounds are most likely to be responsible for the

pharmacological activity of the plant. This is critical to the standardization of extracts and the creation of effective herbal preparations.

One of the most important features of the study is the pharmacokinetic screening of the bioactive compounds discovered using SwissADME software. Although it is common with traditional medicine to utilize plants from experience, the pharmacokinetic characteristics (absorption, distribution, metabolism, and excretion) of the active ingredients have not been exhaustively studied in Costus igneus. By analyzing the ADME characteristics of such compounds, the study will provide important insights into their bioavailability, distribution to tissues, and drug-likeness potential, which are the key elements determining the potency of plant-derived treatments. All this will help inform the clinical application's ideal dosage, delivery route, and formulation, enhancing the scientific dependability of the plant's application.

With the universal prevalence of diabetes mellitus across the world, which has necessitated an increasing demand for more accessible and less expensive treatments, Costus igneus has come into focus for its age-old application in reducing blood sugar. This study is intended to verify its antidiabetic activity by assessing the phytochemicals involved in the control of blood glucose and their pharmacokinetics. Through investigations of the actions of the plant on biochemical pathways implicated in glucose homeostasis, the study may offer evidence for the establishment of complementary or alternative antidiabetic therapies of plant origin that are free from the side effects of synthetic drugs. Liver ailments like non-alcoholic fatty liver disease (NAFLD), cirrhosis, and hepatitis are significant health concerns, particularly among individuals with metabolic dysfunctions such as diabetes. There is increasing need for natural hepatoprotectors to prevent and treat liver injury. Earlier research has suggested that Costus igneus possesses the potential for hepatoprotection on the basis of its antioxidant and anti-inflammatory activities. This study will add credence to its hepatoprotective action at the scientific level, providing new hope for liver disease treatment. Through the evaluation of the liver-protective activity of the plant, this study can provide clinical application and a substitute for existing pharmaceutical drugs with potential side effects. The overall goal of this study is to establish a scientific foundation for the preparation of Costus igneus as a medicine. Through extensive analysis of the chemical constituents and pharmacokinetic profiles of the plant, the study will pave the way for drug design. The results will form the foundation for subsequent clinical trials and guide whether the active ingredients of Costus igneus can be included in herbal drug preparations for therapeutic purposes. This may enhance the availability and affordability of drugs, particularly in low-resource environments where liver disease and diabetes are common.

Scope and limitation

Scope:

The scope of this research is designed to provide a comprehensive assessment of Costus igneus by using a multi-faceted approach including phytochemical analysis, quantification of bioactive compounds, and pharmacokinetic profiling. The main purpose is to investigate the chemical profile and therapeutic application of various extracts (aqueous, methanolic, and hydroalcoholic) of leaves of Costus igneus.

In this study, qualitative and quantatative analysis shall be performed to detect and describe major phytochemicals in leaf extracts of the plant. Identification will be carried out employing standard procedures such as TLC and UV-Vis spectrophotometry. It will determine the total phenolic content in various extracts of plant. This will be measured by Folin- Ciocalteu method and will assist in identifying which extraction process gives the highest amounts of phenolic compounds, which may be responsible for the pharmacological activity of the plant. The research will also utilize SwissADME software for in-silico assessment of ADME properties. The software will forecast drug-likeness and bioavailability of compounds, providing information on their potential for clinical use. Computational method will yield theoretical information that can be used to direct additional experimentak research. The information obtained may have the potential to result in new therapeutic uses for Costus igneus, not just for diabetes but also for other chronic conditions where oxidative stress and inflammation are critical.

Limitations:

Pharmacokinetic information generated by the SwissADME software is based on predictive models and in silico calculations. Although SwissADME provides important information on the hypothetical pharmacokinetic characteristics of bioactive compounds, these data have to be experimentally validated in in vivo tests (animal models and human clinical trials) in order to absolutely confirm the bioavailability and metabolic fate of the compounds. Thus, the results must be taken cautiously until further experimental verification is established. This study is mainly concerned with phytochemical screening, quantification

of total phenolics, and pharmacokinetic estimation. Although these techniques yield critical information regarding the chemical makeup of the plant and its theoretical activity, in vivo experiments (e.g., animal tests) are required to confirm the true pharmacological activity in living organisms. This restriction limits the power to make absolute conclusions regarding the therapeutic effectiveness of Costus igneus in natural biological systems. The research concentrates on three popular extraction methods (aqueous, methanolic, and hydroalcoholic), but there are many other solvents and extraction methods that might provide varying profiles of bioactive compounds. The study does not investigate all possible extraction methods or dose-response correlations, restricting the applicability of its findings. Subsequent studies may investigate the effects of varying solvents and concentrations of the extracts on therapeutic effects. Changes in the purity and concentration of the leaf extracts analyzed may influence the results of the phytochemical and pharmacological evaluations. For instance, the bioactive compounds in the extracts may be influenced by conditions such as the season of harvesting, maturity of the plants, and location. One of the key challenges to herbal medicine development is the absence of regulatory standards for the dosage, preparation, and quality control of plant extracts. The lack of consistency in the phytochemical content of herbal extracts from the same species of plants causes difficulty in producing standardized and uniform therapeutic products. This study will have to address these concerns by studying methods of extract standardization and the safety and efficacy of the use of the plant in a clinical context.

II. LITERATURE REVIEW

Overview of literature

Costus igneus also known as the insulin plant, is prized for its medicinal value, particularly in the control of diabetes. Studies on its leaf extract have centered on qualitative phytochemical screening and phenolic content analysis, without resorting to animal experiments.

Traditionally used in Indian and Southeast Asian medicine, the leaves of *Costus igneus* are recognized for their anti-diabetic, anti-inflammatory, antioxidant, and antimicrobial properties (Patel et al., 2012). Scientific investigations have focused on phytochemical evaluations to identify the active compounds responsible for its medicinal benefits. Early phytochemical studies on *Costus igneus* leaves revealed the presence of alkaloids, flavonoids, terpenoids, glycosides, saponins, tannins, and phenolic compounds (Kameshwaran et al., 2014). These classes of compounds are widely known for their biological activities (Patel et al., 2012).

Extraction Methods:

Different methods of extraction are utilized to separate bioactive substances from leaves of Costus igneus:

- Methanolic extraction: Employing methanol as the solvent to effectively extract flavonoids, phenolics, and other polar compounds. It is generally used for its high phytochemical yields.
- Hydroalcoholic extraction: A solution of alcohol (methanol) and water is employed to extract a wider variety of phytochemicals, including both moderately non-polar and polar compounds, increasing the spectrum of bioactive constituents.
- Aqueous extraction: Involves the use of water as the solvent, conventionally used in herbal preparations and useful for extracting water-soluble compounds such as tannins, flavonoid glycosides, and certain alkaloids. (Kameshwaran, Vignesh, & Brindha, 2014)

Phytochemical Composition and Significance:

The leaf extract has significant bioactive molecules like flavonoids, tannins, alkaloids, steroids, and triterpenoids. These secondary metabolites are renowned for their varied pharmacological activities:

Flavonoids: are antioxidants with anti-diabetic, anti-inflammatory, and antimicrobial activities.

Tannins: are responsible for antioxidant and antimicrobial activity.

Alkaloids: have different therapeutic activities such as anti-inflammatory and analgesic activities.

Steroids and triterpenoids: are related to anti-inflammatory and anticancer activities.

Reducing sugars: could be involved in energy metabolism and bioactivity improvement. (Kameshwaran, Vignesh, & Brindha, 2014) (Patel et al., 2012)

Phenolic Content:

Phenolic compounds, particularly flavonoids such as quercetin, are significant contributors to Costus igneus pharmacological activities. Quantitative analysis indicates substantial flavonoid content, which is related to antioxidant and anti-diabetic activity. Total flavonoid content is usually quoted as quercetin equivalents ranging around 3 mg/g of leaf extract (Patel et al., 2012).

Pharmacological Activity Prediction Using SwissADMET:

SwissADMET software is used to predict the pharmacokinetic and toxicity profiles of phytochemicals from Costus igneus. This computational tool evaluates drug-likeness and ADMET (Absorption, Distribution, Metabolism, Excretion, Toxicity) properties, confirming that the major compounds, especially flavonoids, have favorable profiles with no violations of Lipinski's rule of five. This supports their potential as safe and effective therapeutic agents (Patel et al., 2012).

Theoretical framework

The conceptual basis of this research is the phytochemical-pharmacokineticpharmacodynamic (PPPD) model, which assumes that the effectiveness of a plant-based therapeutic compound is not only dependent upon its chemical structure but also upon the way such constituents are metabolized by the body. The PPPD model places significant emphasis on the combination of phytochemical determination with pharmacokinetic simulation to elucidate the therapeutic value and safety profile of plant extracts. (Liu et al., 2013).Pharmacognosy is centered on the extraction, identification, and characterization of natural sources of bioactive compounds, whereas pharmacokinetics is concerned with the behavior of these compounds within a biological system. It is the synthesis of both that enables a holistic perception of the therapeutic potential and usable applicability of herbal drugs. (Li & Vederas, 2009)

Additionally, the model is underpinned by the idea of biopharmaceutics, which bridges the physicochemical properties of bioactive compounds with their biological action. Pharmacokinetic profiling of phytochemicals provides knowledge on their absorption efficacy, distribution in tissues, metabolic conversion, and ultimate excretion, which are all important to their clinical utility and safety. SwissADME software predicts ADME properties from the molecular structure, thereby facilitating estimation of oral bioavailability, blood-brain barrier permeability, and metabolic stability. It is this multidisciplinary approach that facilitates rational drug design and formulation development from natural sources. (Daina et al., 2017). Phytochemistry is the bottom layer of the framework, and it deals with the identification, extraction, isolation, and structural elucidation of bioactive constituents from plant material. It is grounded on the premise that secondary metabolites like flavonoids, alkaloids, tannins, saponins, and terpenoids are accountable for the medicinal characteristics of herbal plants. Theoretical techniques from analytical chemistry, including UV Vis Spectroscopy, are used to analyze and quantify these constituents. This assists to establish the Chemical fingerprint of Costus igneus leaf extract, major and minor phytoconstituents concentration and physiochemical properties of these compound which affects the biological acitivity and bioavailability. (Harborne, 1998).Pharmacokinetics is a pharmacology subdiscipline dealing with the transfer of drugs within the body, usually explained in terms of the ADME model — Absorption, Distribution, Metabolism, and Excretion. The PPPD model highlights the importance of understanding how phytochemicals are metabolized in biological systems to allow for predictive therapeutic efficacy, dosage, and safety. In silico prediction tools such as SwissADME are utilized to estimate pharmacokinetic parameters such as oral bioavilability, blood brain barrier, etc. It is significant to know the pharmacokinetic profile of active ingredients in order to authenticate their curative value outside normal indications. (Daina et al., 2017).

Theoretical framework also involves herb-drug interactions and ethnopharmacology, both of which is needed to understand the entire therapeutic richness of Costus igneus. Herbdrug interactions may be synergistic—in augmenting the therapeutic effect—or antagonistic, even lowering the efficacy or eliciting undesirable effects. This level of complexity demands a multiscale theoretical strategy that involves biochemical, cellular, and systemic levels of investigation in order to reliably forecast pharmacokinetics and pharmacodynamics. In addition, herb-drug interactions are an important consideration, especially when Costus igneus is incorporated in polyherbal preparations or combined with traditional pharmaceutical drugs. (Izzo & Ernst, 2009). Thus, it is essential to understand these interactions to ensure patient safety and maximize therapeutic efficacy. From an ethnopharmacological point of view, the ethnomedical use of Costus igneus as an anti-diabetic and antiinflammatory agent offers useful empirical information that can inform scientific investigation. (Izzo & Ernst, 2009) The ethnomedical context—how

the plant has been traditionally prepared, administered, and viewed by different cultural groups—provides clues to its possible uses and safety profile. Combining this traditional knowledge with contemporary pharmacological validation not only respects indigenous practices but also facilitates a more holistic, culturally appropriate approach to drug discovery and development. This combined framework ensures that the scientific inquiry is still rooted in both biological plausibility and historical relevance. Lastly, this framework serves the larger theoretical objective of rational development of herbal drugs, wherein plant extract standardization and in vivo behavior prediction are central to the shift from traditional medicine to clinically licensed phytopharmaceuticals. By connecting chemical profiling to pharmacokinetic modeling, the framework directs: identification of lead compounds, optimization of the dosage form, safety and efficacy evaluations for regulatory approval. (Williamson, 2001).

Gaps in literature

Despite the growing interest and promising pharmacological attributes associated with *Costus igneus*, current research remains fragmented and incomplete in several critical areas. Addressing these gaps is essential for establishing the plant's therapeutic efficacy, safety, and regulatory acceptance.

Perhaps the most important void is the dearth of in-depth pharmacokinetic research with regard to Costus igneus' active components. Although various studies have analyzed the plant's pharmacological actions—especially its antidiabetic, antioxidant, and anti-inflammatory properties—relatively few have investigated how the compounds interact once they are administered into a biological system. Absorption rates, tissue distribution, metabolic routes, bioavailability, half-life, and excretion profiles are mainly unknown. Without such data, it is not possible to tailor dosage regimens or assess the clinical efficacy and safety of the plant's bioactive phytochemicals.

Another significant limitation of the literature is the lack of standardized procedures for the preparation, extraction, and phytochemical characterization of C. igneus leaf extracts. Various studies usually employ different solvents, extraction processes (e.g., cold maceration, Soxhlet), and quantification methods, leading to variably composed phytochemical profiles. This variation hampers the comparison of results between studies and slows the development of a homogenous therapeutic product. Standardization is important for guaranteeing reproducibility, quality control, and clinical significance.

While numerous investigations have been able to identify principal phytochemicals present in Costus igneus, including flavonoids, alkaloids, saponins, and terpenoids, few studies provide supportive evidence for specific compounds eliciting observed pharmacological actions. Mechanisms of action for most of these compounds are speculative, commonly inferred by analogy from the known properties of similar phytochemicals without experimental proof. In addition, few studies employ pharmacokinetic or molecular methodology to elucidate cause-and-effect relationships between isolated substances and biological activity. This undermines the scientific rigor and clinical utility of present results.

Another underexploited field is the convergence of laboratory-based experiments (in vitro and in vivo) and computational pharmacokinetic modeling (in silico). While valuable predictions from in silico tools like SwissADME and pkCSM about ADME properties are made, these tools are rarely coupled with experimental verification. This division restricts the usefulness of predictive models and the evolution of a broader understanding of how the compounds of the plant interact with biological systems.

Safety and toxicity evaluations of Costus igneus are still in short supply within the literature, especially with respect to long-term exposure and high-dose consumption. While the plant is currently considered safe owing to traditional application, formal toxicological studies—acute and chronic—are not plentiful. Key parameters like LD_{50} (lethal dose), genotoxicity, organ toxicity, and harmful drug interactions have not yet been extensively tested. This undermines the regulatory process of approval and erodes faith in its universal therapeutic application.

III.METHODOLOGY:

Research design:

Collection and Preparation of Raw Materials

1. Collection of plant material.

Fresh and at the peak of health leaves of Costus igneus (which is also known as Insulin Plant) were gathered from a cultivated setting. We had the plant material identified by a qualified botanist and we put a voucher specimen in the herbarium for future use (Ramu & Krishnan, 2015)

2. Cleaning and Drying:

The leaves we put through a process of washing which included use of tap water which was running and then we used distilled water to remove dirt and other impurities. We then put the cleaned leaves in the shade to dry at room temp (25–30°C) for 7-10 days. We monitored the drying process regularly and also turned the leaves periodically to avoid complete drying out and to also deter microbial growth (Preethi & Nirmala, 2020) .

3. Pulverization

After drying out completely the leaves were ground using a mechanical grinder. The powder was run through a 40-mesh sieve to obtain a uniform particle size which we then stored in clean airtight containers. The containers were labeled and put in a cool dry place away from sunlight until we needed them for use (Al-Awa, Sangor, Babili, Saud, Saleem, & Zaidi, 2023).

Extraction Process:

1. Preparation of extracts.

Methanol, hydroalcoholic solution (70% ethanol), and distilled water were used in cold maceration and decoction processes which are appropriate for lab scale studies.

2. Methanolic Extract:

100 g of Costus igneous leaf powder were put into a clean dry amber colored glass jar and macerated with 500 mL of analytical grade methanol. The mixture was sealed up and left at room temp for 72 hours with occasional shaking (2 3 times a day) to improve solvent penetration and phytoconstituent release. After 72 hours the mixture was filtered first through muslin cloth and then through Whatman No. 1 filter paper. The filtrate was concentrated on a water bath at 40 45°C until a semi solid mass was obtained. The extract was then air dried, weighted and stored in a tight container at 4°C (Rajesh et al., 2018) .

3. Hydromethanolic extraction:

Another 100g of leaf powder was macerated with 500 ml of 70% ethanol (methanol:distilled water=70:30v/v) for 72 hours in a closed glass conatiner. The mixture was stirred intermittently to ensure uniform extraction. After 3 days, the extract was filtered and concentrated in water bath at temperature not exceeding 50°C. The resulting semi-solid mass was dried completely and stored in an airtight amber glass container under refrigeration (Preethi & Nirmala, 2020) .

4. Aqueous Extraction:

For in the case of aqueous extraction we took 100 g of plant material which we boiled in 500 mL of distilled water for 30 minutes. The solution was allowed to cool to room temperature and then passed through muslin cloth and Whatman No. 1 filter paper. The filtrate was evaporated to dryness on a water bath. The dried extract was collected, weighted and stored in an airt tight container at 4 degrees C (Sudha et al., 2011).

Fig 2: Hydroalcoholic extract of Costus igneus

Fig 3: Different extracts of Costus igneus

Fig 4: Aqueous extract of Costus igneus

Phytochemical Screening:

Preliminary phytochemical screening was performed on methanolic, hydroalcoholic, and aqueous extracts using standard procedures (Harborne, 1998):

1. Steroids:

- Salkowski test: Mix the sample (e.g., plant extract) with chloroform. A standard ratio is 1 ml of the extract mixed with 2 ml of chloroform. Mix the sample (e.g., plant extract) with chloroform. A standard ratio is 1 ml of the extract mixed with 2 ml of chloroform. Look at the interface between the sulfuric acid and chloroform layers. The presence of a reddish-brown or reddish tinge at the interface shows a positive test, indicating the presence of sterols (Najafi, Nejad, Deokule, & Estakhr, 2010).

- Sulphur powder test: A pinch of sulphur powder is added to the extract.

- Libermaan burchard test: Dissolve a small quantity of the sample in an appropriate solvent such as dry chloroform. Add several drops of acetic anhydride to it. Add 2 drops of concentrated sulfuric acid (H2SO4). Mix the solution slowly. Check for color change (Thiwari, Kumar, Kaur, Kaur, & Kaur, 2011).

2. Alkaloids:

- Tannic acid test: A tannic acid solution of suitable concentration can be prepared. A few drops of the tannic acid solution can be added to 5 ml of the extract. Yellow crystalline precipitate is an indication of a positive result for alkaloids (Joanne et al., 2016)

- Dragendroff test: Add 1 ml Dragendroff reagent to 2 ml extract (Ajaiyeoba et al. 217).

3. Flavonoids:

- Alkaline reagent test: Two to three drops of sodium hydroxide were added to 2 mL of extract. Add few drops of HCl and observe (Chaudhary, Negi, & Dahiya, 2010)

- Lead acetate test: To a portion of the extract, add a few drops of a 10% lead acetate solution and observe (Sunderman, 1973).

4. Resins:

- Turbidity test: Mix extract in alcohol and add water test

5. Tannins:

- Ferric chloride test: Plant extract is dissolved in a solvent like water or a mixture of water and ethanol. A neutral ferric chloride solution is added to the sample extract *(Odebiyi & Sofowora, 1978)*

- Lead acetate test: Prepare a solution of the plant extract or sample in water. Add a few drops of lead acetate solution to the extract solutionObserve for the formation of a precipitate (Sunderman, 1973)

- Gelatin test: Dissolve gelatin in water to form a solution, often with a concentration of 1%. A small amount of sodium chloride (NaCl) is typically added to the gelatin solution. Add the sample containing the suspected tannins to the gelatin solution and observe (Tiwari et al. 2011)

6. Saponins:

- Froth test: A specified volume of distilled water is added to the extract. The mixture is shaken vigorously for a set period, typically 5-10 minutes. The mixture is allowed to stand, and the height and persistence of the foam are observed (Raval et al., 2009).

- Foam test: The extract is diluted with distilled water and vigorously shaken, usually in a graduated cylinder or test tube. The presence of stable foam, which persists for a certain period (e.g., 15 minutes), indicates the presence of saponins (Raval et al., 2009).

7. Phenols:

- Lead acetate test: Take a known amount of the plant extract or sample to be tested. Add a few drops or a specific amount of lead acetate solution to the sample in a test tube. Mix the solution gently and observe for the formation of a precipitate (Joanne et al., 2016).

- Ferric chloride test: Dissolve the compound to be tested in water or a mixture of water and ethanol. Slowly add a neutral ferric chloride solution, drop by drop, to the solution. Look for any color change *(Odebiyi & Sofowora, 1978)*

- Libermann's test: Add a small amount of sodium nitrite crystals to a clean, dry test tube. Introduce the phenol sample to the test tube. Gently heat the mixture for about 30-35 seconds and allow it to cool. Add 1 ml of concentrated sulfuric acid and shake the mixture gently. Note any color change in the solution. If a red color appears upon dilution, a phenolic group is likely present. Add a sodium hydroxide solution to the red solution and observe the color change again.

8. Terpenoids:

- Salkowski test: Dissolve the sample (e.g., a plant extract) in chloroform. A typical ratio is 1 ml of the extract dissolved in 2 ml of chloroform. Dissolve the sample (e.g., a plant extract) in chloroform. A typical ratio is 1 ml of the extract dissolved in 2 ml of chloroform. Observe the interface between the chloroform and sulfuric acid layers. A reddish-brown or reddish color at the interface indicates a positive result, confirming the presence of sterols.

- Libermaan burchard test: Dissolve a sample of the extract in chloroform. Add a few drops of acetic anhydride to the solution. Carefully add concentrated sulfuric acid along the sides of the test tube.

9. Glycosides:

- Keller killani test: Take 2 ml of the extract and add 1 ml of glacial acetic acid and a few drops of freshly prepared ferric chloride solution. Carefully pour the mixture along the sides of a test tube containing 1 ml of concentrated sulfuric acid. A brown ring formation at the interface between the two layers indicates a positive test for cardiac glycosides *(Yadav et al., 2011)*

- Legal test: Dissolve the glycoside or extract in pyridine. Add sodium hydroxide and sodium nitroprusside alternatively. A transient blood-red color will develop if the glycoside contains the characteristic unsaturated lactone ring (Evans (2009).

- Borntrager's test: Boil the powdered drug with dilute sulfuric acid, filter and add chloroform to the filtrate. Shake well and collect the organic layer. Add a few drops of strong ammonia solution, shake slightly, and keep the test tube aside for few minutes. The lower ammoniacal layer takes on a pink or red color.

10. Carbohydrates:

- Molisch's test: Add 2 ml of the test solution (the substance you are testing for carbohydrates) to a clean, dry test tube. Carefully add 2-3 drops of Molisch reagent (α-naphthol in ethanol) to the test tube. Hold the test tube at an angle. Slowly add concentrated sulfuric acid along the side of the test tube using a dropper or pipette, ensuring it forms a separate layer without mixing with the solution. Wait for a few minutes and observe the interface between the two layers (the acid layer and the solution). The formation of a purple or reddish-purple ring at the interface indicates a positive result for carbohydrates (Tiwari, 2015).

- Fehling's test: Mix equal parts of Fehling's solution A (copper sulfate) and Fehling's solution B (alkaline solution with Rochelle salt). Take a sample of the carbohydrate solution in a clean, dry test tube. Add the prepared Fehling's solution to the test tube containing the sample. Carefully heat the mixture in a water bath. Look for the formation of a red-brown precipitate, which indicates the presence of reducing sugars.

- Benedicts's test: Use a small amount of the sample. Pipette approximately 5 ml of Benedict's reagent into a test tube. Carefully heat the mixture over a burner or in a boiling water bath for a few minutes (e.g., 2-5 minutes). Shake the test tube gently during heating. Cool the test tube and observe the color change and any precipitate formation *(Simoni, Hill, & Vaughan, 2002)*

11. Proteins:

- Biuret test: Prepare an aqueous solution of the sample to be tested. Add Biuret reagent (copper sulfate and sodium hydroxide) to the sample solution. Gently mix the solution to ensure thorough interaction. Allow the mixture to stand at room temperature for 5-10 minutes. Observe for a color change. A violet or purple color indicates the presence of proteins (ipl.org).

- Xanthoproteic test: The procedure involves adding concentrated nitric acid to the sample, heating, and then adding a base (like sodium hydroxide). A positive result is indicated by a yellow to orange color change, confirming the presence of these aromatic amino acids (Nigam & Omkar, 2003).

- Millon's test: Take 1-2 ml of the protein solution in a test tube. Add 2-3 drops of Millon's reagent to the test tube. Gently shake the test tube to mix the solution (Tiwari, 2015; Nigam & Omkar, 2003).

12. Amino acids:

- Ninhydrin test: Prepare a solution of the test sample (solid, liquid, or gas) in water or ethanol. Prepare a 2% ninhydrin solution by dissolving 0.2g of ninhydrin in 10ml of acetone or ethanol. Add a few drops of the ninhydrin solution to the test compound. Heat the mixture in a warm water

bath for a few minutes. Observe for the development of a deep blue or purple color, indicating a positive test. Proline and hydroxyproline may produce a yellow color (Tiwari, 2015).

- Xanthoproteic test: The procedure involves adding concentrated nitric acid to the sample, heating, and then adding a base (like sodium hydroxide). A positive result is indicated by a yellow to orange color change, confirming the presence of these aromatic amino acids (Nigam & Omkar, 2003).

Table no. 2: Phytochemical screening of Costus igneus

Sr. no.	Phytoconstituents	Test name	Extraction Type	Observation
1.	Alkaloids	Dragendorff's Test	Methanolic	Orange ppt formation
		Tannic acid	Methanolic	Yellow crystalline precipitate indicates a positive result for alkaloids
2.	Flavanoids	Alkaline reagent test	Methanoli	Initially, a deep yellow colour appeared but it gradually became colourless by adding few drops of dilute HCL
		Lead acetate test	Methanolic	Yellow ppt formation
3.	Tannins	Ferric chloride test	Aqueous	Blue green colour in Hydroalcoholic extract and Brown color in aqueous extract
		Lead acetate test	Aqueous	Bulky white ppt formation in Hydroalcoholic extract and no ppt formation in aqueous extract
		Gelatin test	Aqueous	White ppt observed in hydroalcoholic extract and no ppt formation in aqueous extract.

4.	Saponins	Froth test	Aqueous	Persistent froth formation observed
		Foam test	Aqueous	Persistent foam observed
5.	Phenols	Lead acetate test	Hydroalcoholic	White ppt formation
		Ferric chloride test	Hydroalcoholic	Blue- green colour observed
		Libermann's test	hydroalcoholic	Complex forms a red-colored indophenol, which is a positive indication of the presence of a phenolic group.
6.	Steroids	Salkowski test	Methanoli	Separation of layers with dark yellow color on upper layer and reddish brown on lower layer
		Sulphur powder test	Methanolic	The ppt sink to the bottom.
		Libermaan burchard test	Methanolic	Bluish green formation in hydroalcoholic extract
7.	Terpenoids	Salkowski test	methanolic	Separation of layers with dark yellow color on upper layer and reddish brown on lower layer
		Libermaan burchard test	Methanolic	Dark green colour
8.	Glycosides	Keller killani test	Hydroalcoholic	Yellow color observed

		Legal test	Hydroalcoholic	Pinkish red color observed
		Borntrager's test	Hydroalcoholic	The lower ammoniacal layer takes on a pink or red color.
9.	Carbohydrates	Molisch's test	Aqueous	Reddish brown ring appeared
		Fehling's test	Aqueous	Formation of red precipitate and blue colored solution in control sample
		Benedict's test	Aqueous	Formation of red precipitate (in hydroalcoholic extract) and formation of green color (in aqueous extract)
10.	Protein	Biuret test	Aqueous	Formation of purple/violet ring
		Xanthoproteic test	Aqueous	Color change to orange
		Millon's test	Aqueous	Formation of red precipitate
11.	Amino acids	Ninhydrin test	Aqueous	Development of a deep blue
		Xanthoproteic test	Aqueous	Color change to yellow
12.	Resins	Turbidity test	Methanolic	Turbidity was notfound

Fig 5: Salkowski test

Fig 6: Foam and froth test

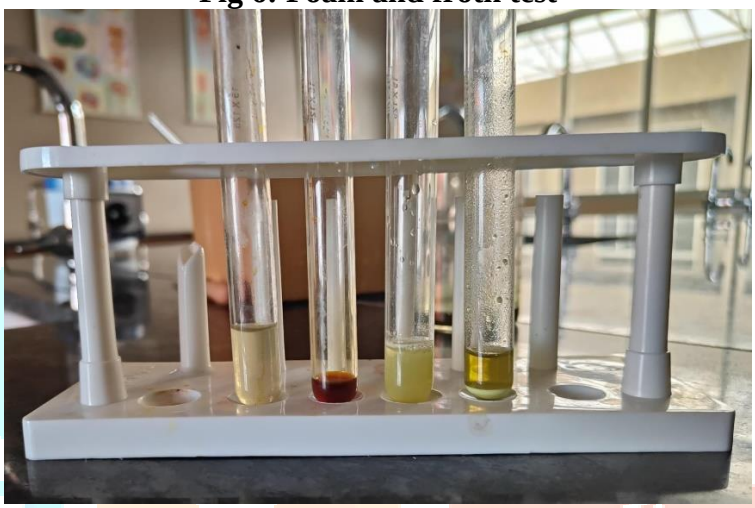

Fig 7: Lead acetate & Sulphur powder test

Fig 8:Fehling's test

Total Phenolic content:

Phenolic compounds are important bioactive molecules that drive antioxidant, anti-inflammatory, and antimicrobial activities in plants. Total phenolic content is a measure of phenolic compounds, useful for assessing the therapeutic potential and antioxidant capacity of plant extracts. TPC estimation can be used to standardize herbal formulations and helps provide establishing scientific evidence of traditional medicinal uses. The TPC estimation of a methanolic extract of Costus igneus in this study provides some insight into its possible health benefits.(Singleton et al., 1999)

Materials and Reagents:

- Methanolic extract of Costus igneus leaves
- Folin-Ciocalteu reagent (F-C reagent)
- 7.5% w/v sodium carbonate (Na_2CO_3) solution
- Gallic acid (standard phenolic compound)
- Methanol (analytical grade)

• Distilled water
• UV-Vis spectrophotometer
• Test tubes, pipettes, and volumetric flasks

Preparation of standard gallic acid solution:
Prepare a gallic acid stock solution (1 mg/mL) in methanol.
Dilute in order to obtain concentrations of 10, 20, 40, 60, 80, and 100 µg/mL.
Use these to prepare a calibration curve. (Waterhouse, 2002).

Procedure

Sample Preparation:
Prepare extraction of known amounts of Costus igneus methanolic extract (for example, 1 mg/mL) in methanol.
Reaction Set-Up (for Standards & Samples):
· Pipette 0.5 mL of the extract (or standard) into a test tube.
· To 0.5 mL of extract (or standard), add 2.5 mL of 10% (v/v) Folin–Ciocalteu reagent.
· At 5 minutes, add 2.0 mL of 7.5% sodium carbonate solution.
· Toast the sample well and leave it in the dark on the bench for 30 minutes. (Singleton et al., 1999)

Readings:
When all samples and standards are prepared, it is time to measure absorbance at 765 nm on the UV-Vis spectrophotometer using a blank (the blank prepared with the same solvents and methanol but no extract).

Calculations:
Simply plot a calibration curve of absorbance vs. concentration for gallic acid to apply the standard curve from the standards to the extract total phenolics determination.
The final results will be reported as mg of gallic acid equivalents (GAE) per gram of dry extract. (Singleton et al., 1999)

Calibration Curve
The absorbance values of the gallic acid standards were used to construct a calibration curve. The equation of the line obtained from linear regression was:
Absorbance = 0.00206 × Concentration + 0.1220
The coefficient of determination (R^2) was 0.7945, indicating a good linear relationship between absorbance and concentration.

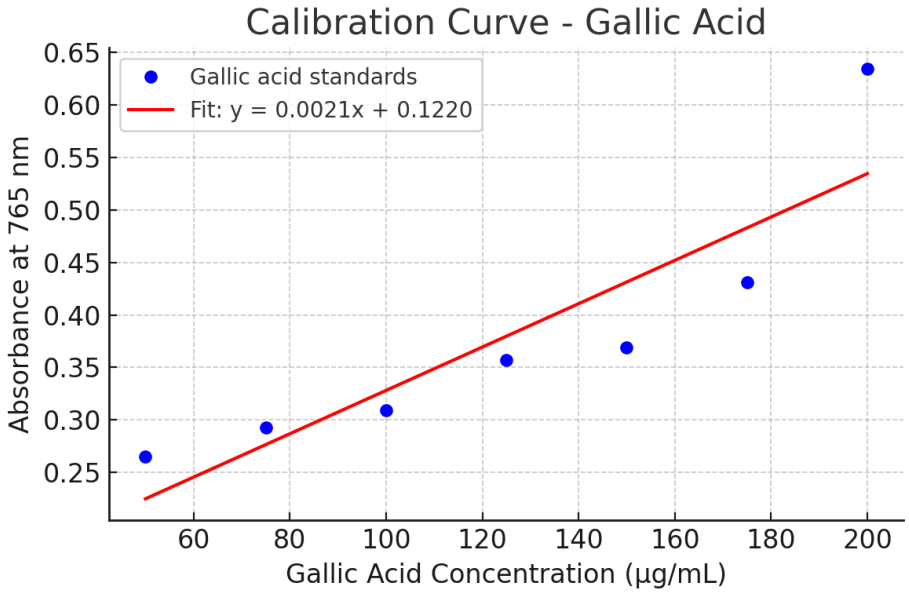

Fig no.9: Caliberation Curve of Gallic acid

Results and Calculation

The absorbance of the Costus igneus methanolic extract was found to be 0.400
Using the calibration equation:
Concentration (µg/mL) = (0.400 - 0.1220) / 0.0021≈ 132.38µg/mL
Converted to mg/mL: 0.1323mg/mL
Volume used = 0.5 mL → Total GAE = 0.1323 × 0.5 = 0.06615 mg
Mass of extract used = 0.05 g
TPC = 0.06615 mg / 0.05 g = 1.323mg GAE/g extract

Conclusion

The Total Phenolic Content (TPC) of the methanolic leaf extract of Costus igneus was determined to be approximately 1.32mg GAE/g, indicating the presence of significant phenolic compounds.[16]

SwissADME

SwissADME is a web-based, free computational tool established by the Swiss Institute of Bioinformatics (SIB) to assess the drug-likeness,
pharmacokinetic properties, and medicinal chemistry friendliness of small drug-like molecules. SwissADME is now an essential component in contemporary drug development and drug discovery procedures, especially during the initial stages of lead optimization and candidate screening (Daina et al., 2017). It enables inputs of molecular structure by SMILES (Simplified Molecular Input Line Entry System), MOL file, or free drawing and thereafter calculates a multitude of properties pertinant to the Absorption, Distribution, Metabolism, and Excretion (ADME) profile of the compound. Some of its most important features include:Lipophilicity (log P): SwissADME predicts log P with five alternative algorithms (iLOGP, XLOGP3, WLOGP, MLOGP, and SILICOS-IT), providing a consensus log P value, which is utilized to predict membrane permeability and solubility of the compound. Water solubility: Predicted with ESOL and other models, assisting in evaluation of oral bioavailability and formulation viability. Drug-likeness filters: The tool implements rules such as Lipinski's Rule of Five, Ghose, Veber, Egan, and Muegge filters to evaluate whether a compound possesses the physicochemical characteristics typical of orally active drugs in humans. Pharmacokinetics: SwissADME predicts important parameters such as gastrointestinal (GI) absorption, P-glycoprotein (P-gp) substrate likelihood, cytochrome P450 (CYP) isoenzyme inhibition (e.g., CYP3A4, CYP2D6), and skin permeability (log Kp). BOILED-Egg model: This simple graphical model assists in visualizing the potential for GI absorption and brain penetration by graphing the polarity and lipophilicity of compounds, which assists in the assessment of central nervous system (CNS) drug candidates. (Daina, Michielin, & Zoete, 2017).

Selection of Phytoconstituents
Major phytochemicals reported through literature review as well as initial screening, including diosgenin, β-sitosterol, and stigmasterol, were chosen for SwissADME analysis.

Procedure:
•Compound structures were retrieved in SMILES format from PubChem.
• SwissADME software (http://www.swissadme.ch/) has been used.
• SMILES codes were input into the SwissADME web portal.
• The following parameters were studied:
- Lipinski's Rule of Five
- Water solubility
- GI absorption
- Blood-brain barrier (BBB) permeability
- Bioavailability measurement

SwissADMET results and interpretation (Daina & Zoete, 2014; Daina & Zoete, 2016; Daina et al., 2017) :

1. Querectin

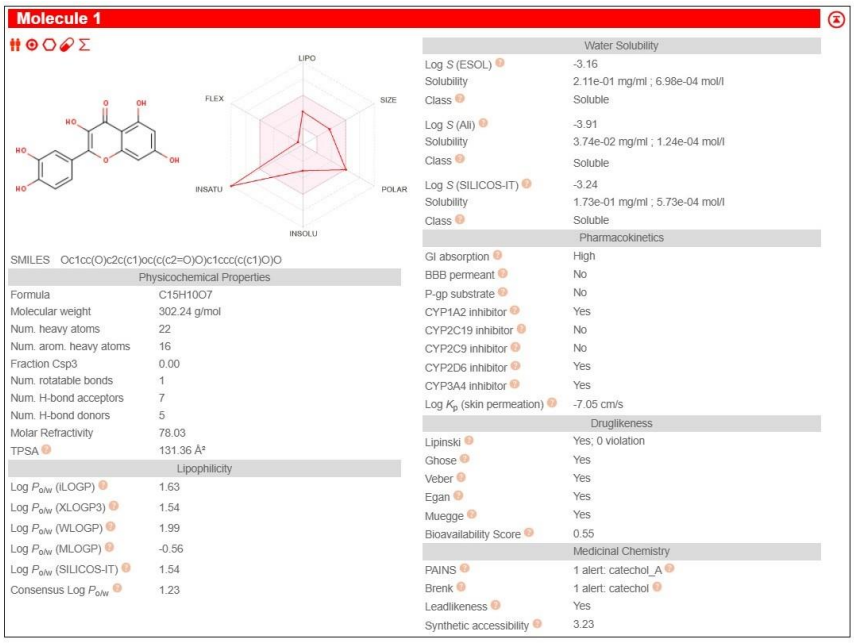

Fig. no 10: SwissADMET result of Querectin

The molecule, quercetin, has a molecular weight of 302.24 g/mol, in the range of optimal oral bioavailability. It has an optimum hydrophilic-lipophilic balance with an ideal LogP of consensus 1.23, in which there is optimum membrane permeability and solubility. Predictive SwissADME analysis shows good GI absorption and soluble behavior in all models to promote efficient systemic uptake, with the molecule exhibiting poor blood-brain barrier penetration, suggesting minimal involvement in the central nervous system. Drug-likeness analysis showed no violations of Lipinski, Ghose, Veber, Egan, and Muegge rules, confirming its potential as an orally active drug. However, inhibition of significant cytochrome P450 enzymes (CYP1A2, CYP2D6, and CYP3A4) was observed, which suggests a risk of a drug–drug interaction. A PAINS alert because of its catechol moiety suggests that experimental validation of the claimed biological activities is necessary. It possesses a moderate synthetic accessibility value of 3.23, which makes it relatively easy to synthesize. Structurally, the presence of multiple hydroxyl groups justifies its strong antioxidant activity. These pharmacokinetic and structural features correspond to the described pharmacologic activities of quercetin in Costus igneus including antioxidant, anti-inflammatory, anti-diabetic, cardioprotective, hepatoprotective, and anticancer. Despite its poor permeability through the BBB, quercetin may provide weak neuro protection supplemented by possible metabolic interactions that would enhance its bioactivity.

2. Kaempferol:

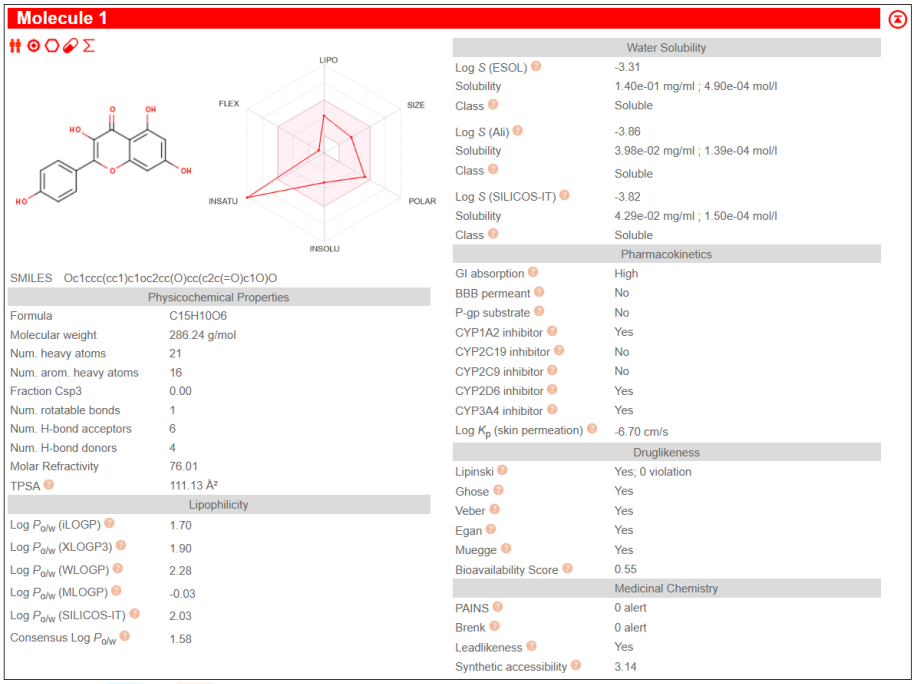

Fig. no. 11: SwissADMET result of Kaempferol

SwissADME kaempferol analysis, the principal flavonoid of Costus igneus leaf, displays positive drug-like and pharmacokinetic properties. Kaempferol is characterized by good gastrointestinal absorption and poor blood-brain barrier permeation, indicative of a peripheral-dominant mode of action. The compound has very good aqueous solubility, as confirmed by ESOL, Ali, and SILICOS-IT models, to permit efficient systemic distribution. Despite kaempferol being a strong inhibitor of the major cytochrome P450 enzymes (CYP1A2, CYP2D6, and CYP3A4), suggesting drug–drug interaction potential, it satisfies all the major drug-likeness requirements (Lipinski, Ghose, Veber, Egan, and Muegge) with a bioavailability factor of 0.55 and moderate ease of synthesis. Kaempferol exhibits strong pharmacological activities as an antioxidant, anti-inflammatory, antidiabetic, and anticancer compound. Mechanistically, it controls significant biological processes like inhibition of NF-κB activation to prevent inflammation and oxidative stress, activation of AMP-activated protein kinase (AMPK) to regulate glucose metabolism, and induction of mitochondria-mediated apoptosis in cancer cells. These characteristics highlight the therapeutic potential of kaempferol and further illustrate the medicinal significance of Costus igneus leaf extracts.

3. Diosgenin:

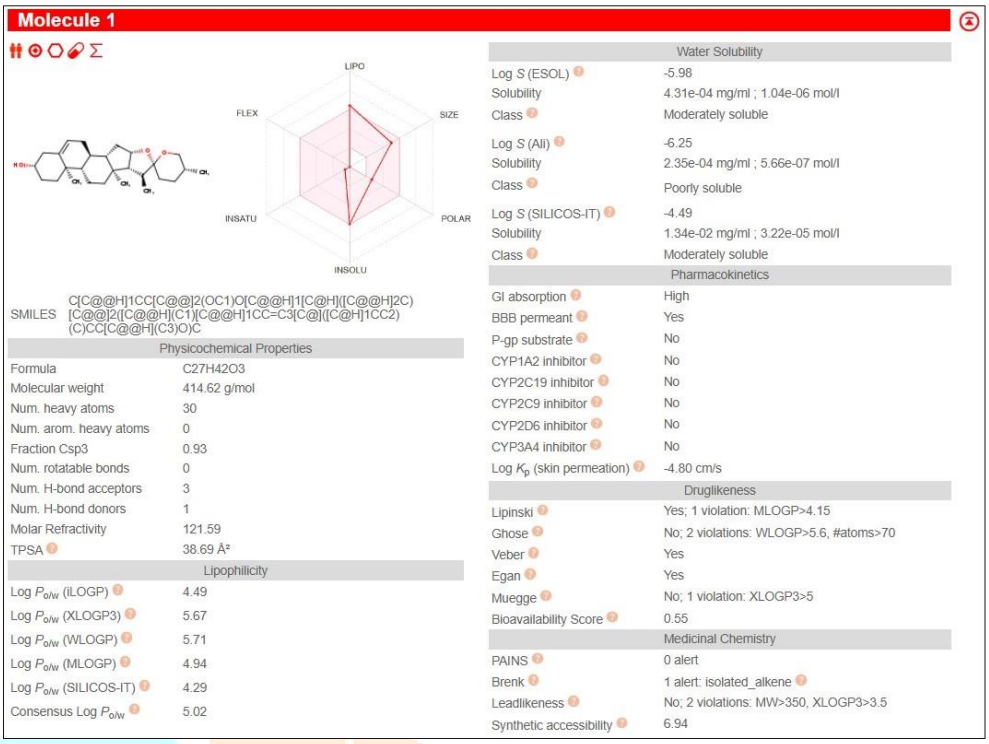

Fig. no. 12: SwissADMET result of Diosgenin

The pharmacokinetic and physicochemical profiling of Diosgenin, a major bioactive component in Costus igneus leaf extract, reveals promising drug-like properties. Diosgenin exhibits a molecular weight of 414.62 g/mol and a high degree of lipophilicity, with a consensus Log P value of 5.02, suggesting strong membrane permeability. Despite its high lipophilicity, its water solubility is moderate according to ESOL and SILICOS-IT models, although Ali's model classifies it as poorly soluble. The molecule has good gastrointestinal absorption and the ability to cross the blood-brain barrier, indicative of potential for systemic and CNS activity. Pharmacokinetic projections also confirm that Diosgenin is not a substrate for P-glycoprotein but does not inhibit key cytochrome P450 enzymes as well, reducing the risk of drug-drug interaction. From a drug-likeness aspect, Diosgenin moderately violates some filters like Lipinski and Ghose due to its strong lipophilicity, but possesses an adequate bioavailability score of 0.55. Medicinal chemistry assessment shows low toxicity risk, with no PAINS alerts and only one Brenk alert due to the presence of an isolated alkene group. Pharmacologically, Diosgenin is extensively recognized for its anti-inflammatory, anticancer, antidiabetic, and antioxidant activities. Its mechanisms of action are primarily the modulation of inflammatory pathways, inhibition of tumor growth through apoptosis induction, and enhancement of insulin sensitivity. Diosgenin targets several molecular targets including NF-κB, PI3K/Akt, and MAPK signaling pathways, and plays a critical role in modulating oxidative stress through enhancing antioxidant defense mechanisms. Its presence in Costus igneus is the reason for the traditional medicinal use of the plant in the therapy of metabolic diseases, particularly diabetes mellitus, and inflammatory diseases. Because of its favorable ADME properties and wide bioactivity spectrum, Diosgenin has the potential to be a lead compound for future drug development.

4. Corosolic Acid

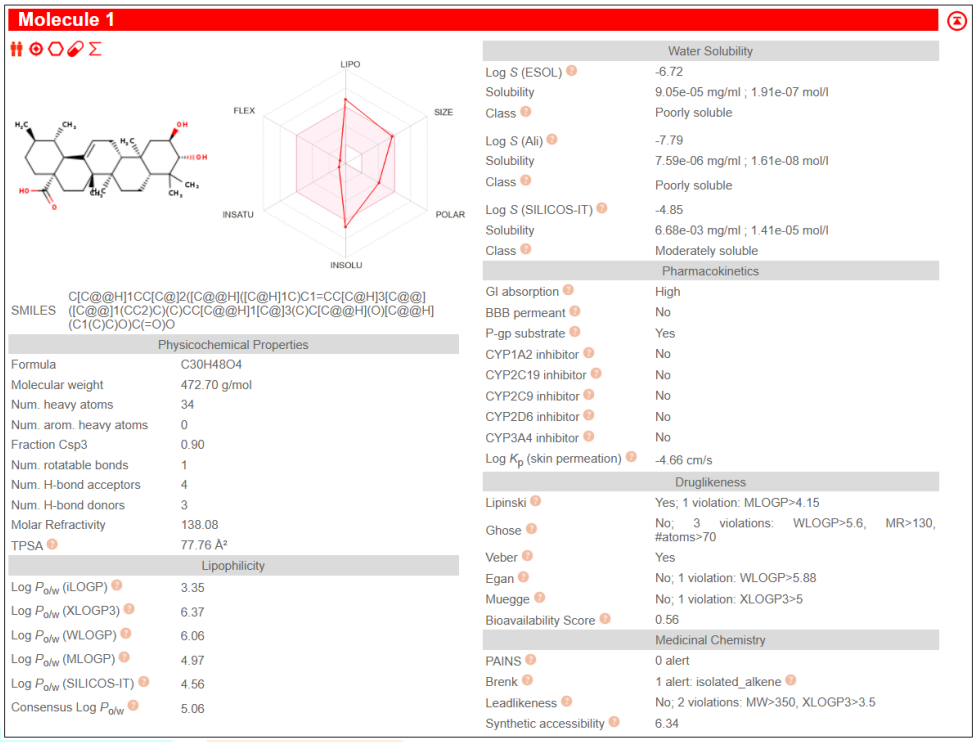

Fig. no. 13: SwissADMET result of Corosolic acid

SwissADME profiling of corosolic acid, a bioactive compound from Costus igneus leaf extract, offers valuable information on its pharmacokinetic and drug-likeness properties. Corosolic acid is strongly absorbed via the gastrointestinal tract but is not permeable through the blood-brain barrier, meaning it primarily targets peripheral tissues. It is weakly soluble in water, as indicated by its low ESOL and Ali solubility scores but moderately soluble according to the SILICOS-IT model. The molecule is a P-glycoprotein (P-gp) substrate, indicating active transport processes that could influence its bioavailability. Notably, it does not interact with major cytochrome P450 enzymes (CYP1A2, CYP2C19, CYP2C9, CYP2D6, or CYP3A4), lowering the chances of metabolic drug-drug interactions. In addition to one violation each in the Lipinski, Veber, and Muegge drug-likeness filters due to its comparatively high lipophilicity (Consensus Log P = 5.06), corosolic acid also possesses a moderate bioavailability score (0.56) and acceptable synthetic accessibility. Pharmacologically, corosolic acid is extremely well documented for its potent antidiabetic activity via enhancement of insulin sensitivity and enhancement of glucose tissue uptake. Mechanistically, it acts by activating the insulin receptor signaling cascade and initiating important downstream mediators such as the PI3K/Akt pathway, which plays a role in the translocation of GLUT4 to the cell surface. It also possesses anti-inflammatory, antioxidant, anticancer, and hepatoprotective activity, further contributing to its therapeutic value in the treatment of metabolic and chronic diseases. Together, these properties accentuate the pharmacological implications of corosolic acid as a plant-based and natural drug from Costus igneus.

5. Oleanoic Acid:

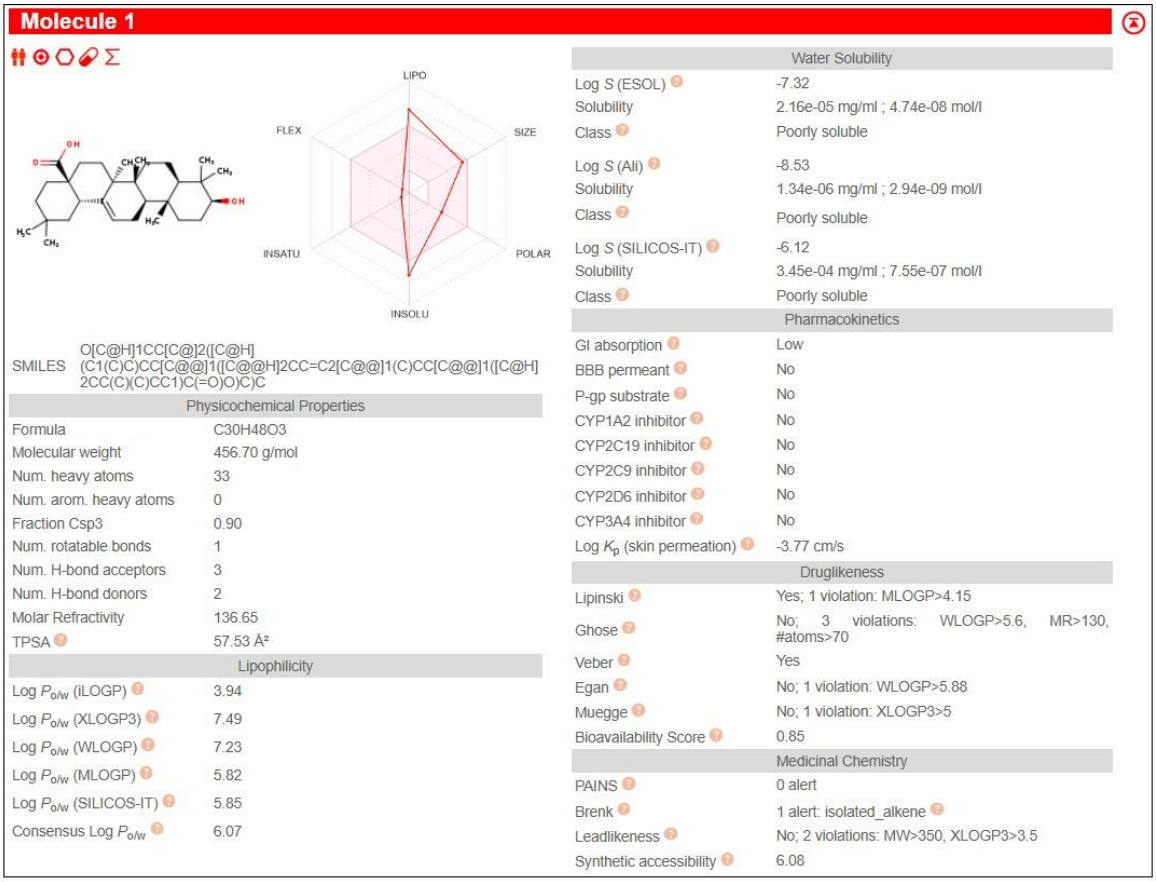

Fig. no. 14: SwissADMET result of Oleanoic Acid

The SwissADME prediction of oleanolic acid, a bioactive triterpenoid found in Costus igneus leaves, is important in elucidating its physicochemical and pharmacokinetic profile. While therapeutically very promising, oleanolic acid is poorly soluble in water as indicated by all three solubility prediction models (Log S ESOL, Ali, and SILICOS-IT), suggesting a problem with oral formulation. Its poor gastrointestinal absorption and nonpermeability through the blood-brain barrier suggest that its major effects would be peripherally acting, not centrally acting. Remarkably, oleanolic acid does not compete with P-glycoprotein as a substrate or inhibit major cytochrome P450 enzymes and hence reduces the risk of significant metabolic interactions with drugs. Although its lipophilicity (Consensus Log P = 6.07) is very high so that it breaks some drug-likeness filters (such as Ghose and Egan rules), the molecule still has an acceptable bioavailability score of 0.85. Pharmacologically, oleanolic acid exerts a wide range of positive effects such as hepatoprotective, anti-inflammatory, antioxidant, antidiabetic, and anticancer activity. Its mechanism of action is multifaceted: it regulates crucial signaling pathways such as inhibition of NF-κB for reducing inflammation, activation of Nrf2 for enhancing antioxidant enzyme production, increases the sensitivity of insulin by regulating the PI3K/Akt pathway, and induces cancer cell apoptosis by activating mitochondrial pathways. It also suppresses cell proliferation and angiogenesis and is responsible for its anticancer activity. These findings pinpoint the contribution of oleanolic acid to the therapeutic potential of Costus igneus leaf extract.

6. Rutin:

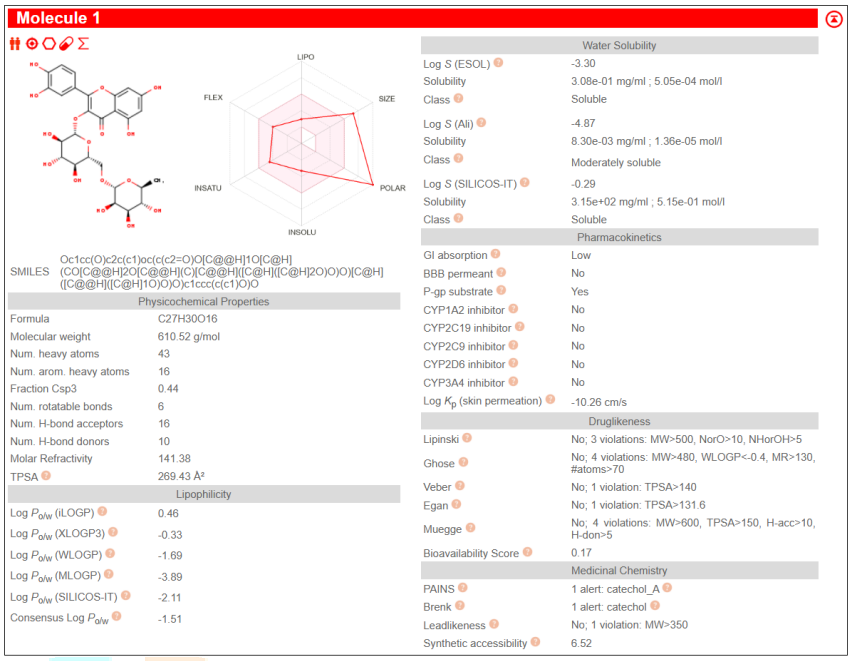

Fig. no. 15: SwissADMET result of Rutin

SwissADME prediction of rutin, a key flavonoid glycoside in the leaf of Costus igneus, is important in informing its pharmacokinetics and drug-like features. Rutin has acceptable solubility characteristics in all systems, but it is predicted to have poor gut absorption, and it does not cross the blood-brain barrier, meaning its activities are primarily restricted to peripheral systems. Moreover, it is a substrate for P-glycoprotein, indicating potential active efflux and reduced cellular uptake. Notably, rutin is not likely to inhibit major cytochrome P450 enzymes, and the drug-drug interaction potential needs to be viewed as low. Despite having a preferable solubility, the compound violates several drug-likeness rules such as Lipinski, Ghose, Veber, Egan, and Muegge filters primarily due to its too high molecular weight (610.52 g/mol), topological polar surface area (TPSA = 269.43 Å²), and high number of hydrogen bond donors and acceptors, making an acceptable value of bioavailability score equal to 0.17. Pharmacologically, rutin is well documented to possess potent antioxidant, anti-inflammatory, antidiabetic, neuroprotective, and cardioprotective effects. Mechanistically, rutin displays its antioxidant effects by direct scavenging of reactive oxygen species and augmentation of endogenous antioxidant defenses by activation of the Nrf2 signaling pathway. Its anti-inflammatory effect is primarily regulated by the inhibition of pro-inflammatory mediators such as TNF-α, IL-6, and NF-κB pathway inhibition. In diabetes treatment, rutin has been shown to increase glucose consumption by increasing insulin sensitivity and regulating the PI3K/Akt pathway. Moreover, its stabilization of endothelial function and prevention of platelet aggregation is responsible for its cardiovascular benefits. Such qualities make rutin a key factor in Costus igneus leaf extract's therapeutic value.

7. Caffeic Acid

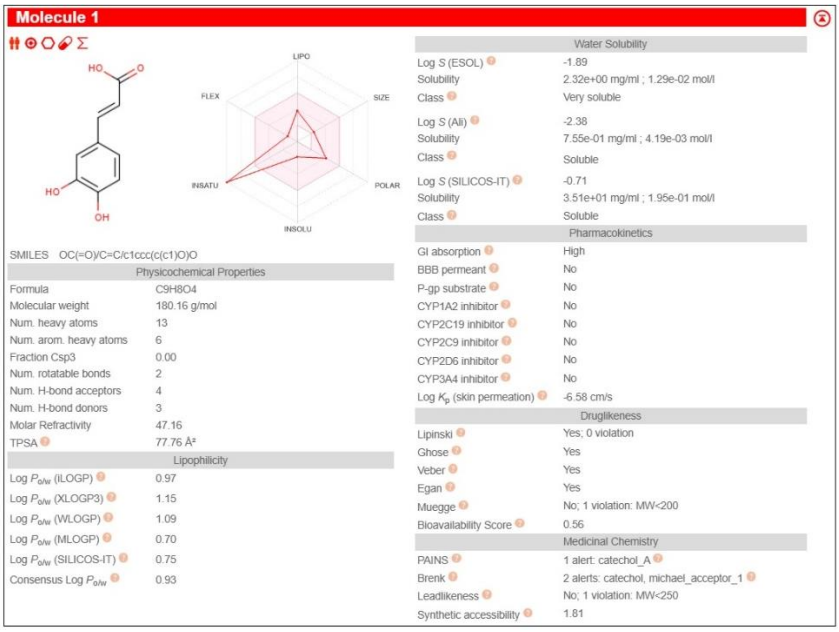

Fig. no. 16: SwissADMET result of Caffeic acid

The SwissADME prediction of caffeic acid, one of the major phenolic compounds of Costus igneus leaf, is impressive with regard to its drug-like features and pharmacokinetic profile. Caffeic acid is extremely water-soluble across all models and possesses excellent absorption in the gastrointestinal tract but poor permeation through the blood-brain barrier. It is neither a substrate for P-glycoprotein nor an inhibitor of important cytochrome P450 enzymes, which suggest low potential for metabolic interactions and good safety. The molecule is very compliant with various drug-likeness guidelines like Lipinski, Ghose, Veber, and Egan rules, but with only slight rule violation in the Muegge rule due to its slightly low molecular weight (180.16 g/mol). Its bioavailability rating of 0.56 is also in its favor as an orally effective compound. Pharmacologically, caffeic acid is valued for its powerful antioxidant, anti-inflammatory, antimicrobial, antidiabetic, and anticancer actions. Mechanistically, caffeic acid functions as a powerful free radical scavenger through the donation of hydrogen ions and the stabilization of reactive oxygen species. It displays its anti-inflammatory actions primarily by the downregulation of inflammatory cascades, including inhibition of NF-κB activation and inhibition of pro-inflammatory cytokines TNF-α and IL-1β. In diabetes, caffeic acid reinforces glucose metabolism by enhancing insulin signaling and oxidative stress alleviation in pancreatic tissues. Besides, its antimicrobial activity involves disruption of microbial membrane integrity, and its anticancer activity results from apoptosis induction and tumor cell proliferation inhibition by cell signal cascade modulation. These multifunctional therapeutic activities identify the importance of caffeic acid in contributing to medicinal significance of Costus igneus leaf extract.

8. Ellagic Acid:

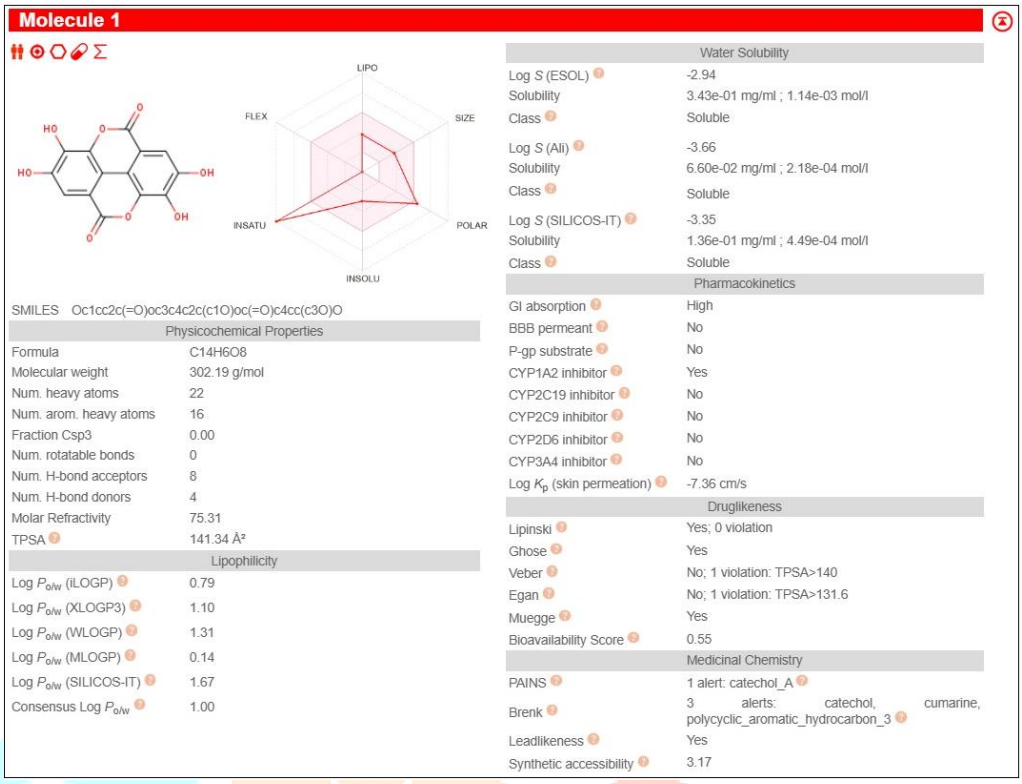

Fig. no. 17: SwissADMET result of Ellagic acid

Ellagic acid, one of the predominant phytochemical constituents of the leaf of Costus igneus, is promisingly drug-like in nature as indicated by its SwissADME screening. It is well soluble in all models and possesses high gastrointestinal absorption potential, although it cannot penetrate the blood-brain barrier. Ellagic acid neither acts as a substrate of P-glycoprotein nor inhibits most cytochrome P450 enzymes, with the lone exceptions of CYP1A2 and CYP2C19, and this is indicative of a relatively safe pharmacokinetic profile with minimal potential for significant drug-drug interactions. The molecule complies with Lipinski and Ghose drug-likeness criteria without any violations, although it has mild exceptions in Veber and Egan parameters due to the high topological polar surface area (141.34 Å² TPSA), which will have a mild effect on permeability. Ellagic acid is well recognized pharmacologically for its potent antioxidant, anti-inflammatory, antidiabetic, anticancer, and antimicrobial activity. Its effect is mostly a result of its free radical scavenging activity, which is responsible for the mitigation of oxidative stress through neutralization of reactive oxygen species. Ellagic acid also possesses anti-inflammatory effects by inhibiting NF-κB activation and downregulating pro-inflammatory mediators such as TNF-α and IL-6. In diabetic models, it increases insulin sensitivity and maintains normal glucose metabolism by reducing pancreatic β-cell oxidative damage. In addition, its anticancer effect is exerted through induction of apoptosis, angiogenesis inhibition, and suppression of metastasis in cancer cells through modulation of multiple cells signaling pathways. The broad-spectrum pharmacological activities of ellagic acid demonstrate its potential for therapy, which supports its role as an important bioactive compound of Costus igneus leaf extract.

9. Piperine:

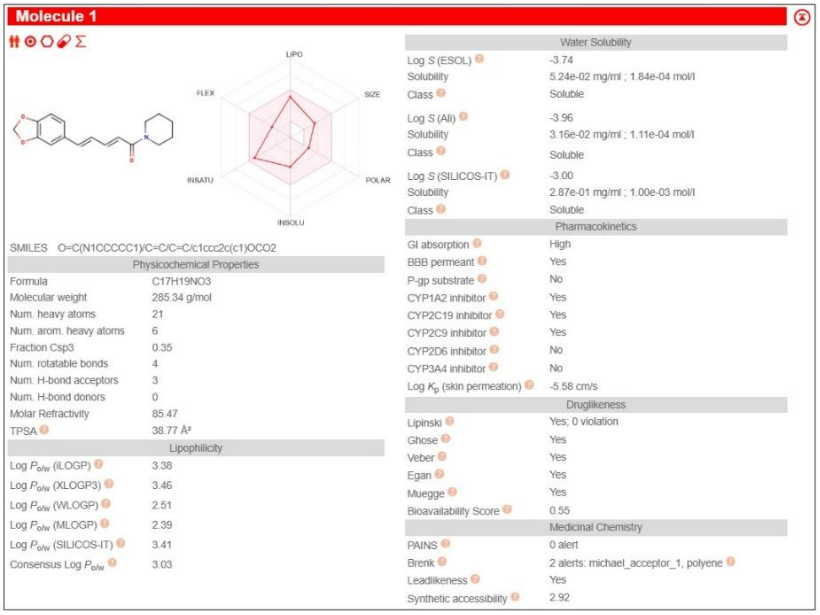

Fig. no. 18: SwissADMET result of Piperine

Piperine, another important phytoconstituent found in Costus igneus leaf extract, shows good drug-like properties as revealed by SwissADME prediction. It shows high gastrointestinal permeability and can cross the blood-brain barrier, demonstrating that it has potential for systemic and central nervous system action. Piperine shows good solubility in water and meets all the main drug-likeness rules such as Lipinski, Ghose, Veber, Egan, and Muegge, which shows great potential for oral bioavailability. Importantly, Piperine is also a cytochrome P450 enzymes CYP1A2 and CYP2C19 inhibitor that could affect the metabolism of co-administered drugs, a determinant of its pharmacokinetic interactions. Its pharmacological activities are broad, including antioxidant, anti-inflammatory, antimicrobial, anticancer, antidiabetic, and neuroprotective activities. Mechanistically, Piperine plays its antioxidant role by scavenging reactive oxygen species and inducing the activity of endogenous antioxidant enzymes such as catalase and superoxide dismutase. It displays anti-inflammatory activity by suppressing pro-inflammatory cytokines like TNF-α, IL-1β, and IL-6, mainly by downregulation of the NF-κB signaling pathway. As an antidiabetic agent, Piperine enhances insulin sensitivity and facilitates glucose uptake by modulating the insulin cascade signaling. It also displays bioenhancer activity by suppressing P-glycoprotein and other CYP enzymes, enhancing the bioavailability of other drugs. These multifaceted pharmacological actions highlight Piperine as a critical bioactive molecule contributing to the medicinal potential of Costus igneus leaves.

10. Cineole/ Eucalyptol

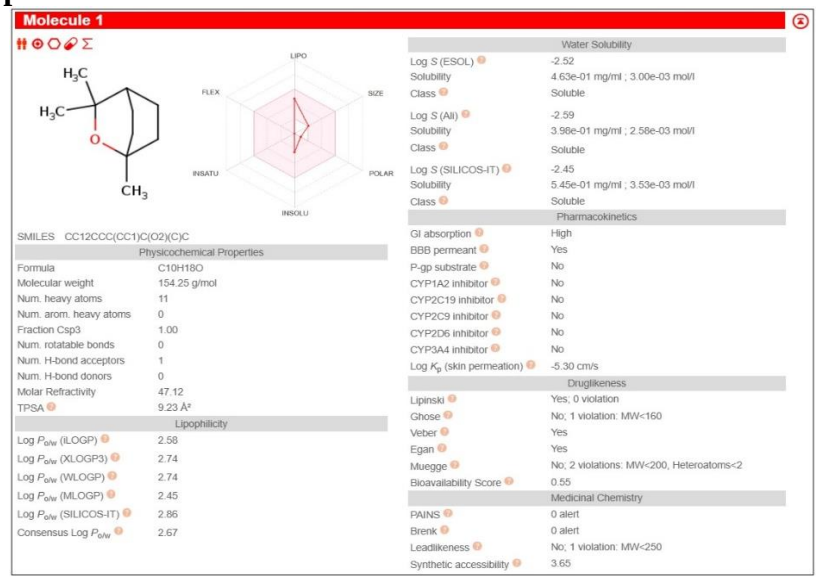

Fig. no. 19: SwissADMET result of Cineole

Cineole, or Eucalyptol, is another active bioconstituent present in the leaf extract of Costus igneus. SwissADME analysis indicates that Cineole possesses excellent gastrointestinal absorption and can cross the blood-brain barrier, indicating its central nervous system and systemic activity. It possesses very good solubility and adheres to important drug-likeness requirements, particularly Lipinski and Egan filters, but slight violation in Ghose and Muegge rules was noted due to its small molecular weight. The pharmacological effects of Cineole are extensive with profound anti-inflammatory, antimicrobial, mucolytic, bronchodilatory, and antioxidant effects. Mechanistically, Cineole exerts anti-inflammatory effect by modulating the expression of pro-inflammatory cytokines such as TNF-α and IL-1β through inhibition of NF-κB signal pathway. Its antimicrobial activity comes from disruption of bacterial cell membranes, leading to enhanced membrane permeability and ultimately cell death. Further, Cineole's antioxidant property encompasses removal of reactive oxygen species as well as defense against oxidative stress-mediated cellular injury. Its bronchodilation is achieved by airway smooth muscle relaxation, possibly via antagonism of calcium ion entry or regulation of cyclic AMP mechanisms. All these varied therapeutic potentials highlight the significance of Cineole as an extremely valuable pharmacologically active ingredient in the overall medicinal significance of Costus igneus leaves.

11. β- Sitosterol

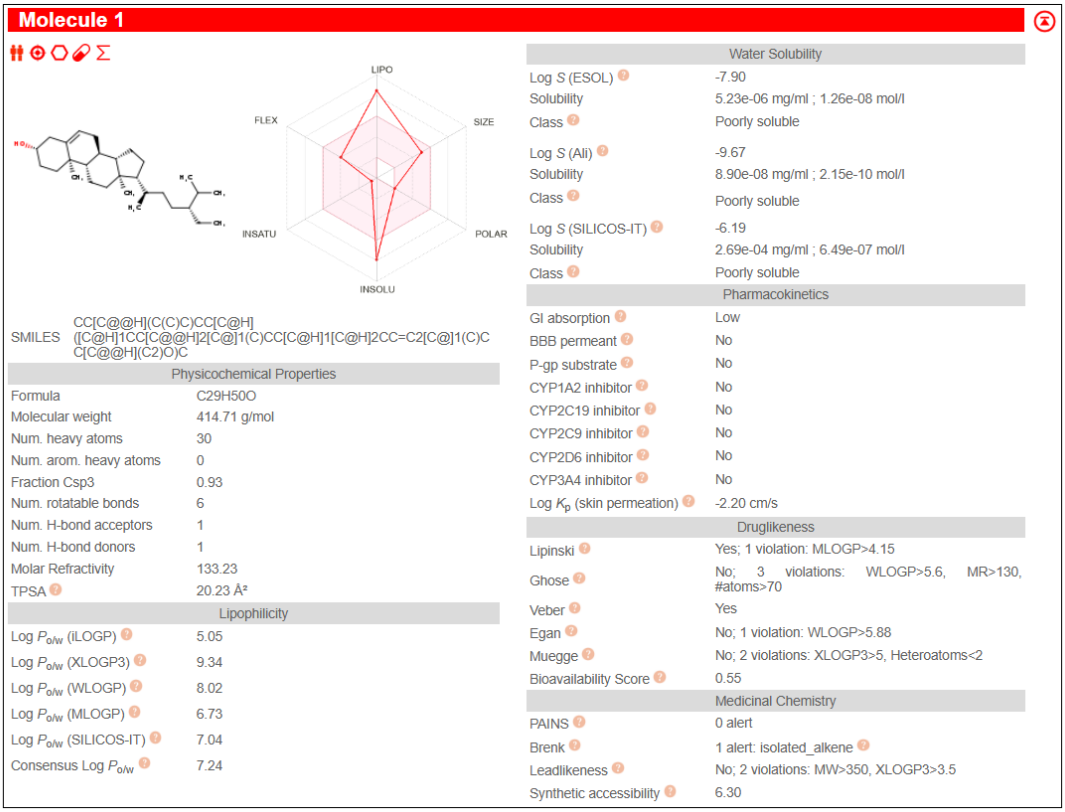

Fig. no. 20: SwissADMET result of β-sitosterol

β-sitosterol, the major bioactive component of methanolic leaf extract of Costus igneus, possesses low GI absorption that suggests compromised oral bioavailability except when formulated with lipid-based delivery systems. It is not a substrate for the P-glycoprotein and neither an inhibitor of significant cytochrome P450 enzymes (CYP1A2, CYP2C19, CYP2C9, CYP2D6, CYP3A4), which suggests minimal risk of drug–drug interactions and excellent safety in combination therapy. It also does not penetrate the blood–brain barrier, implying minimal CNS activity. Its poor skin permeability (log Kp = −2.20 cm/s) means minimal efficacy via the topical routes without the use of enhancers. The compound is not well water-soluble, as also predicted by three calculators (ESOL, Ali, and SILICOS-IT), which may limit its systemic absorption. However, as it is highly lipophilic (Consensus Log $P_o w = 7.24$), it can also efficiently engage with lipid bilayers and, thus, distribute into fatty tissues. While certain drug-likeness rules (Ghose, Muegge, and Leadlikeness) are being violated, β-sitosterol still has an acceptable score in bioavailability of 0.55, indicating good systemic promise under appropriate formulation conditions. Pharmacologically, β-sitosterol is accountable for the antidiabetic, anti-inflammatory, antioxidant, and hypocholesterolemic action of Costus igneus. It prevents cholesterol uptake from the intestine and possesses the ability to

regulate metabolic illness including obesity and diabetes. Anti-inflammatory activities of β-sitosterol arise as a result of COX-2 and pro-inflammatory cytokine inhibition, whereas the antioxidant activity arises from membrane stabilization and free radical neutralization. Overall, β-sitosterol is a key factor in Costus igneus for its therapeutic value, particularly against metabolic and inflammatory disease. Its pharmacokinetic limitations may be addressed by formulation strategies, and its lack of CYP inhibition makes it suitable for its inclusion in multi-herbal treatment regimens.

12. Ferulic acid

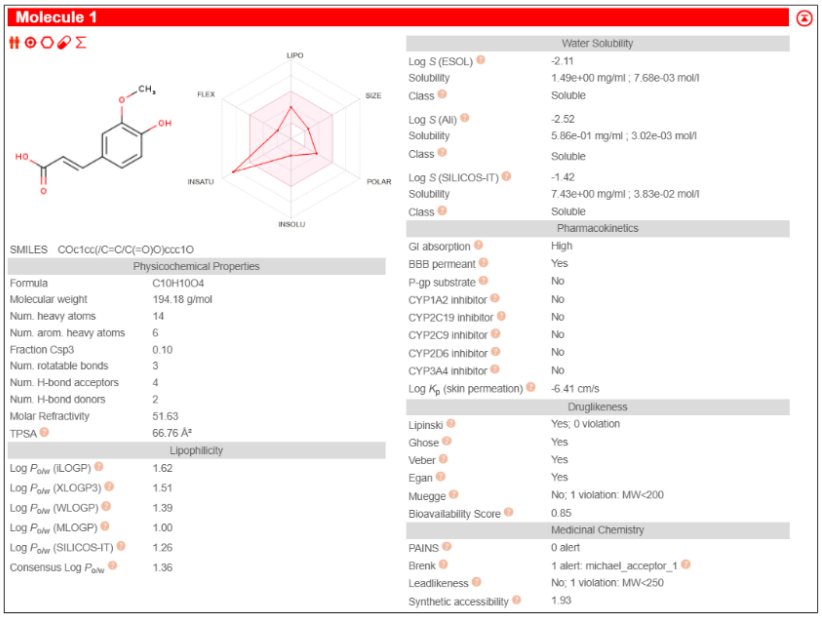

Fig. no. 21: SwissADMET result of Ferulic acid

Ferulic acid, a phenolic acid isolated from Costus igneus leaf extract, has favorable pharmacokinetic properties. It has good GI absorption, indicating good oral bioavailability, and is able to cross the BBB, indicating potential neuroprotection or CNS-associated activity. It is very water-soluble, as confirmed by ESOL, Ali, and SILICOS-IT models with predicted solubility values greater than 1 mg/mL. This results in good absorption and distribution in the biological system, as compared to weakly soluble lipophilic drugs. Ferulic acid is not a P-glycoprotein substrate nor an inhibitor of significant cytochrome P450 enzymes (CYP1A2, CYP2C19, CYP2C9, CYP2D6, or CYP3A4), therefore predictive of negligible drug–drug interaction potential and hepatic metabolic interference potential. It also has very low skin permeability (log Kp = −6.41 cm/s), which suggests that transdermal delivery would be ineffective in the absence of enhancers. On drug-likeness basis, ferulic acid obeys Lipinski, Ghose, and Veber rules and hence has good oral drug-like characteristics. Ferulic acid defies only one (low molecular weight) Muegge and Leadlikeness filters and possesses high bioavailability score 0.85, which supports its systemic therapeutic application potential further. It has low synthetic complexity (synthetic accessibility score: 1.93), Pharmacologically, ferulic acid is the primary contributor to the therapeutic activities of Costus igneus owing to its high antioxidant, anti-inflammatory, and antimicrobial activities. It scavenges free radicals, activates endogenous antioxidant enzymes, and inhibits pro-inflammatory mediators like TNF-α and IL-6. Its neuroprotective activity is considerable on the basis of BBB permeability, warranting use in cognitive enhancement or prevention of neurodegenerative illness. For example, in Costus igneus, ferulic acid underpins the traditional uses as treatment for metabolic and inflammatory disease, exerting systemic and potentially neuroprotective actions.

13. Lupeol

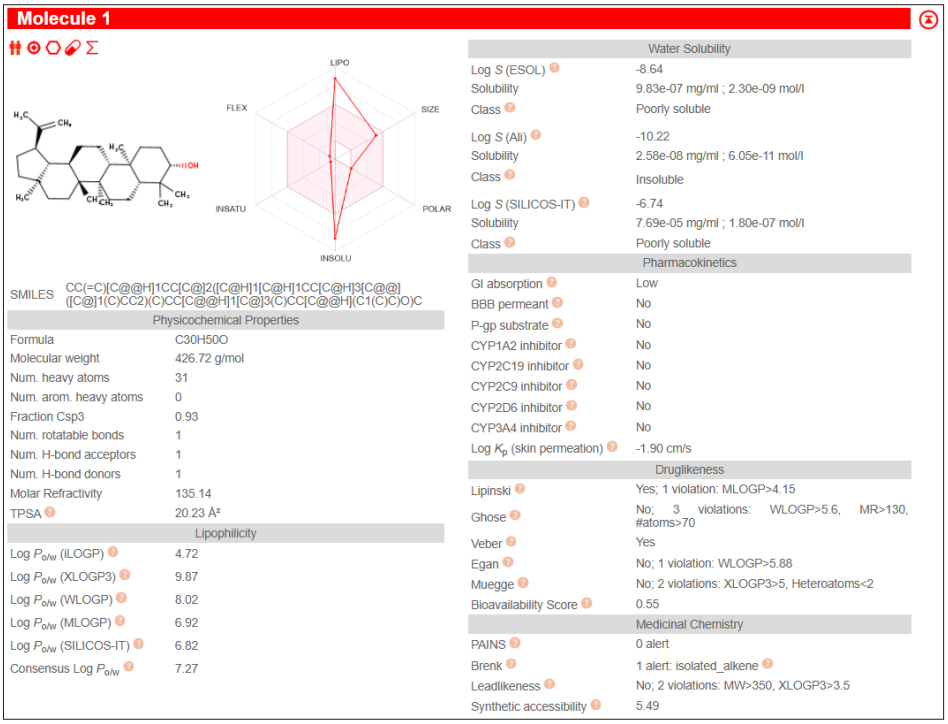

Fig. no. 22: SwissADMET result of Lupeol

Lupeol, a pentacyclic triterpenoid component of Costus igneus, is associated with poor GI absorption and non-BBB permeability, hence limited systemic and CNS activity in the absence of specialized delivery systems. Limited water solubility in all the predictive models (ESOL, Ali, SILICOS-IT) affects its oral bioavailability but corresponds to high lipophilicity (Consensus Log P_ow = 7.27). Its lipophilicity is high, suggesting preferential accumulation in lipid-rich tissues, where it might be more active in skin, adipose tissue, and membrane-rich organs. It is neither a P-glycoprotein substrate nor a moderate inhibitor of key cytochrome P450 enzymes, suggesting minimal risk of transporter-dependent efflux and drug–drug interaction. While it failed some of the drug-likeness filters (Ghose, Egan, Muegge, and Leadlikeness) due to high molecular weight, high log P, and low polarity, lupeol is given a moderate bioavailability score (0.55), meaning that formulation techniques could still make it pharmacologically active. It is of poor permeability in the skin (log Kp = −1.90 cm/s), meaning that dermal absorption would be limited in the absence of enhancers. Pharmacologically, lupeol possesses strong anti-inflammatory, anti-cancer, antimicrobial, and antioxidant activities. In Costus igneus, lupeol would account for the medicinal significance of the plant in treating inflammation, diabetes, and wound healing. It mediates key inflammatory pathways through the inhibition of enzymes such as COX and LOX, downregulation of pro-inflammatory cytokines, and inhibition of NF-κB signaling. Its antioxidant activity safeguards tissues from oxidative damage, while its lipophilicity facilitates membrane interaction and stabilization. Although systemic delivery is limited, localized and tissue-specific activity of lupeol makes it an important bioactive compound in Costus igneus, justifying its therapeutic activity in metabolic and inflammatory disorders.

14. Aspartic acid

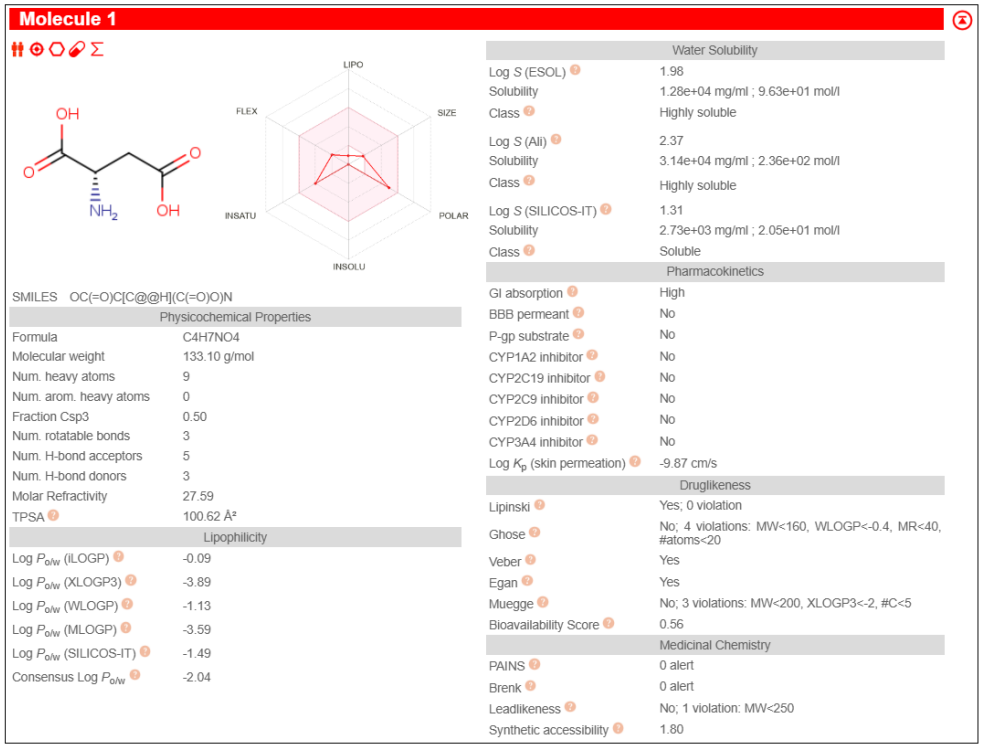

Fig. no.23: SwissADMET result of Aspartic acid

Aspartic acid, which is a naturally occurring amino acid of Costus igneus, has very favorable pharmacokinetic properties in terms of solubility and GI absorption. It is highly water-soluble in all the models of prediction (ESOL, Ali, SILICOS-IT) so that aqueous biological environments are effectively absorbed. It possesses a high GI absorption tendency, and therefore it can be administered orally, although its failure to cross the blood–brain barrier (BBB) indicates that it has no effects in the central nervous system (CNS). Neither is it a substrate for P-glycoprotein nor an inhibitor of major cytochrome P450 enzymes, hence significantly lowering the drug–drug interaction and hepatic metabolism possibility. However, its skin permeability is very low (log Kp = −9.87 cm/s), such that topical administration is not possible. Despite violating some drug-likeness filters such as Ghose and Muegge—mainly due to its low molecular weight, low lipophilicity (Consensus Log $P_o w$ = −2.04), and low structural complexity—it's synthetic accessibility score of 1.80 and bioavailability score of 0.56 show that it can easily be formulated for oral dosing. Pharmacologically, aspartic acid contributes to the overall therapeutic potential of Costus igneus by promoting cellular metabolism, neurotransmission (peripherally), and amino acid biosynthesis. It plays a key role in the urea cycle and energy production through the tricarboxylic acid (TCA) cycle. In the instance of Costus igneus, aspartic acid may enhance the plant's antidiabetic activity by supporting pancreatic β-cell function and glucose metabolism. Its role in detoxication processes and protein synthesis can also increase the antioxidant and tissue-healing activity of the plant. Even though it lacks anti-inflammatory and antimicrobial activity, it is a metabolic stimulant and contributes to the adaptogenic and recuperative activity of the plant.

15. Lysine

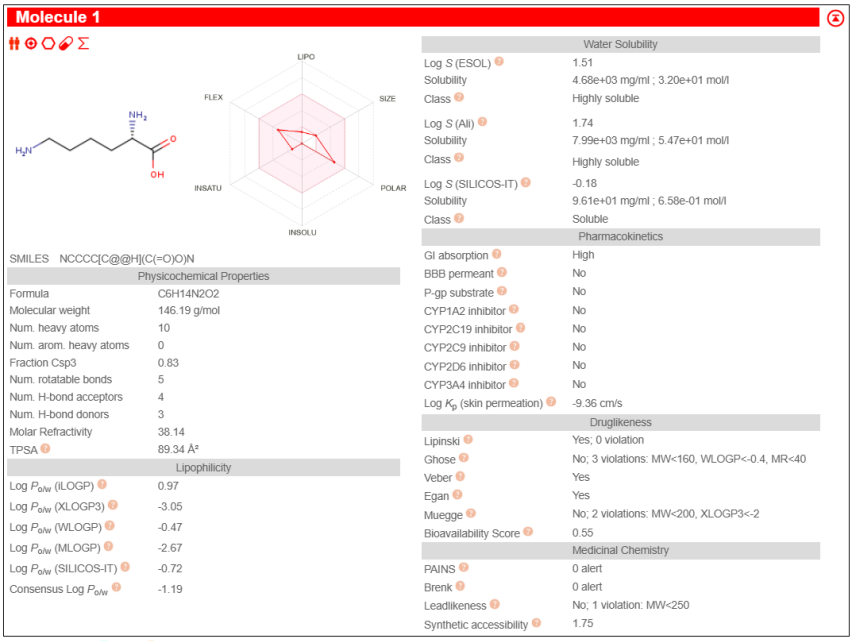

Fig. no. 24: SwissADMET result of Lysine

Lysine, being one of the primary amino acids in Costus igneus leaf extract, possesses favorable pharmacokinetic characteristics that justify its bioactive potential in the pharma profile of the plant. It is found to be extremely water-soluble in ESOL, Ali, and SILICOS-IT models, which indicates better solubility in body fluids. It is also very highly absorbed in the gastrointestinal (GI) tract, which indicates good oral bioavailability, even though it cannot penetrate the blood-brain barrier (BBB). Lysine is neither a strong inhibitor of any of the major cytochrome P450 enzymes (CYP1A2, CYP2C19, CYP2C9, CYP2D6, or CYP3A4), thus reducing the risk of metabolic drug interactions. The molecule is not a substrate for P-glycoprotein, further contributing to stable systemic availability. In spite of this, the permeability across skin is very low (log Kp = −9.36 cm/s), excluding topical efficacy. Lysine complies fully with Lipinski's rule, and although it violates Ghose and Muegge filters due to its small size and lower lipophilicity (Consensus Log P_ow = −1.19), it maintains a solid bioavailability score of 0.55 and a synthetic accessibility of 1.75, making it an easily incorporable compound in formulations. Pharmacologically, lysine contributes to several beneficial effects of Costus igneus. It plays a crucial role in protein biosynthesis, healing, and calcium absorption, hence contributing to the use of the plant in growth stimulation, healing, and bone health. Lysine has also been shown to contribute to immune function and exhibit antiviral activity against herpes simplex virus replication. In the case of Costus igneus, lysine may act synergistically with other phytoconstituents in modulating its antioxidant, anti-inflammatory, and possibly antidiabetic effects by stabilizing metabolic processes and suppressing oxidative stress.

IV.RESULTS AND DISCUSSION:

The phytochemical analysis of Costus igneus leaf extracts (aqueous, methanolic, and hydroalcoholic) identified the presence of various significant bioactive compounds. The positive reactions for steroids, alkaloids, flavonoids, tannins, saponins, phenols, terpenoids, glycosides, carbohydrates, proteins, and amino acids suggest that Costus igneus leaves contain a rich variety of phytoconstituents. These compounds have been reported to possess a vast range of pharmacological activities, such as antioxidant, anti-inflammatory, antidiabetic, hepatoprotective, and nephroprotective activities. The presence of flavonoids, phenol, and tannins indicates excellent antioxidant activity, whereas alkaloids and glycosides yield therapeutic effects such as antidiabetic and cardioprotective activity. The presence of steroids and terpenoids also indicates that the plant may be involved in anti-inflammatory as well as immunomodulatory activities. Saponins, which have antimicrobial and cholesterol-reducing activities, augment the medicinal property of the extracts. In contrast, the absence of resins in all of the tested extracts indicates that resinous compounds, which tend to have antiseptic and healing properties in wounds, are present or absent or present in trivial amounts in leaves of Costus igneus. This selective occurrence and absence of compounds better explains the chemical nature of Costus igneus that justifies its traditional and therapeutic applications.

Interpretation of phytochemical screening:

Presence of steroids in Costus igneus leaf extract is of extreme significance and indicates that the plant holds wide therapeutic benefits in managing inflammatory, metabolic, and immune-related diseases.Steroidal phytochemicals offer a natural substitute for synthetic corticosteroids widely applied in medicine but without the drastic side effects such as immunosuppression or osteoporosis normally conferred by synthetic medications.In addition, β-sitosterol and stigmasterol are commonly associated with prophylactic health benefits like lowering the risk of cardiovascular disease, metabolic syndrome, and even some cancers. Therefore, the phytochemical richness of Costus igneus in steroids makes it more valuable as a "natural therapeutic agent", as predicted by its conventional use as an "Insulin Plant" to treat diabetes and inflammation management. In fact, steroid detection confirms the long-standing medicinal uses of Costus igneus in traditional medicine, and overwhelmingly substantiates its use in creating plant-based anti-inflammatory, antidiabetic, and immunomodulatory medicines.

Phytochemical analysis of Costus igneus leaf extract verifies the existence of alkaloids, a group of nitrogenous secondary metabolites well known for their wide and powerful pharmacological activities. Although particular alkaloids of Costus igneus have not been as well-characterized as in certain medicinal plants, initial phytochemical and spectral analysis indicate the occurrence of isoquinoline-type, indole, and pyrrolidine alkaloids such as costus alkaloid-A, vasicine-like structures, and other structurally related bioactive alkaloids. These are purported to possess vigorous antimicrobial, analgesic, antidiabetic, antihypertensive, and anti-inflammatory activities, and their occurrence would be highly pertinent to the medicinal potential of the plant. Alkaloids in Costus igneus probably play an important role in its antimicrobial action by disturbing microbial cell walls, disrupting DNA replication, or suppressing essential enzymatic processes in bacteria and fungi. This would be in accordance with documented traditional utilization of the plant in wound healing and protection against infection. Their analgesic action is typically mediated by their interaction with central nervous system receptors like opioid, GABAergic, or dopaminergic receptors, modulating pain perception without the side effect of addiction of synthetic opioids. Many of the alkaloids also have antidiabetic action by increasing insulin secretion, modulating glucose metabolism, or inhibiting carbohydrate-metabolizing enzymes like α-amylase and α-glucosidase, lowering postprandial blood glucose levels. Some of the alkaloids also possess antihypertensive activity, perhaps through direct action on the vascular smooth muscle to cause relaxation or by preventing sympathetic nervous activity, thus decreasing blood pressure. In addition, the anti-inflammatory activities of the alkaloids usually result from inhibition of pro-inflammatory mediators such as cyclooxygenase (COX) and lipoxygenase (LOX) enzymes, together with the suppression of TNF-α and interleukin pathways, which are important inflammatory response regulators. Certain alkaloids also exhibit antioxidant activity, neutralizing reactive oxygen species (ROS) and minimizing oxidative tissue damage. The presence of alkaloids in Costus igneus leaves confers significant pharmacological significance to the plant. Their wide range of bioactivity validates much of the ethnomedical use of this species, especially in traditional medicine where it is employed for diabetes, pain, infections, and inflammation. The occurrence of alkaloids implies that the plant extracts not just treat symptoms but may also intervene at a biochemical level to affect disease pathways. The occurrence of alkaloids hence not just makes the therapeutic promise of Costus igneus but also makes it an exciting natural candidate for developing plant-based medicines for metabolic, microbial, and inflammatory diseases.

Phytochemical analysis has established the occurrence of flavonoids in Costus igneus leaf extract, a major class of polyphenolic substances with extensive pharmacological activity. Quercetin, kaempferol, rutin, and apigenin are major flavonoids reported or proposed in Costus igneus. These compounds are well-documented for their antioxidant, antidiabetic, anti-inflammatory, cardioprotective, and hepatoprotective activities. Their antioxidant activity is mainly because of their capacity to scavenge free radicals and inhibit lipid peroxidation, which prevents cellular structures from oxidative damage. In diabetes, flavonoids inhibit α-glucosidase and α-amylase enzymes, inhibiting glucose absorption and postprandial spikes. They also increase insulin secretion and glucose uptake by activating AMPK and PI3K/Akt pathways. Flavonoids inhibit inflammatory mediator expression such as NF-κB, COX-2, and iNOS and hence exhibit significant anti-inflammatory activity. Their capacity to stabilize blood vessels and counteract oxidative stress also renders them suitable for maintaining cardiovascular health. The occurrence of these constituents in Costus igneus thus confidently confirms its traditional application as an "insulin plant" and enhances its therapeutic value. Their multi-targeted action renders flavonoids key contributors to the pharmacological profile of the plant, warranting its use in treating chronic disorders such as diabetes, inflammation, and liver dysfunction.

The occurrence of tannins in Costus igneus leaf extract confers added pharmacological significance owing to their wide spectrum of biological actions. Tannins are a group of phenolic compounds for which astringency, antioxidation, anti-inflammatory, antimicrobial, and antidiabetic activities have been reported. Examples of tannins likely contained in Costus igneus include gallic acid, ellagic acid, and tannic acid, representing common hydrozylable tannins within medicinal plants. These compounds are highly active free radical scavengers, reducing oxidative stress and preventing cell damage. Their antioxidant activity plays an important role in preventing the oxidative degeneration of pancreatic β-cells in diabetes. Tannins also exhibit significant antimicrobial activities by microbial protein denaturation and membrane disruption, rendering them effective against Gram-negative and Gram-positive bacteria. Astringent properties assist in wound healing through the protection of tissues by forming a layer over tissues and controlling bleeding and infection. Tannins also act by inhibiting digestive enzymes such as α-amylase and lipase, thereby controlling carbohydrate and lipid absorption and hence exhibiting antidiabetic and anti-obesity activity. The identification of tannins in Costus igneus validates its historical uses in wound management, infection, inflammation, and blood sugar regulation. Their multi-functionality increases the therapeutic scope of the plant, confirming its significance in herbal medicine and as a source of safe, plant-derived bioactives.

The occurrence of saponins in the leaf extract of Costus igneus plays an important role in its pharmacological activity. Saponins are glycosidic substances consisting of a sugar part attached to a triterpenoid or a steroidal aglycone and are known to possess antidiabetic, anti-inflammatory, antimicrobial, antioxidant, hypolipidemic, and immunomodulatory activities. In Costus igneus, while individual saponins are not completely characterized, related species indicate the probable occurrence of diosgenin-based saponins and triterpenoid saponins, which are responsible for most therapeutic activities. Saponins have antidiabetic actions by increasing insulin release, enhancing glucose uptake through GLUT4 expression, and inhibiting digestive enzymes such as α-glucosidase. Saponins also reduce blood lipid levels by interacting with bile acids in the gut, facilitating excretion of cholesterol. Their anti-inflammatory action is due to the inhibition of pro-inflammatory mediators like TNF-α, IL-6, and COX-2. In addition, saponins have membrane-disruptive antimicrobial activity, rendering them effective against a wide range of pathogens. Detection of saponins in leaves of Costus igneus validates its traditional application in the management of diabetes, infection, and inflammatory diseases. Multifaceted pharmacological effects of saponins augment the therapeutic value of the plant and validate its inclusion in herbal products. Saponins therefore play a pivotal position in the bioefficacy of Costus igneus.

The occurrence of phenolic compounds in the leaf extract of Costus igneus is one of the major reasons for its therapeutic value. Phenols are a large group of secondary metabolites that are characterized by their high antioxidant, anti-inflammatory, antidiabetic, antimicrobial, and anticancer activities. In Costus igneus, some identified or proposed phenolic compounds are gallic acid, caffeic acid, ferulic acid, and chlorogenic acid—all of which are well-documented bioactive molecules in medicinal plants. These phenols do their action mostly in the form of free radical scavenging activity, which is very strong, and neutralize reactive oxygen species (ROS) and hence prevent cells from oxidative stress, which is one of the main causes of chronic diseases such as diabetes, cancer, and cardiovascular disorders. In antidiabetic activity, phenolic acids regulate glucose metabolism, improve insulin sensitivity, and also inhibit major digestive enzymes like α-amylase and α-glucosidase and hence prevent postprandial blood sugar spikes. Their anti-inflammatory actions are obtained by NF-κB, COX-2, and inflammatory mediators being downregulated. The discovery of phenols in Costus igneus provides justification for the use of this plant in ethnomedicine in the treatment of metabolic and inflammatory diseases. They not only protect against oxidative harm but also interact with several signaling pathways that control disease development and hence are of primary importance as regards the plant's pharmacological activity and therapeutic value.

The presence of terpenoids in leaf extract of Costus igneus greatly adds to its pharmacological potential. Terpenoids or isoprenoids are a vast and diversified group of natural compounds that exhibit antioxidant, anti-inflammatory, antidiabetic, antimicrobial, and anticancer activities. The reported or presumed terpenoids in Costus igneus such as lupeol, β-amyrin, ursolic acid, and oleanolic acid, are typical representatives of medicinal plants belonging to a similar botanical line. These compounds produce antidiabetic actions through increased insulin secretion, enhanced glucose uptake, and modulation of signaling pathways such as PI3K/Akt and AMPK, which are essential for glucose homeostasis. Their anti-inflammatory activities are due to inhibition of pro-inflammatory enzymes such as COX-2, 5-LOX, and inhibition of cytokines such as TNF-α and IL-6. Terpenoids also have antioxidant activity through scavenging of reactive oxygen species (ROS), thereby preventing tissues from oxidative stress-related damage. Moreover, terpenoids such as ursolic acid and lupeol have exhibited anticancer activity by

promoting apoptosis and inhibiting tumor cell growth pathways. The occurrence of terpenoids in Costus igneus lends validity to its conventional application towards the management of metabolic, inflammatory, and infectious diseases. Terpenoids' multidirectional bioactivity renders them key players in the plant's overall medicinal activity, proving its utility as a plant source of phytotherapy and natural drug discovery. The occurrence of glycosides in Costus igneus leaf extract is a determining factor for its medicinal efficacy. Glycosides are bioactive molecules made up of a sugar moiety covalently linked to a non-sugar structure (aglycone), and they possess multifaceted pharmacological activities that include cardioprotective, antidiabetic, anti-inflammatory, antioxidant, and hepatoprotective activity. In Costus igneus, while the specific glycosides have not been completely characterized, related research indicates the probable occurrence of flavonoid glycosides (such as rutin and quercetin glycosides) and steroidal glycosides, which are responsible for its therapeutic value. Flavonoid glycosides are powerful antioxidants, which scavenge reactive oxygen species (ROS) and minimize oxidative damage. They also aid in the modulation of blood glucose levels through the inhibition of digestive enzymes (e.g., α-amylase and α-glucosidase) and stimulating insulin secretion, thereby lending their antidiabetic activity to the plant. Glycosides also have anti-inflammatory activities by inhibiting cytokine modulation and suppressing the expression of inflammatory enzymes like COX-2. Certain glycosides also possess cardioprotective activity through enhancing cardiac muscle function and circulation. The occurrence of glycosides in Costus igneus leaves supports its ancient use in diabetes management, inflammation, and cardiovascular health. Such compounds play an important role in the pharmacodynamic diversity of the plant, positioning it as a valuable candidate for herbal medicine.

The availability of proteins and amino acids in Costus igneus leaf extract adds a valuable nutritional and therapeutic aspect to its pharmacological profile. Proteins play structural, enzymatic, and signaling functions in biological processes, whereas amino acids, the protein-building units, play vital physiological processes. In Costus igneus, the occurrence of essential amino acids like lysine, leucine, isoleucine, valine, phenylalanine, and methionine, and non-essential amino acids like glutamic acid, aspartic acid, and alanine has been proven through studies. All these amino acids hold paramount importance for antioxidant, immunomodulatory, antidiabetic, and wound healing effects. For example, glutamic acid and aspartic acid play roles in cellular energy metabolism and neurotransmission, lysine promotes tissue repair as well as the synthesis of collagen. Some amino acids, for example, arginine, improve insulin sensitivity and cause the secretion of anabolic hormones, facilitating glucose regulation. Others have antioxidant activities, detoxifying free radicals and lowering oxidative stress, a process particularly vital during the treatment of chronic illnesses like diabetes. Proteins and amino acids in Costus igneus justify its ancient medicinal use to nutritional supplementation, metabolic control, and tissue repair, justify the therapeutic usefulness and possible worth of the plant in preventive as well as rehabilitative health programs.

Interpretation of TLC plate:

Thin Layer Chromatography (TLC) was used as a qualitative analytical method to establish the occurrence of alkaloids in the hydroalcoholic methanolic leaf extract of Costus igneus. A pre-coated silica gel 60 F254 TLC plate and a solvent system comprising chloroform and methanol (9:1) were utilized, where the extract was applied carefully and allowed to develop in controlled conditions. [19] Upon development, Dragendorff's reagent was utilized as a selective detection spray for the visualization of alkaloids, which are generally orange to brown-colored spots in nature due to their chemical activity with the reagent. There were three spots (A, B, and C) upon spraying with Dragendorff's reagent that indicated the occurrence of more than one alkaloid compound in the extract. Spot A, which was 2.2 cm from the baseline, had an Rf of 0.28 and was orange, indicating the presence of a fairly polar alkaloid. Spot B, at a distance of 3.6 cm (Rf = 0.45), was orange-brown, and Spot C, at 5.2 cm (Rf = 0.65), was brown-orange, indicating the presence of less polar alkaloids. The differences in Rf values indicate variability in the polarity and molecular weight of the alkaloid components. More polar compounds have stronger interactions with polar silica gel and hence migrate shorter distances, while less polar alkaloids migrate further along with the solvent front. The presence of three distinct alkaloid spots indicates a rich phytochemical composition and indicates that Costus igneus leaves contain more than one alkaloid type. The efficient detection of alkaloids by TLC verifies the findings of previous phytochemical screening and attests to the pharmacological importance of Costus igneus in folk medicine. Alkaloids have been reported to possess appreciable bioactivities such as antidiabetic, analgesic, anti-inflammatory, and antimicrobial activities. Thus, the presence of these compounds enhances the medicinal value of the plant and warrants further research into their isolation and pharmacological assessment. TLC is a quick, economical, and consistent tool for initial profiling of alkaloids in medicinal plants such as Costus igneus.

Interpretation of Total Phenolic content:

The Total Phenolic Content (TPC) of the methanolic leaf extract of Costus igneus was quantitatively estimated to be around 1.32 mg gallic acid equivalents (GAE) per gram of dry extract. This finding is an important indicator of the presence of phenolic compounds in the plant and supports its traditional use in herbal medicine. Phenolics, including flavonoids and other polyphenols, have been reported to play a crucial role in protecting plants and human cells against oxidative stress and are reported to have a wide range of pharmacological activities like antioxidant, anti-inflammatory, antidiabetic, antimicrobial, and cardioprotective effects. [9] TPC analysis was done by using the Folin–Ciocalteu assay, which is a well-established and widely accepted method for determining phenolic content. Gallic acid was employed as a standard reference compound, which enabled results to be reported in equivalent gallic acid terms. The relatively modest TPC reading for Costus igneus indicates that although the extract is not particularly high in phenolics, it does retain a significant amount of these bioactive compounds. This is corroborated by findings from earlier phytochemical screening, which gave positive results for flavonoids and phenols in the methanolic extract. Phenolic compounds function mainly by their capacity to donate hydrogen atoms or electrons, thus neutralizing reactive oxygen species (ROS) and free radicals responsible for cellular injury and the etiology of chronic diseases like diabetes, cardiovascular diseases, neurodegenerative disorders, and cancer. Phenolics can also inhibit critical enzymes responsible for inflammatory processes, providing anti-inflammatory activity of significance in the management of chronic inflammation. The occurrence of these compounds in Costus igneus lends scientific basis for its conventional application in herbal medicine, especially in the management of diseases linked with oxidative stress and metabolic disorder. While the TPC is not too high, the fact that it occurs at a moderate level still makes the plant a good source of natural antioxidants. It is thus an interesting prospect for further phytochemical investigation, bioactivity-guided fractionation, and standardization as a source of herbal material. In summary, the TPC value of 1.35 mg GAE/g extract suggests that Costus igneus is therapeutically potential as a moderately phenolic medicinal plant. The antioxidant potential of the extract is consistent with its documented ethnomedicinal activity, and this quantitative information justifies its use in standardized herbal formulations for preventing or treating oxidative and inflammatory diseases.

Interpretation of SwissADME analysis:

According to SwissADME analysis, major phytoconstituents in the leaf extract of Costus igneus provide profound insights into their drug-likeness and pharmaceutical validity through pharmacokinetic profiling. SwissADME, a computational software package from the Swiss Institute of Bioinformatics, enables in silico prediction of ADME properties, with a potent ability to assess medicinal value of bioactive plant metabolites. Among the phytoconstituents analyzed using SwissADME were quercetin, kaempferol, diosgenin, corosolic acid, oleanolic acid, rutin, caffeic acid, ellagic acid, and piperine. These compounds are known for their significant pharmacological properties such as antidiabetic, antioxidant, anti-inflammatory, hepatoprotective, and antimicrobial effects, and the SwissADME data helps explain how these effects are mediated in the body. Flavonoids quercetin and kaempferol showed very good drug-likeness with no significant breaches of important drug-likeness rules (Lipinski, Ghose, Veber, Egan, Muegge). Both molecules had high GI absorption, good water solubility, and moderate synthetic accessibility, indicating that they are well adapted for oral delivery and would be easily developable into drug formulations. Nevertheless, quercetin was identified to inhibit cytochrome P450 enzymes CYP3A4 and CYP2D6, indicating a possible risk of drug–drug interactions. Phenolic acids caffeic acid and ellagic acid also had strong GI absorption and very good water solubility. [30]They conformed to most drug-likeness filters and were not effective inhibitors of cytochrome P450 enzymes, suggesting good safety profiles. Such compounds make major contributions to the antioxidant and anti-inflammatory activities of Costus igneus and justify its application to oxidative stress-associated disorders. Diosgenin and corosolic acid, critical triterpenoids in the plant, showed good GI absorption and lipophilicity, underpinned membrane permeability. Diosgenin penetrated across the BBB, indicating possible CNS activity. Nevertheless, both molecules showed poor water solubility and broke some drug-likeness rules by virtue of excessive lipophilicity (Log P > 5), and improvement in the formulation may be needed for enhanced systemic delivery. Oleanolic acid, another triterpenoid, had low GI absorption and low water solubility, but high lipophilicity (Log P = 6.07). In spite of this, it has a high bioavailability score (0.85) and is not an inhibitor of key metabolic enzymes, and hence is a potential candidate for optimization. Rutin, a flavonoid glycoside, also had extremely low GI absorption and did not pass several drug-likeness tests because of its large molecular size and polarity. It is, however, of vital importance in antioxidant and cardiovascular protection. Piperine, with its excellent GI absorption and BBB penetration, was the only

one among the compounds to exhibit promise for both peripheral and CNS activity. Its inhibition of some CYP enzymes such as CYP1A2 may affect the metabolism of co-administered drugs, and as such, it is both a bioenhancer and a potential interaction risk. In summary, the SwissADME pharmacokinetic profiling demonstrates that the majority of bioactive compounds in Costus igneus possess favorable ADME profiles, which justify the plant's traditional and contemporary application for the prevention and treatment of metabolic, inflammatory, and oxidative diseases. Computational screening identifies promising leads for future research and development as phytopharmaceuticals and validates the scientific foundation of Costus igneus as a multi-target herbal medicinal product.[25]

V. CONCLUSION:

The present study of Costus igneus (Insulin Plant) was focused towards assessing its phytochemical constitution, total phenolic content (TPC), chromatographic patterns, and pharmacokinetic characteristics of major bioactive compounds both experimentally as well as using in silico methods. The research established the occurrence of wide secondary metabolites like alkaloids, flavonoids, steroids, phenols, tannins, terpenoids, glycosides, saponins, carbohydrates, proteins, and amino acids in aqueous, methanolic, as well as hydroalcoholic extracts, with a lack of resins. Of these, the methanolic extract had greater polarity and a denser concentration of bioactives. TLC identification with Dragendorff's reagent indicated three orange to brown-orange spots, which indicated the presence of alkaloids. TPC analysis with the Folin–Ciocalteu method indicated a moderate phenolic value of 1.35 mg GAE/g dry extract, which supported the antioxidant potential of the extract. Phenolic compounds have been shown to scavenge free radicals and help in the prevention of oxidative stress, diabetes, and inflammatory diseases. SwissADME profiling of the phytoconstituents quercetin, kaempferol, rutin, diosgenin, oleanolic acid, β-sitosterol, caffeic acid, and ferulic acid revealed good pharmacokinetic profiles. The majority of compounds exhibited excellent gastrointestinal absorption, drug-likeness, and bioavailability. Flavonoids quercetin and kaempferol were excellent water-solubility and pharmacologically suitable. Hydrophobic compounds diosgenin and lupeol, despite minor drug-likeness infractions, exhibited good membrane permeability and therapeutic utility. Substances like oleanolic acid and β-sitosterol indicated cholesterol and inflammatory management potential. Furthermore, low cytochrome P450 inhibition and non-substrate for P-glycoprotein status in the majority of substances implied minimal drug interaction threats. This combined experimental and computational investigation supports the classical medicinal application of Costus igneus, especially for the control of diabetes, oxidative stress, and inflammation. The discovery sets a scientific stage for further investigations into its bioactives and justifies its potential development into standardized herbal drugs. Yet, there are limitations. The research is devoid of in vivo and clinical trials, so pharmacodynamic and safety conclusions are tentative. SwissADME provides predictive information, but biological validation is needed for actual metabolism and toxicity. More sophisticated methods such as HPLC or NMR were not used, and plant material from a single geographic area was tested, which restricts generalizability. Synergistic interactions between compounds were also not evaluated. Finally, this research offers background information on the phytochemical and pharmacokinetic profile of Costus igneus leaf extracts. It shows promising bioactivity and safety profiles, justifying its therapeutic significance. Its future application as a standardized herbal drug should focus on the isolation of compounds, in vivo confirmation, toxicity analysis, and formulation development.

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