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Formulation and Evaluation of Poly Herbal Mouth Wash against Different Mouth Disorder

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Abstract: ***Our oral cavity is a sweet able place to grow different types of bacterial species either harmless or harmful for human. Many of us are suffering different types of mouth disease and feeling insecure or hesitate to speak with another due to the unpleasant odor of mouth. To maintain a good oral hygiene mouthwash plays a important role in our daily lifestyle. From ancient age medicinal plants are considered as a store room of different types of biological activity in Ayurveda, Unani and Siddha, and have important role to cleanse tooth and prevent different human pathogens are responsible for unpleasant odor, inflammation of teeth root, dental plaque. Herbal mouthwash contains different medicinal plant part extract have various pharmacological activities and shows significant advantages which helps to cure different types of oral disorder and helps to maintain oral hygiene. Herbal mouthwash is suitable for any age group due to less or no other side effect instead of chemical mouthwash. Aqueous extract of Neem, Turmeric, Clove, Fennel, Guava leaves, Pudina, Ginger, Tulsishows effective antimicrobial, antifungal activity, anti-inflammatory and anti-plaque properties. In this research work herbal mouth wash was evaluated depends on various parameter like color, pH, Visual appearance, Phase separation, Homogeneity and antibacterial properties.***

Keywords: Herbal Mouthwash, Ayurveda, Neem, Turmeric, Anti-inflammatory activity, Antibacterial activity

I. INTRODUCTION

Ancient Egyptians think that an unclean body was impure and emphasize on hygiene and beauty for maintain their health. In 40-90 AD a Greek physician and surgeon Pedanius Dioscorides suggestedin his medical text book a mouth wash mixture contain a decoct of the leaves of the olive tree, milk, the juice of pickled olives, gum myrrh with wine and oil, pomegranate peelings, nutgalls, and vinegar for treatment of bad breath.The Romans used human urine as a hidden component in their cleanser and purchased Portuguese urine because they believed it to be stronger. From The ancient Romans age to 18th century urine add as a main ingredient in toothpaste and mouthwash because presence of ammonia and its cleaning ability. Main ingredient in toothpaste and mouthwash because presence of ammonia and its cleaning ability.(1)

In twenty first century human being is very much cautious about their hygiene, health and beauty. They prefer to choose harmless herbal mouthwash instead of chemical mouthwash. Mouthwash prepared from different parts of plant extract are known as herbal mouthwashes recognized as useful for human which helps to remove bad odour and reduce dental cavity, plaque and gingivitis, mucositis due to presence of different active constituent with zero side effect.(2)

Most of the Chemical mouthwashes contain artificial drugs which act as an instant tooth whitener, sanitizer, pain reliever and have several side effect.(3) However, they have the tendency to discolour teeth and may cause harmful impacts and unsafe for children.

Herbal mouthwash always preferable due to its non-irritating, non-staining, and alcohol-free nature. It does not contain any harmful chemicals and totally safe for children. It doesn't show any bad effects after swallowing by mistake for people of any age(4).

II. MATERIALS AND METHODS

A. Collection Of Plant Parts And Chemical Ingredients

Plants samples of the following species like *Azadirachta indica* (Neem), *Ocimum teniflorum* (Tulsi), *Psidium guajava* (Guava), *Mentha arvensis* (Pudina), flower buds of *Syzygium aromaticum* (Clove), seeds of *Foeniculum vulgare* (Fennel), and stems of *Glycyrrhiza glabra* (Liquorice), *Zingiber officinale Roscoe* (ginger) and *Curcuma longa* (turmeric) rhizomes and Rose (*Rosa rubiginosa*)were randomly collected from local herbal garden and chemical like sorbitol (sweetener), Sodium benzoate (preservative) were randomly collected from local market.

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B. List of Ingredients

TABLE 1: INGRADIENT DETAILS: PLANT PARTS &CHEMICAL INGRADIENTs

Sr. No.	Ingredients	Chemical Constituents	Plant parts Extractives and Chemical raw material	Uses
1.	Neem (*Azadirachta indica*) Family- Meliaceae Plant part-leaves	Azadirachtin, Nimbolinin, Nimbin, Nimbidin, Nimbidol, Sodium Nimbinate, Gedunin, Salannin, And Quercetin (5)		Reduce plaque and gum disease. growth of microorganisms. prevents gum disease and bleeding.(5)
2.	Turmeric (*Curcuma longa)* Family-Zingiberaceae Plant part-Rhizome(root)	Turmerone, turmeroneoids, and curcumin(6)		Bacteriostatic, antibacterial, and antimicrobial characteristics. Turmeric reduces inflammation, burning, and ulcers(6)
3.	Clove (*Eugenia caryophyllus*) Family-Myrtaceae Plant part-Leaves	Eugenol, caryophyllene, and methyl amyl ketone,Eogenol acetate,2-Heptanone,Gallic acid,Heptyl acetate.(7)		Tooth analgesic, prevents cavities, treats bad odours, and improves circulation. Effectively used in preventing cavities(7)

4.	Fennel (*Foeniculum vulgare)* Family-Apiaceae Plant part-Seeds	Trans-anethole (31.49%), 2-pentanone (25.01%), fenchone (11.68%) and benzaldehyde-4-methoxy (8.01%)(8)		Increases the amount of saliva secreted in the mouth and assists with removing foul odour from the mouth.(8)
5.	Guava leaves (*Psidium guajava)* Family-Myrtaceae Plant part-Leaves	Pentacyclic triterpenoid guajanoic acid, beta-sitosterol, uvaol , oleanolic acid , and ursolic acid (9)		Helps to reduce plaque and the swelling of inflamed gums. treatment for mouth ulcers that's effective.(9)
6	Ginger (*Zingiber officinale Roscoe)* Family-Zingiberaceae. Plant part-Stem rizome	Carbohydrates (50–70%), lipids (3–8%), terpenes, and phenolic compounds. Terpene components of ginger include zingiberene, *β*-bisabolene, *α*-farnesene, *β*-sesquiphell,(10)		Help your teeth and gum line become whiter and stronger. The antibacterial properties of ginger prevent the growth of bacteria and plaque. Anti-Inflammatory (10)
7	Tulsi (*Ocimum sanctum)* Family-Lamiaceae. Plant part-Leaves	Eugenol,euginaluroso lic acid, carvacrol, linalool,limatrol (11)		It works effectively to reduce plaque and gingivitis.(11)

8	Pudina (*Mentha arvensis)* Family-Lamiaceae Plant part-Leaves	Menthol, menthone, and cineole,(+ -)-Menthyl acetate (12)		Pudina is an excellent gingivitis treatment. It has antiviral, antibacterial, and analgesic properties.(12)
9	Sorbitol	---		Give off a warming, gently sweet, and refreshing mouthfeel. Against dental caries resistance.(13)
10	Sodium benzoate	----		Anti-corrosive and preservative(14)
11	Rose water Family-Rosa rubiginosa Plant part-Flower	----		Contains antibacterial and anti-inflammatory properties that help to disinfect the mouth (15)

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C. Aqueous Extraction Process Of Plants Parts

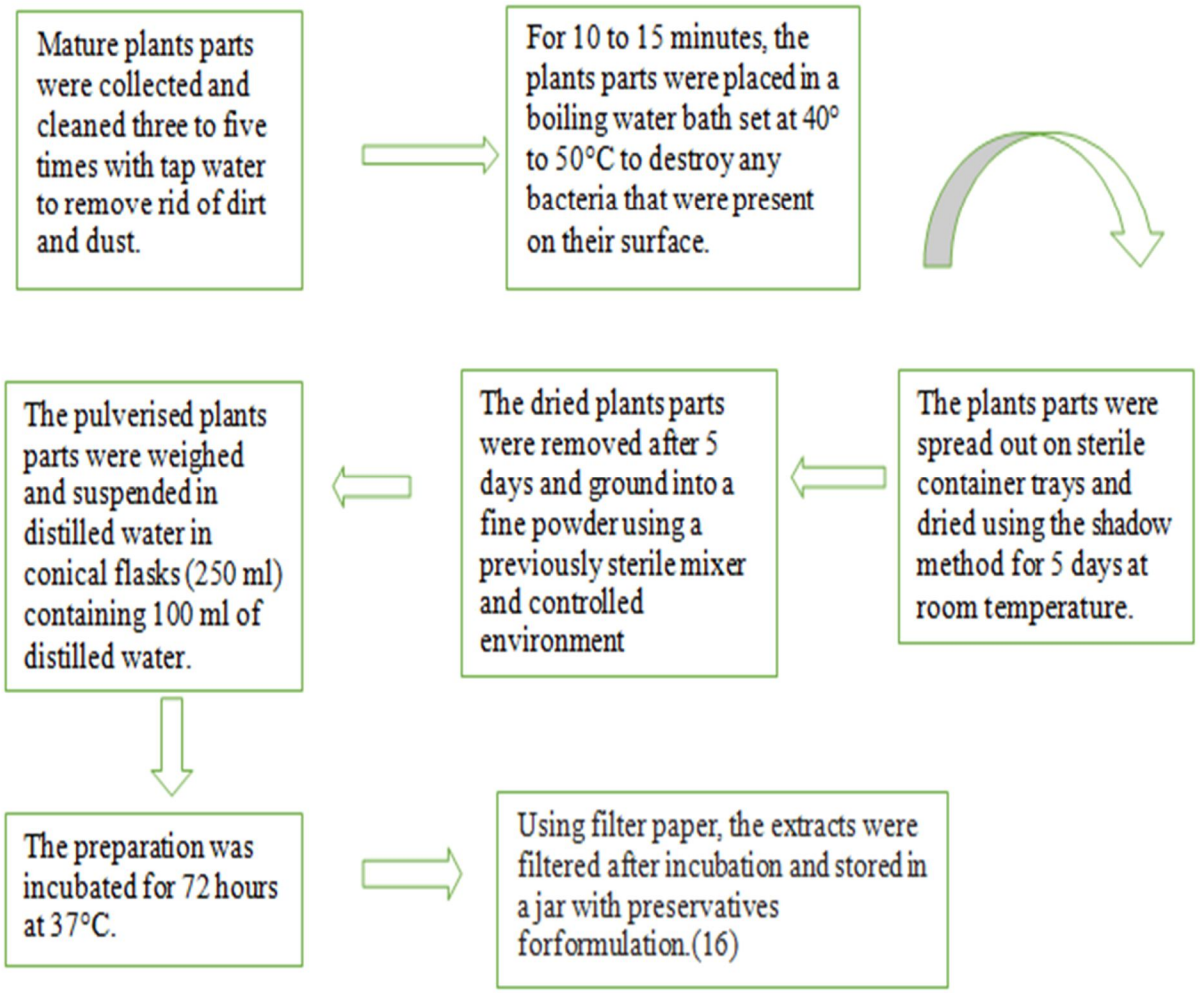


FIGURE 1: FLOW DIAGRAM OF EXTRACTION PROCEDURE.

D. Water soluble extractive of herbal mouthwash

TABLE 2: PERCENTAGE OF WATER SOLUBLE EXTRACTIVE FOUND FROM PLANTS PART

SL NO	INGREDIENTS	Weight of Sample taken in 100 ml water	PETRIDISH WEIGHT (BLANK)	PETRIDISH WEIGHT WITH EXTRACTIVE	% FOUND
1.	Neem	1.0 gm	42.38 gm	42.51 gm	13.6%
2.	Turmeric	1.0 gm	41.78 gm	41.93 gm	15.2%
3.	Clove	1.0 gm	43.35 gm	43.55 gm	20%
4.	Fennel	1.0 gm	40.14 gm	40.36 gm	21.9%
5.	Guava leaves	1.0 gm	40.98 gm	41.07 gm	9.4%
6.	Ginger	1.0 gm	39.23 gm	39.45 gm	22.4%
7.	Tulsi	1.0 gm	44.13 gm	44.27 gm	14.4%
8.	Pudina	1.0 gm	43.23 gm	43.43 gm	20%

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E. Phytochemical Screening

TABLE 3: PHYTOCHEMICAL SCREENING OF DIFFERENT PLANT EXTRACTIVE

Sr. No.	Name of Herbs	Tests	Observation	Images
1	Neem	Aqueous Extract + Ferric chloride 5% solution	Dark colorization occurred due to the presence of tannin. A=Plant extract B=$Fecl_3$ C=Reactant	
2	Clove	Aqueous Extract + Ferric chloride 5% solution	Dark colorization occurred due to the presence of tannin. A=Plant extract B=$Fecl_3$ C=Reactant	
3	Turmeric	Aqueous Solution of turmeric + boric acid	Yellowish brown due to presence of curcumin. A=Plant extract B=Boric acid C=Reactant	
4	Guava	Aqueous Extract + 1% $FeCl_3$	Blackish-greencolourisationdue to presence of tannin. A=Plant extract B=$FeCl_3$ C=Reactant	

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5	Tulsi	5 mg Extract of Ocimum sanctum in test tube + few drops HCl	Yellowish-Green colour due to presence of flavonoids. A=Plant extract B=HCl C=Reactant	
6	Ginger	Few ml ginger extract + few drops Wagner,s reagent.	Green precipitate confirms the present of alkaloids. A=Plant extract B= Wagner's reagent C=Reactant	
7	Fennel	2ml extract + 1ml Mayer's reagent.	Light green precipitateformed , indicating the presence of alkaloids. A=Plant extract B= Mayer's reagent C=Reactant	
8	Pudina	1ml extract + two drops ferric chloride	Green – blackish colour indicate presence of catechol tannins. A=Plant extract B= $Fecl_3$ C=Reactant	

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F. Formulation Table

TABLE 4: FORMULATION TABLE

Sl. No.	Ingredient name	Each 50 ml contain	Purposes
1	Neem	4 ml	Antimicrobial
2	Turmeric	4 ml	Antibacterial and antimicrobial
3	Clove	2 ml	Analgesic
4	Fennel	7 ml	Anti-inflammatory, Antifungal
5	Guava leaves	3 ml	Anti-bacterial
6	Ginger	2 ml	Anti-inflammatory and anti bacterial
7	Tulsi	8 ml	Strong Anti-inflammatory
8	Pudina	5 ml	Antiseptic, cooling agent
8	Sorbitol	10 ml	Sweetener
9	Sodium benzoate	0.025 gm	Preservative
10	Rose water	q.s	Emollient, cooling agent

III. EVALUATION OF PREPARED HERBAL MOUTHWASH

1) Colour and Odour: Visual inspection was used to examine physical characteristics, including colour and odour.

2) pH: A digital pH metre was used to determine the pH of the herbal mouthwash produced a standard buffer was used to calibrate the pH metre. 1 millilitre of mouthwash was weighed, dissolved in 50 ml of purified water, and the pH of the mixture was determined.

3) Test for microbiological growth in developed mouthwash: A control was made and manufactured mouthwash was inoculated in agar media plates using the streak plate method. The plates were put in the incubator, where they would be kept for 24 hours at a temperature of 37±2°C. After the incubation period, the plates were removed and tested for microbial growth.

4) Test for stability: The purpose of the stability test is to make sure that the mouthwash formulations can keep the same properties throughout the long term before conducting an antimicrobial test. Prior to antimicrobial testing, many mouthwash formulations undergo stability testing. This test evaluated the mouthwash formulation's visual appearance, physical separation, and homogeneity. The mouthwashwas then stored at temperature at 30°C while the appearance was assessed on each 10 days up to 1 month and the results were recorded.

IV. RESULTS AND DISCUSSION

After 1 month study the appearance of mouthwash maintained its colour and homogeneity. There was no phase separation observed in formulated mouthwash. The pH of mouthwash shows slightly acidic in nature and maintained its pH. The formulated mouthwash was showing a positive activity on different kinds of human pathogen like E. coli(-ve) and S. aureus(+ve).

Since they did not produce any microbial growth when this was placed in the agar medium, the formulation was free of microorganisms. This mouthwash is made entirely from herbs and does not contain alcohol. Stability tests for the formulation were conducted for both chemical and physical changes. There was no observable, significant differences in the formulation's qualities. Alcohol use is a known risk factor for head and neck cancer, as is tobacco use. It has always been controversial whether using mouthwash with alcohol increases the risk of developing cancer. Neem, clove, and other important plant extracts are antibacterial ingredients that, when added to frequent brushing and flossing, have been shown to reduce plaque and gingivitis.

For various mouthwash concentrations amount, the agar diffusion method was used to determine their antibacterial activity. For E. coli (-ve), the zone of inhibition was found to be 8 mm for 50 µl, 11 mm for 100 µl, 18 mm for 150 µl, and 20 mm for 200 µl, respectively. For S. aureus, the zone of inhibition was found to be 5 mm for 50 µl, 10 mm for 100 µl, 13 mm for 150 µl, and 15 mm for 200 µl, respectively. These findings demonstrated the significant antibacterial activity of the herbal mouthwash and the capability of the current product to prevent bacterial growth in the oral cavity.

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TABLE 5: STABILITY STUDY DATA

TEMPERATURE & HUMIDITY	EVALUATION PARAMETERS	OBSERVATION (In DAYS)			
		Initial	10	20	30
30^0 C ± 2^0C / 60% ± 5% RH	Visual appearance	Dark brown	Dark brown	Dark brown	Dark brown
	Phase separation	Nil	Nil	Nil	Nil
	Homogeneity	Good	Good	Good	Good
	pH	6.53	6.50	6.48	6.45
	Wt./ml	1.05	1.05	1.05	1.05

TABLE 6: RESULT OF ANTIBACTERIAL STUDY

ORGANISM	ZONE OF INHIBITION (mm)			
E. coli(-ve)	50 µl	100 µl	150 µl	200 µl
	8	11	18	20
S. aureus(+ve).	5	10	13	15

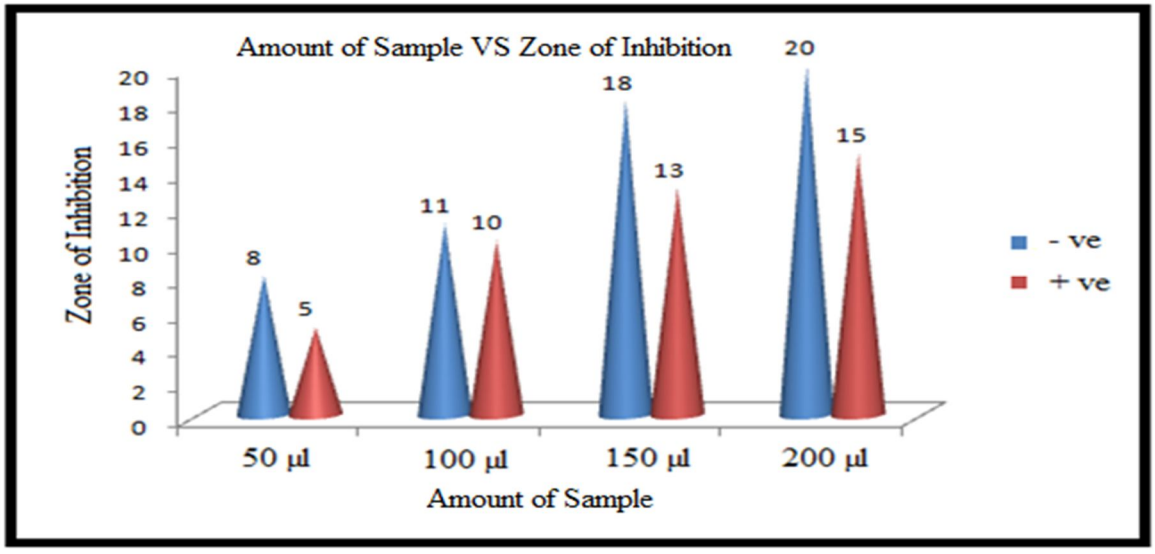


FIGURE 2: BAR DIAGRAM OF AMOUNT OF SAMPLE USED VS ZONE OF INHIBIOTION

V. CONCLUSION

In a study, the active components of several extracts were combined to create an efficient formulation for polyherbal mouthwash. It is alcohol-free. Neem, Tulsi, guava leaves, fennel, turmeric, clove, ginger, pudina and rose water extracts were used in the current research for their potential anti-bacterial properties. The zone of inhibition findings provided additional proof that this herbal mouthwash was an effective plaque inhibitor. Patients favoured it for its taste and ease of use.

The results of the physicochemical screening indicate that the colour and smell of the current herbal formulation are acceptable, with better after effects and a pleasant smell.

It has been shown that the natural herbs included in this mixture have therapeutic advantages for treating oral hygiene problems and bad breath. These plants have a long history of successful use, according to numerous studies. It is simple to rinse one's mouth with this herbal mouthwash and prevent a variety of oral health issues.

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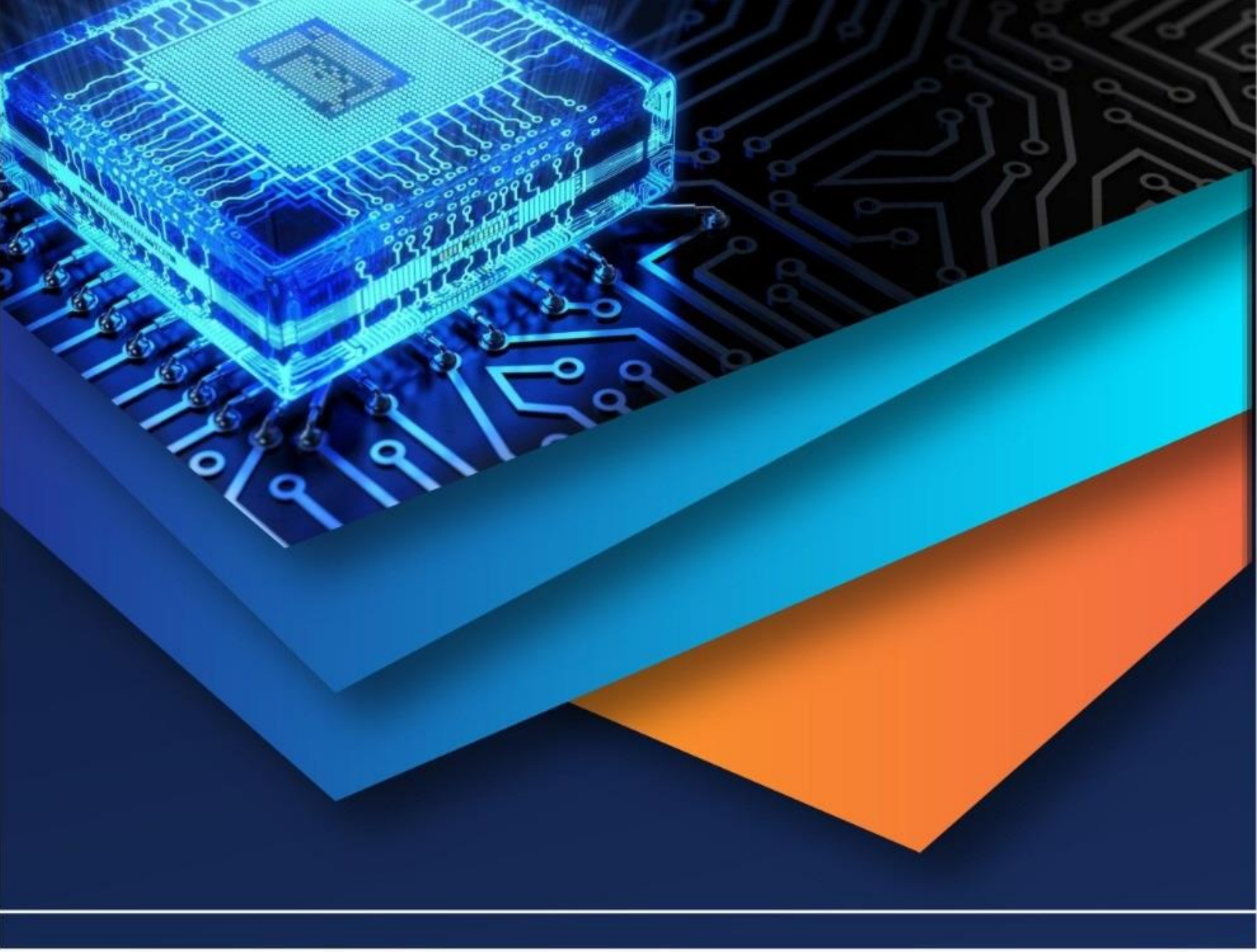
CONFLICT OF INTEREST**:-** Nil

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