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Pharmacognostic and Phytochemical Evaluation of Ganoderma Lucidum Extract

Monika Gorakh Gore[1], Pallavi Gangadhar Rithe[2], Vidya Raosaheb Kale[3]
[1, 2]*Student,* [3]*Assistant Professor, Yashodeep Institute of Pharmacy Chh. Sambhajinagar*

Abstract: ***Ganoderma Lucidum, commonly referred to as Reishi or the Mushroom of Immortality, has historically been esteemed in traditional medicine for its numerous healing applications. These applications are primarily associated with its polysaccharides (beta-glucans) and triterpenoids (Ganoderic acids). Even though it is widely sought after, the quality of Reishi that can be bought frequently differs. This analysis examines the existing knowledge on Ganoderma lucidum based on published studies. It includes the identification of the mushroom, its chemical composition, and its cited therapeutic benefits. We subsequently outline a technique for isolating its essential compounds: hot water is employed to extract the hydrophilic polysaccharides, while ethanol is utilized for the lipophilic triterpenoids. We qualitatively assessed the existence of the primary chemical groups and subsequently measured them accurately using UV spectrophotometry. The data collected here provides a scientific foundation for ensuring the consistency of Reishi products. It additionally aids in organizing additional research and developing new products that incorporate Ganoderma lucidum***

Keywords: Ganoderma Lucidum, Reishi Mushroom, Pharmacognosy, Phytochemical Screening, Polysaccharides, Triterpenoids, Standardization

I. INTRODUCTION

Ganoderma lucidum is a well-known medicinal mushroom commonly called Reishi (Japan) or Lingzhi (China), which belongs to the family Ganodermataceae, a type of shelf fungus. It has been used for over 2,000 years in traditional Asian medicine and is often referred to as the “mushroom of immortality.” People once called it the Mushroom of Immortality - used for ages in traditional healing systems across China. Its structure is tough, taste is bitter and Kidney-like top with a shiny red-brown finish. Found sitting on rotting oak or beech trees where climates are mild or warm-temperate. Most of its health reputation ties back to two key groups of active compounds. Among the active elements in G. lucidum are beta-glucans, which influence immune function quite strongly, while ganoderic acids offer protection against inflammation and support liver health. Though widely sold around the world, actual product quality shifts dramatically - depending on where it's grown, what it's grown on, or how it's processed. Because of these inconsistencies, solid scientific benchmarks become essential. To address that issue, this work looks closely at commercial samples through detailed chemical analysis, measuring two main compounds using separate extraction techniques.

II. TAXONOMY

Synonyms: Reishi (Japanese), Lingzhi (Chinese) Mushroom, Mushroom of Immortality.
Biological source: Ganoderma Lucidum is a mushroom originated from China, Japan, and Korea, and is now widely cultivated in Asia, Europe, and North America for its medicinal and nutritional properties.

Table no. 1 Ganoderma Lucidum

Reign	Fungi
Phylum	Basidiomycetes
Class	Agaricomycetes
Order	Polyporales
Familia	Ganodermataceae
Genus	Ganoderma
Species	Ganoderma Lucidum

Figure 1 Ganoderma Lucidum

III. DISCOVERY

The species was formally reported by William Curtis in 1781 (initially as Boletus lucidus), but its traditional usage predates this by millennia. The oldest Chinese pharmacopoeia, the Shen Nong Ben Cao Jing (Eastern Han dynasty, 25-220 AD), classified Lingzhi as a superior medicine used to promote longevity and enhance vital energy, reserved primarily for royalty and nobility

IV. CULTIVATION/ SOURCES

Ganoderma lucidum is mainly cultivated on hardwood logs, sawdust bags, or agricultural waste substrates. Controlled environmental conditions temperature (25–30°C), humidity (85–95%), and low light are required for fruiting body development. Commercially, it is grown in China, Korea, and Japan and is now widely cultivated in Asia, Europe, and North America for its medicinal and nutritional properties.

Variability in cultivation methods leads to differences in polysaccharide and triterpenoid content.

The cultivation process of Ganoderma lucidum typically involves the following steps:

1) Spawn preparation: The spawn is prepared by inoculating a sterile substrate, such as grain or sawdust, with Ganoderma lucidum mycelium.
2) Substrate preparation: The substrate is prepared by mixing a combination of ingredients, such as hardwood sawdust, rice bran, and water.
3) Bagging: The substrate is filled into bags, which are then sealed and sterilized.
4) Inoculation: The spawn is inoculated into the sterilized substrate.
5) Incubation: The inoculated substrate is incubated at a controlled temperature (around 2530°C) and humidity (around 60-70%) to allow the mycelium to grow.
6) Fruiting: The substrate is transferred to a fruiting room where it is exposed to a controlled environment (around 20-25°C and 80-90% humidity) to induce fruiting.
7) Harvesting: The fruiting bodies are harvested when they are mature, usually 7-10 days after fruiting.
8) Drying: The harvested fruiting bodies are dried to a moisture content of around 10% to preserve them for storage and transportation

V. MICROSCOPICAL DESCRIPTION

Diagnostic microscopic features include double-walled, brown, ellipsoid basidiospores which are truncate (flat) at the apex, possessing distinct germ pores. Examination of the powder confirms the presence of these spores alongside fragments of the dark, thick-walled hyphae and the laccate cuticular layer, serving as key markers for species identification and quality control.

VI. MORPHOLOGY

Ganoderma lucidum (Lingzhi/Reishi) is a woody, polypore fungus known for its kidney-shaped, fan-like cap that features a shiny, varnished reddish-brown to dark brown exterior. Its morphology features a stalked (stipitate) form, a white-to-yellowish pore surface under the cap, and spore prints that appear brown with truncated spores.

Morphological Characteristics of G. lucidum:

1) Fruiting Body (Basidiocarp): Typically features a firm, cork-like, woody feel with a glossy, polished look. Typically, it is fan-like to kidney-shaped and can reach sizes of up to 35 cm in diameter, featuring a clear cap (pileus) and stem (stipe).
2) Coloration: The cap starts as white or yellow and matures into shades of red, reddishbrown, or dark brown with a white edge.
3) Stipe (Stalk): Usually found, brown to reddish-brown, frequently lateral, and may reach up to 1.5 times the cap's diameter.
4) Pores/Hymenium: The lower side features small pores, averaging around 4–5 pores for every millimetre. They seem white to grayish-white, changing to brown as they age.
5) Spores: Basidiospores are oval-shaped, brown, and possess a thick double wall with a flat or truncated end. They usually measure about (8.2–12.1) × (4.8–8.9) µm
6) Internal Context: The inner tissue is cork-like, displaying hues from light buff to cinnamon.
7) Mycelium: During growth, the fungus may develop either a filamentous structure or a pelleted shape (compact, spherical clusters of hyphae).

The morphology, growth, and market prospects of Ganoderma lucidum may differ somewhat depending on environmental factors, and accurate identification is frequently verified through DNA testing.

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VII. CHEMICAL CONSTITUENT

Ganoderma lucidum (Reishi mushroom) contains over 400 bioactive compounds, primarily triterpenoids (ganoderic acids) and polysaccharides (ganoderans), which deliver potent anti-inflammatory and antioxidant effects. These constituents alleviate inflammation by inhibiting inflammatory mediators, modulating immune cells (macrophages, T cells), and suppressing signaling pathways like NF-κB and MAPK.

A. Chemical Constituents are

1) Triterpenoids (Ganoderic Acids): More than 100 triterpenes exist in G. lucidum, responsible for its bitter taste and key anti-inflammatory and antitumoral activities, with notable examples including ganoderic acids A, B, C, and D.
2) Polysaccharides: These act as immunomodulators, strengthening immune response while decreasing oxidative stress by increasing enzymes like superoxide dismutase (SOD).
3) Other Compounds: Phenols (for antioxidant action), sterols, glycoproteins, and minerals (selenium, zinc, calcium) also contribute to its therapeutic properties.

B. Anti-Inflammatory Mechanisms

1) Reduction of Proinflammatory Cytokines: Polysaccharides decrease the levels of IL-6, IL-1β, and TNF-α.
2) Inhibition of Inflammatory Enzymes: Triterpenes inhibit enzymes involved in inflammation.
3) NF-κB and MAPK Inhibition: The compounds modulate signaling pathways, which are critical in reducing chronic inflammation.
4) Antioxidant Activity: By increasing antioxidant enzyme activity, the compounds reduce ROS-induced tissue damage.
5) Neuro-inflammation Protection: Triterpenoids have been shown to reduce neuroinflammation and neuronal apoptosis.

VIII. RAW MATERIAL AND REAGENTS

Fungal matter known as Ganoderma lucidum arrives already dried, in powdered form, shipped straight from a vendor. Before anything else happens, that batch gets checked by experts to confirm its species identity. Out in the field, fresh caps are laid into drying units set precisely at forty degrees Celsius - left there one full day so water levels drop enough for safe keeping. Once brittle and dry, they go spinning inside a motor-driven grinder until broken down completely.

That crumbly mix passes through a tight filter, each hole barely wider than 160 threads per inch. After sorting, what remains is kept away from light, sealed without gaps in a closed jar, sitting still on a shelf under normal indoor conditions till it's time to test. From a lab supply source, ethanol at 95 percent arrives alongside deionized water, phenol, strong sulfuric acid, plus vanillin - each meant for test use. Glucose steps in when measuring polysaccharides; oleanolic acid handles triterpenoid levels. These reference materials anchor the numbers later on.

IX. EVALUATION PARAMETERS (METHODOLOGY)

The following pharmacognostic and phytochemical evaluation parameters have been reported in literature for the standardization of Ganoderma lucidum. These techniques help in identifying the mushroom, assessing its quality, and verifying its chemical constituents.

A. Pharmacognostic Evaluation

1) Macroscopic Examination

The crude drug (dried basidiocarps) was subjected to macroscopic examination to determine its morphological and organoleptic characters, which were compared against standard pharmacopeia monographs for authentication.

- General Appearance: The basidiocarps are corky to woody in texture, consisting of a distinct pileus (cap) and stipe (stalk).
- Pileus (Cap): The cap is reniform (kidney-shaped) to semi-circular, convex, measuring approximately 2–10 cm in diameter. The upper surface is covered with a hard, crust-like cuticle that is laccate (shiny and varnish-like).
- Color: The surface color ranges from reddish-brown to mahogany, often graduating to a lighter yellowish tint toward the margin.
- Hymenophore (Underside): The lower surface is whitish to pale brown, containing minute, round pores (tubes) rather than gills.

- Stipe (Stalk): The stalk is typically lateral or eccentric, cylindrical or flattened, with a color ranging from dark reddish-brown to nearly black, possessing the same laccate (shiny) appearance as the cap.
- Fracture: The fracture is fibrous and tough. The inner context (flesh) is firm and pale brown to cinnamon in color.
- Organoleptic Properties:
  1. Odor: Mild, distinct, and earthy (fungal).
  2. Taste: Distinctly bitter and lingering, which indicates the presence of bioactive triterpenoids.

2) Microscopic Evaluation

Thin sections of Ganoderma lucidum fruiting body, mounted in glycerin or lactophenol cotton blue, show a trimitic hyphal system with generative, skeletal, and binding hyphae. Generative hyphae are thin-walled with clamp connections, while skeletal hyphae are thick-walled and yellowish. Binding hyphae are highly branched and interwoven. Numerous brown, double-walled basidiospores (8–12 µm) with rough outer surfaces are seen. These features help confirm the authenticity of the fungus.

3) Physiochemical Evaluation

- Loss on Drying- Heating at 105°C in a hot air oven until constant weight to determine the moisture content, essential for storage stability.
- Total Ash Value- Incineration of the dried material at 55O°C in a muffle furnace.

Measures the total inorganic content and overall mineral content.

- Acid-Insoluble Ash– Treating the total ash residue with dilute HCl and re-incinerating.

Measures contamination by non-physiological inorganic matter, such as soil or sand.

B. Phytochemical Analysis

The phytochemical evaluation is designed to characterize the chemical composition of the extracts through sequential analysis: extraction, qualitative screening, and quantitative analysis.

• Dual-Solvent Extraction –

Two distinct extraction methods performed on separate portions of the authenticated G. lucidum powder to ensure selective isolation of the primary bioactive compounds. I. Polysaccharide-Rich Extract (Aqueous Extraction):

Dried Ganoderma lucidum fruiting body powder
↓
Defatting (Treatment with 95% Ethanol)
↓
Filtration and Maceration
↓
Hot Water Extraction (85–100°C for 2–3 hours)
↓
Filtration
↓
Concentration of filtrate
↓
Add 95% Ethanol (3–4× volume)
↓
Overnight precipitation of polysaccharides
↓
Centrifugation / Drying
↓
POLYSACCHARIDE EXTRACT

II. Triterpenoid-Rich Extract (Ethanol Extraction):

Dried Ganoderma lucidum fruiting body

↓
Powdering
↓
Place powder in Soxhlet apparatus
↓
Alcoholic Extraction (Maceration or reflux)
(Ethanol/Methanol, 4–6 hrs.)
↓
Collect alcoholic extract and filtrate
↓
Evaporate the solvent (Rotary Evaporator)
↓
Dry the residue
↓
TRITERPENOID EXTRACT

• Qualitative Phytochemical Screening-

The crude extracts subjected to standard chemical tests to confirm the presence of major secondary metabolite classes. These tests used colour changes to spot key groups. This screening includes indicator tests for:

I. Alkaloids: Utilizing reagents like Dragendorff's or Mayer's reagent (formation of a precipitate).
II. Triterpenoids/Steroids: Utilizing the Liebermann-Burchard test (colour change from red to blue-green).
III. Flavonoids: Utilizing the Shinoda test (Mg + HCl reduction).
IV. Carbohydrates/Glycosides: Utilizing Molisch's test.

• Quantitative Spectrophotometric Analysis: The concentrated extracts quantitatively analysed to determine the total content of the primary bioactive markers using validated colorimetric assays and a UV-Vis Spectrophotometer.

I. Total Polysaccharides Content (TPC) – Phenol-Sulfuric Acid Method:

A sudden burst of colour appears when phenol meets broken-down carbs, thanks to a strong acid splitting them first. The process relies on that vivid change to track carbohydrate levels accurately.

Start off by making a series of glucose solutions - each one stronger than the last - to build a reference pattern. This set becomes the measuring stick for what comes next. Start here with a set amount of the sugar extract - pour in some 5 percent phenol mix. Next, drop in strong sulfuric acid; that breaks down the big sugar molecule while warming things up right away.

A cool-down happens first, bringing the mix to room temperature so colours stay steady. After that settles, a reading takes place - absorbance checked at 490 nanometres using a blank as reference.

Using the glucose standard curve's regression formula, scientists figure out how much polysaccharide is present. This amount shows up as milligrams of glucose equivalent for every gram of extract tested. Numbers come from matching sample results to that curve. Results always land in mg GE per g of material.

II. Total Triterpenoids Content (TTC) – Vanillin-Perchloric Acid Method :

A colour appears when the triterpenoid's ring system meets vanillin, driven by perchloric acid pulling out water. What emerges is a mix you can see, shaped by intense acidity guiding the change.

A series of Oleanolic Acid solutions, each with a defined concentration, forms the basis for the reference pattern. This reference helps match sample levels to known values. Starting from low to high, these points build a measurable trend. Because consistency matters, every solution is handled the same way. The plotted response creates a line that reflects concentration changes.

Matching unknowns becomes possible once the shape stabilizes. Precision comes from careful measurement at each step.

A small portion of the triterpenoid extract, or a reference sample, has its liquid removed until solid remains. That leftover material gets mixed into a suitable liquid medium. After that, vanillin mix goes in along with perchloric acid. A warm water bath brings out the purple-red tint in the mix, followed by quick cooling. After that, readings take place at 548 nanometres using a reference sample for comparison.

What you get here depends on the oleanolic acid reference line. That number then shows how much triterpenoid exists in each gram of extract. Results appear as milligrams of oleanolic acid equivalent for every gram. Math comes straight from matching samples to that standard pattern.

Table no.2 ASSAY

Assay	Target Compound	Standard used for Calibration
Phenol Sulfuric Acid Method	Total Polysaccharides	Glucose (Results expressed as mg Glucose Equivalent/g extract)
Vanillin Perchloric Acid Method	Total Triterpenoids	Oleanolic Acid (Results expressed as mg Oleanolic Acid Equivalent/g extract)

X. MEDICINAL USES OF GANODERMA LUCIDUM

Ganoderma lucidum helps because it works in many ways inside the body. Its effects mostly come from two parts - polysaccharides and triterpenoids. One benefit shows up in how it supports immune function. Another way appears through reducing inflammation. Some changes happen at a cellular level over time. Effects can also show when stress on cells decreases. The full picture includes shifts in energy balance too. Here are some of the key benefits:

1) Immunomodulatory: It enhances the immune response by stimulating macrophages, NK cells and lymphocytes, helping the body fight infections. • Hepatoprotective: Triterpenoids (Ganoderic acids) stabilize hepatocyte (liver cell) membranes, accelerate the metabolism of toxin.
2) Anti-inflammatory: Reduces chronic inflammation and swelling, offering potential relief for conditions like asthma and arthritis.
3) Cardiovascular: Ganoderic acids reduce total cholesterol and LDL (bad cholesterol). Used for managing hypertension and hypercholesterolemia
4) Weight Loss: Supports metabolism by helping to reduce overall fat accumulation in the body.
5) Anticancer Potential: Polysaccharides and triterpenoids show apoptosis-inducing and anti-proliferative effects against various cancer cells.
6) Anti-Diabetic Effect: Helps regulate blood glucose by improving insulin sensitivity and reducing oxidative stress in pancreatic cells.
7) Anti-cancer: some compounds trigger cell death in tumor's while slowing their growth, this effect appears across several types of cancer cells when exposed to certain mushroom extracts.

A. *Some Additional Benefits*

1) Anti-Allergic Effect: Triterpenoids help inhibit histamine release and reduce allergic reactions such as asthma and dermatitis.
2) Neuroprotective Activity: Protects nerve cells from oxidative damage and may help in conditions like Alzheimer's and Parkinson's disease.
3) Anti-Aging Properties: Its antioxidant polysaccharides help maintain skin elasticity, reduce wrinkles, and promote cell regeneration.

XI. SIDE EFFECTS OF GANODERMA LUCIDUM

While Ganoderma lucidum mushroom are generally considered safe to eat, there are some potential side effects to be aware of:

1) Gastrointestinal Disturbances :May cause stomach upset, nausea, or diarrhoea in some individuals, especially at higher doses.
2) Dry Mouth and Throat Irritation :Long-term intake can lead to dryness of the mouth, throat, and nasal passages.
3) Skin Rashes or Allergic Reactions :Some people may experience itching, redness, or mild allergic reactions due to hypersensitivity.
4) Headache and Dizziness :Mild dizziness or headache can happen, especially when consuming concentrated extracts.
5) Liver Toxicity (Rare, High Doses) :At high dose, can cause liver toxicity.

6) Blood Pressure Fluctuations :It can slightly drop your blood pressure, which could be a problem if you're already on blood pressure meds.
7) Bleeding Risk :Ganoderma can thin your blood a little, it's risky to take if you're already on blood thinners like warfarin or aspirin.
8) Digestive Discomfort from Spores :Reishi spore powder, if not processed properly, may irritate the digestive tract.
9)

XII. RESEARCH GAP

Right now, Ganoderma lucidum gets used a lot across India, yet hard proof about its plant-based qualities remains scarce. Different growing materials change how strong each batch turns out. Heat levels during drying play a role too, altering active compounds unpredictably. Alcohol-based extractions pull out triterpenoids; water pulls polysaccharides - rarely are both checked at once. Most research skips comparing these two together within one test run. Without side-by-side results, judging quality becomes guesswork. So, consistency slips. A clearer way to measure what's actually inside store-bought versions would help everyone relying on it.

XIII. EXPECTED OUTCOMES

A. *Expected Pharmacognostic and Physicochemical Results*

Table no.3 Parameter

Parameter	Expected Result Range (Based on Literature)
Loss on Drying	Expected: 10-12%
Total Ash Value	Expected: 5-7%
Acid Insoluble Ash	Expected: 1.0%
Qualitative Screening	Expected: Positive for Triterpenoids, Carbohydrates, and Steroids.

B. *Expected Phytochemical Quantification Results: A. Extraction Yield*

1) Aqueous Extract Yield (Polysaccharide-Rich): After we take out the fat and use hot water, we should get a good amount back—probably about 10-30% of the original powder. This part will mostly be sugary compounds and proteins that dissolve in water.
2) Ethanol Extract Yield (Triterpenoid-Rich): The alcohol extraction won't give us as much, maybe 1-5%. It'll be a bit sticky or gummy, which makes sense since triterpenoids and fatty acids don't mix with water. B. Quantitative Assay Outcomes: Measuring light absorption gives clear signs of what chemicals are present. This method pins down exactly which substances show up every time. Details emerge when samples react to different wavelengths. What you see links directly to specific compounds
3) Total Polysaccharides (TPC): Accuracy comes from tracking these responses carefully A hefty dose of total polysaccharides usually lands between 150 and 500 milligrams of glucose equivalent for every gram of raw extract. Quality tends to climb when numbers sit in that span. Stronger batches often reflect better healing promise down the line.
4) Total Triterpenoids (TTC): These compounds show up clearly when tested in alcohol-based extracts. A single gram of raw extract often holds between 10 and 60 milligrams, measured as oleanolic acid equivalents. Testing this bitter part matters because levels shift depending on how and where the mushrooms grow. What you get can differ a lot from one batch to another.

XIV. POSSIBLE DOSAGE FORMS

So, Ganoderma lucidum extract shows up in all sorts of products—capsules, pills, tinctures, syrups, creams, even gels you put on your skin. If they want to boost your immune system, they usually go for the polysaccharide-rich stuff you swallow. But for things like inflammation or liver protection, they use extracts rich in triterpenoids. Choosing the right form just depends on how well it dissolves, how stable it is, how much your body can use, and what health problem it's supposed to help.

Since our ethanol extraction will give us a good amount of those fat-loving triterpenoids, and because we know they're good for fighting inflammation, a gel you put on your skin makes the most sense for what we're doing here.

1) Rationale: Ganoderic acids, which form a heavy extract of the triterpenoid class, reduce inflammation by inhibiting NF-Kb activation; this is known to trigger immune responses - alongside reducing production of histamines. As a topical application, the extract gets to work on areas of pain or inflammation without ever going through the liver process.

2) Approach to Formulation: The use of a cold gel formulation with either Carbopol 934 or 940, even as Emulsion-gel formulations, comes first. To this will be added the thick ethanol extract that will form a thin layer of rapid drying, light weight and deep transdermal delivery of herbal constituents.

3) Possible Application: Joint pain relief, skin conditions, localized swelling management can be possible. With the use of triterpenoids found in Ganoderma, which is a plant derived chemical with steroid-like effects. Such constituents have the ability to calm irritated tissues. Rather than using artificial drugs, the formulation uses natural alternatives. It may be effective without the need for conventional medicine.

XV. CONCLUSION

The findings of this review and the proposed experimental procedure indicate that Ganoderma lucidum may have therapeutic properties.

These properties come largely due to the polysaccharides in Ganoderma lucidum that improve the immune system and triterpenoids that protect the liver. The raw Ganoderma lucidum can be identified, and the physicochemical parameters can be determined. This allows controlling the quality of the material obtained from suppliers. In the experimental part, the proposed procedure will involve a dual solvent for extraction followed by quantitative analysis using Phenol-Sulfuric Acid method for polysaccharides and Vanillin-Perchloric Acid for triterpenoids. This will help determine the quantity of these substances for standardization purposes. This review suggests the need for standardization of Ganoderma lucidum.

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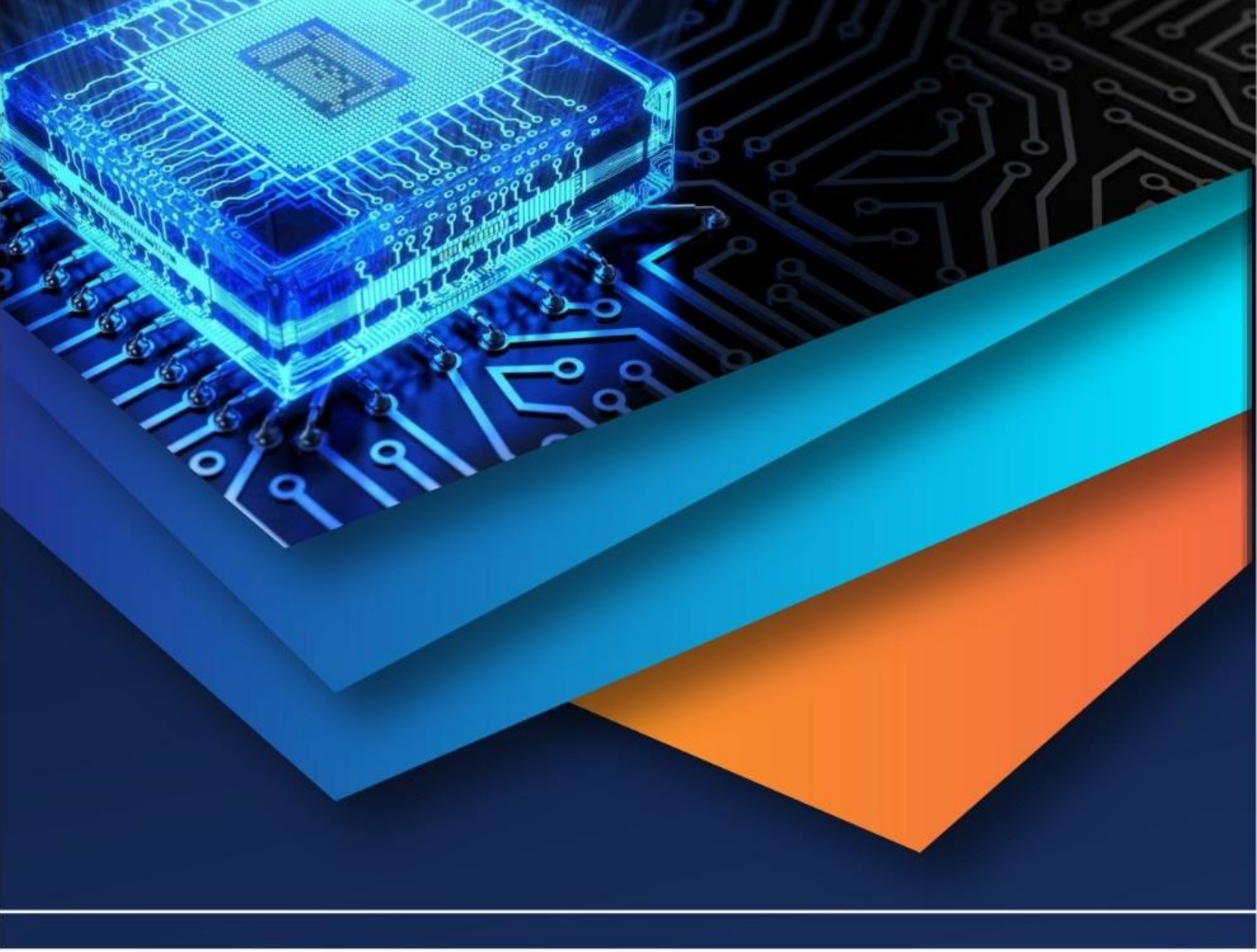

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