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Bioremediation of Hydrocarbon Contaminated Soil using Sewage Sludge

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Abstract*: This research were evaluated the study on bioremediation of soil contaminated with petroleum hydrocarbons. The objective of this study was to investigate whether organic nutrients enhance the process of microbial degradation. The samples were collected from the workshops where soil samples contain waste oil spills and Sewage Sludge was applied at 3different proportions namely0%, 30% and 60%. A laboratory scale batch reactor was developed and maintained upto 90days at room temperature. Bacterial count was done to determine the development of petroleum degrading heterotrophic bacteria. Spectrophotometer was used to determine the petroleum hydrocarbons present. The results indicate that the application of organic materials and SS at 30% and 60% concentration were able to remediate the soil by 80.38%, 95.44% and 97.57% respectively.

Keywords: Include at least 5 keywords or phrases

I. INTRODUCTION

This document is a template. For questions on paper guidelines, please contact us via e-mail. Environmental pollution is one of the greatest threats to society in the future. One of the primary causes of environmental degradation in a country could be attributed to rapid growth of population, which adversely affects the natural resources and environment. Increasing economic development and a rapidly growing population in India has taken the country from 300 million people in 1947 to over one billion people today was putting a strain on the environment, infrastructure, and the country's natural resources. Rapid industrialization further worsens the situation.

It was the industrial revolution that gave birth to environmental pollution as we know it today. The emergence of large factories and consumption of immense quantities of coal and other fossil fuels gave rise to unprecedented air pollution and the large volume of industrial chemical discharges added to the growing load of untreated human waste. It was estimated that 10,000 more chemicals are introduced worldwide annually and industrialized countries generate more than 90% of the world's annual total of 325-375 million tons of oxic and hazardous waste originating from chemical and petrochemical industries.

Every year spillage of tones of fuel and lubricating oil has become a major environmental hazard. In October 2014, a single oil spillage of 546 tonnes was reported at Mid-Valley Pipeline of Louisiana, United States. A recent incident highlighted how time consuming a cleanup operation of oil spillage can be. According to CNN International, in Nov 2013, oil-spillage occurred in Philippines due to a typhoon and till date the cleaning up process is going on. There are also long-term effects on ecosystems related to the release of toxic components over a prolonged period as the oil breaks up and the concentration of toxicants in organisms towards the top of the food chain increases

II. MATERIALS COLLECTION

A. Sample Collection

Soil samples were collected from a nearby automobile shop in Chidambaram town. Soil samples were collected in a sterile polythene bag. Samples are collected for a maximum depth of 10cm with a scoop. And it is preserved in a room temperature before starting the experiment. Sewage sludge used as an inoculums. Sewage sludge is collected from a nearby pumping station.

B. Sample Preparation

A measured quantity of 50 gm soil sample was transferred into a conical flask to which a distilled water of 200ml was added. The conical flask was tightly closed and placed in rotary shaker for a period of 48hours at 250rpm. The samples were filtered using a standard filter paper. And the filtered samples were analyzed.

III.EXPERIMENTAL SET UP

A. Preparation Of Growth Medium

5gram of soil samples inoculated with sewage sludge at different proportions was added to a mineral salt broth medium (MSM) of 200 ml, which composed of 1L of $(NH_4)_2SO_4$, 1g; $KH_2\ PO_4$, 0.2g; $K_2\ HPO_4$, 1.6g; $MgSO_4\ .7H_2O$, 0.2g; NaCl, 0.1g; $FeSO_4$, 0.1g and $CaCl_2.2H_2O$, 0.02g and 10g/L of glucose were added and p^H of the medium was adjusted to 7. The broth medium containing hydrocarbon polluted soil was incubated at room temperature with orbital shaker of 120RPM

Figure 1 Photographic View Experimental Setup

B. Enumeration of Total Heterotrophic Bacteria

Samples were prepared by adding 1ml biomass to 9ml sterile distilled water. Serial dilution of the sample was carried out up to 10^{-4} dilutions. An aliquot (0.1ml) of the dilution was inoculated into a Petri dish containing Plate Count Agar (PCA) in duplicate for total cultivable bacteria. The inoculums were spread on the plate using sterile bent glass wood. The plates were incubated at 28°C for 48 hours. Colonies that were formed during this incubation period were counted.

C. Determination of Hydrocarbon

Hydrocarbon content of the biomass samples was determined by 10ml of sample in 20ml toluene in a 200ml flask. After shaking for 30 minutes on orbital shaker 120rpm, the liquid of the extract was measured at 420nm using spectrophotometer. The TPH in sample was estimated using the standard curve derived from fresh engine oil diluted with toluene.

IV. RESULT AND DISCUSSION

A. Physicochemical Analysis

The physicochemical properties of the soil and organic nutrients used for the bioremediation trials are detailed in table 1, 2 and 3.

Table 4.1 Characteritics Of Hydrocarbon Polluted Soil

S. No.	SAMPLE PARAMETER	SITE1	SITE2	SITE3	SITE4	SITE5	SITE6	REMARK
1	MOISTURE CONTENT%	0.908	1.88	9.8	5.49	2.33	9.78	mg/l
2	pH	7.16	8.28	7.83	7.64	7.35	7.55	-
3	EC	1.411	1.412	0.335	0.190	0.186	0.270	Milli mho
4	NO_2	3	1	1.5	1	2	4	mg/l
5	PO_4	3.5	0.5	3	4	1.5	2	mg/l
6	SO_4	45	48	21	9	42	60	mg/l
7	Ca	48.77	61.11	19.90	11.24	11.90	18.11	mg/l
8	Na	51.62	31.45	16.71	8.66	6.13	17.79	mg/l
9	K	76.48	37.75	62.16	20.74	19.78	37.38	mg/l
10	Li	1.42	4.58	0.46	0.91	0.90	1.12	mg/l
11	TOC	185.0	64.46	67.01	41.18	76.62	102	mg/l

Table 4.3 Characteristics Of Inoculums For Sewage Sludge

S. No.	SAMPLE PARAMETER	SEWAGE SLUDGE	REMARK
1	MOISTURE CONTENT%	204	mg/l
2	p^H	6.25	-
3	EC	3.2	Milli mho
4	NO_2	100	mg/l
5	PO_4	2.1	mg/l
6	SO_4	210	mg/l
7	Ca	338	mg/l
8	Na	161	mg/l
9	K	85	mg/l
10	Li	311	mg/l
11	TOC	192.8	mg/l
12	COD	35600	mg/l
13	BOD	56	mg/l
14	DO	14.5	mg/l
15	HARDNESS	20	mg/l
16	CHLOREIDES	370	mg/l
17	TS	1500	mg/l
18	TSS	200	mg/l
19	TDS	1300	mg/l

B. Bioremediation of Petroleum Hydrocarbon Contaminated Soil Samples using and SS

Inocula carry SS containing directly enriched petroleum hydrocarbon degrading microorganisms at 30% and 60% application rates. At the end of the 90th day, bioremediation experiment results revealed that higher amount of petroleum hydrocarbons loss up to 68.98%, 81.69% and 92% (for 30% SS) and 80.38%, 95.44% and 97.57% (for 60% SS) for sample A, B and C were found respectively.

The petroleum hydrocarbons contaminated soil amended with SS recorded the highest reduction of petroleum hydrocarbon of nearly 97.57% compared with the 30% which was an efficiency of 92%. The reasons for results obtained might be due to differences in the nutrient content, particularly N and P in these two organic nutrients in stimulating indigenous microorganisms. Addition of N and P to a petroleum hydrocarbons polluted soil has been shown to accelerate the biodegradation of the petroleum hydrocarbons in soil. SS with the highest concentration of N and P proves to be effective inoculums in bioremediation of petroleum hydrocarbon contaminated soil. N and P are found to be an important nutrients required by a hydrocarbon utilizing bacteria to carry out effective and efficient bioremediation activities of xenobiotics in the soil environmental.

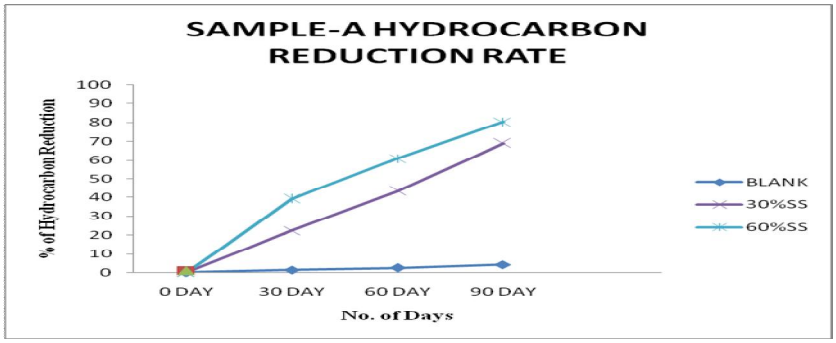

Figure 2 Percentage Of Hydrocarbon Reduction Rate Sample-A

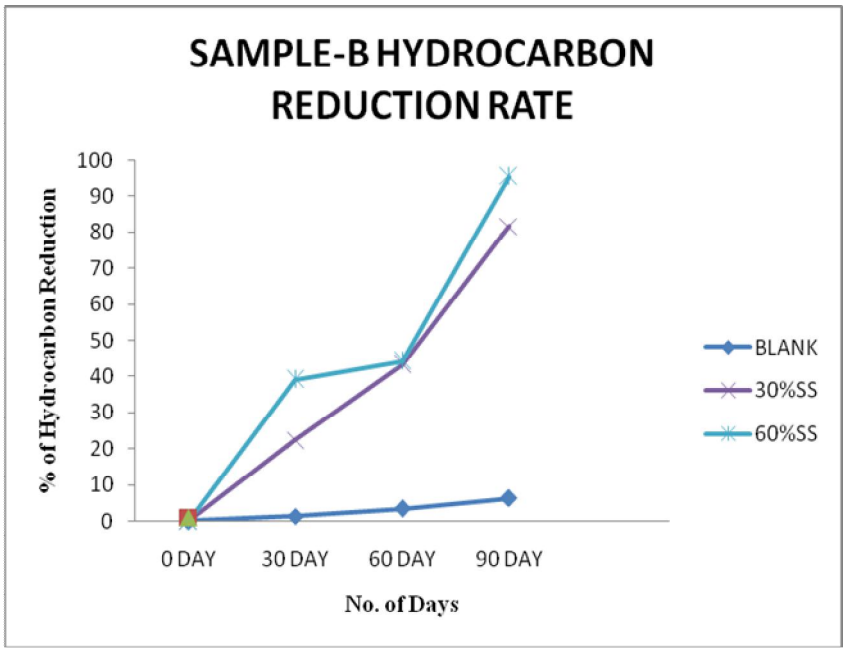

Figure 3 Percentage Of Hydrocarbon Reduction Rate Sample-B

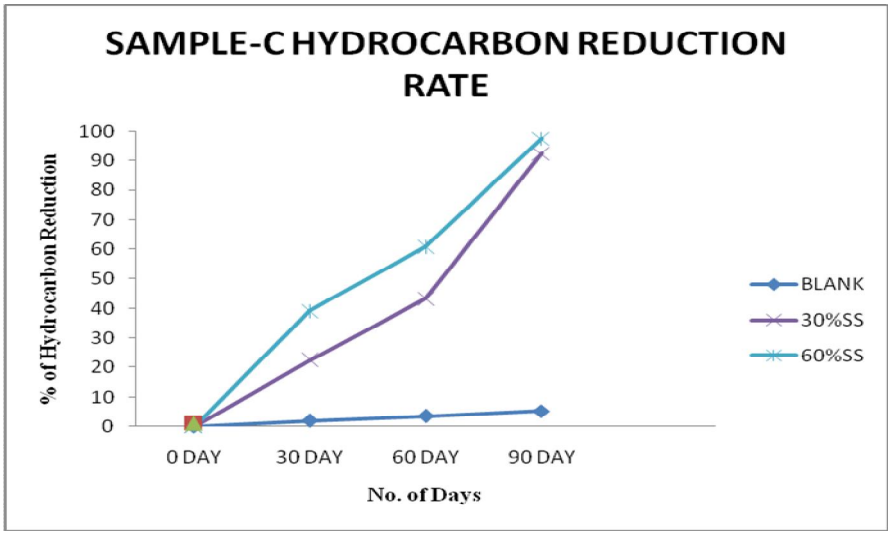

figure 4 percentage of hydrocarbon reduction rate sample-c

C. Microbial Counts

Counts of hydrocarbon degrading bacteria in soil contaminated with petroleum hydrocarbon organic nutrients is shown figure (10, 11, 12). The count of total heterotrophic bacteria (THB) in soil contaminated with petroleum hydrocarbon and amended with SS ranged from $1.3X10^5$cfu/g to $2.54X10^7$cfu/g. The unamended soil contains THB ranging between $3.2X10^5$cfu/g to $3.44X10^7$cfu/g for sample-A. It was relatively lower compared to organic nutrients amended soil. Microbial count was done at the end of the 90^{th} day. The THB count was found higher in nutrients amended soil compared to the un-amended polluted soil, it might be due to the presence of appreciable quantities of nitrate and phosphate in the organic nutrients. The higher microbial population counts in petroleum hydrocarbon contaminated soil amended with organic nutrient is accompanied by significant hydrocarbon bioremediation, indicating that the indigenous soil microbes utilized a portion of the C supplied by the diesel fuel as potential nutrients source.

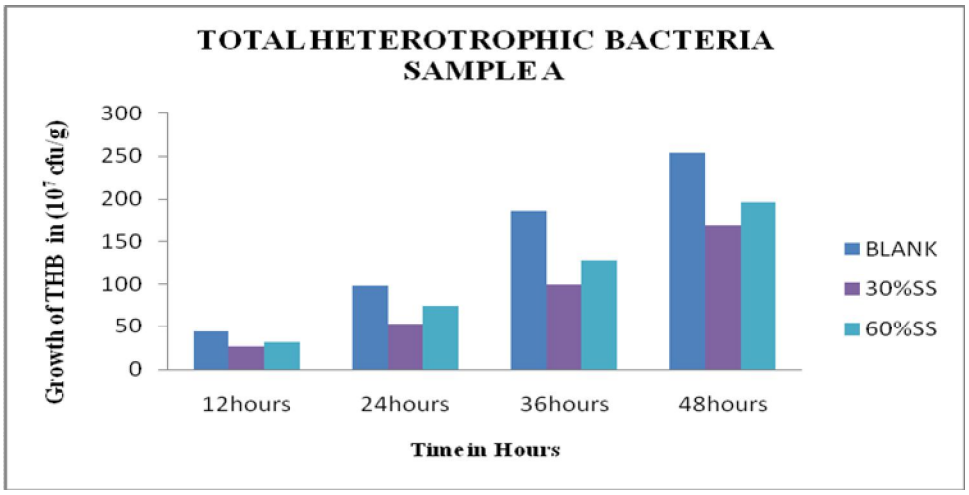

Figure 5 Enumeration Of Total Heterotrophic Bacteria Sample-A

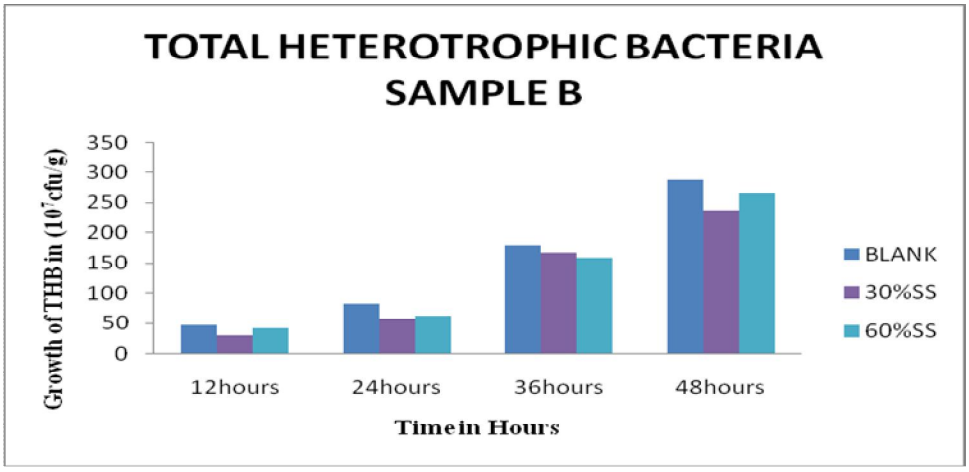

Figure 6 Enumeration Of Total Heterotrophic Bacteria Sample-B

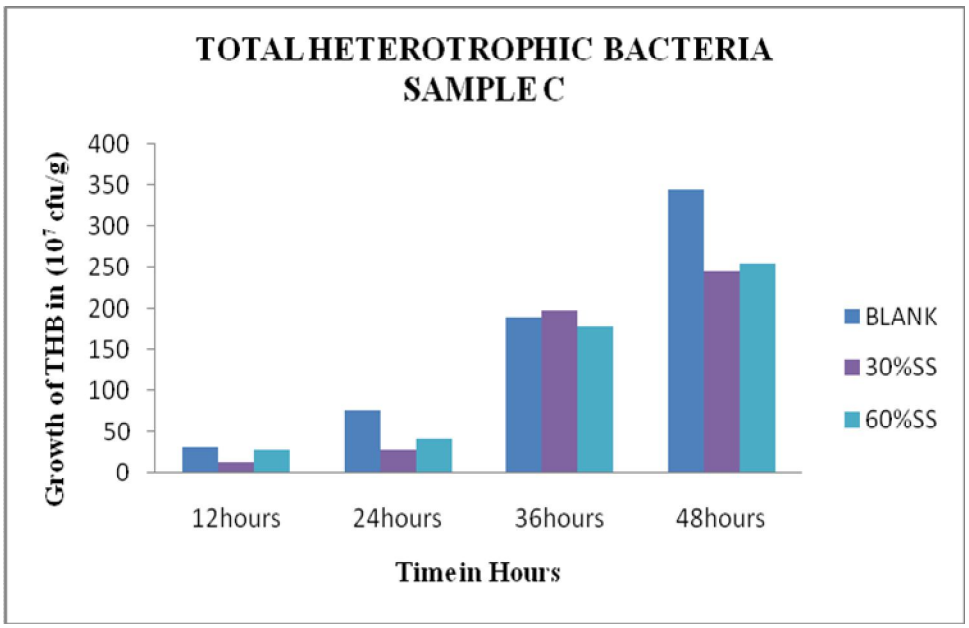

Figure 7 Enumeration Of Total Heterotrophic Bacteria Sample-C

V. CONCLUSION

The results of this study showed that soil contaminated with petroleum hydrocarbon can be treated with SS, whose reduction potential is 92% and 97.57% respectively. Reduction in total petroleum hydrocarbon in contaminated soil by 60% SS proves to be more effective. The study demonstrated the potential of 30% SS in enhancing the growth of indigenous microorganisms in the soil which in turn increases the bioremediation rate of petroleum hydrocarbon. Organic nutrient proved to enhance the multiplication of indigenous microbes thus enabling rapid biodegradation of the contaminated soil. Taking in to account that the amount of limiting nutrients such as Nitrate and Phosphate presented in the polluted soil is low. The use of organic nutrient as Nitrate and Phosphate source can enhance bioremediation process as well as solve the problem of waste management.

VI. ACKNOWLEDGMENT

The heading of the Acknowledgment section and the References section must not be numbered.
Causal Productions wishes to acknowledge Michael Shell and other contributors for developing and maintaining the IEEE LaTeX style files which have been used in the preparation of this template. To see the list of contributors, please refer to the top of file IEEETran.cls in the IEEE LaTeX distribution.

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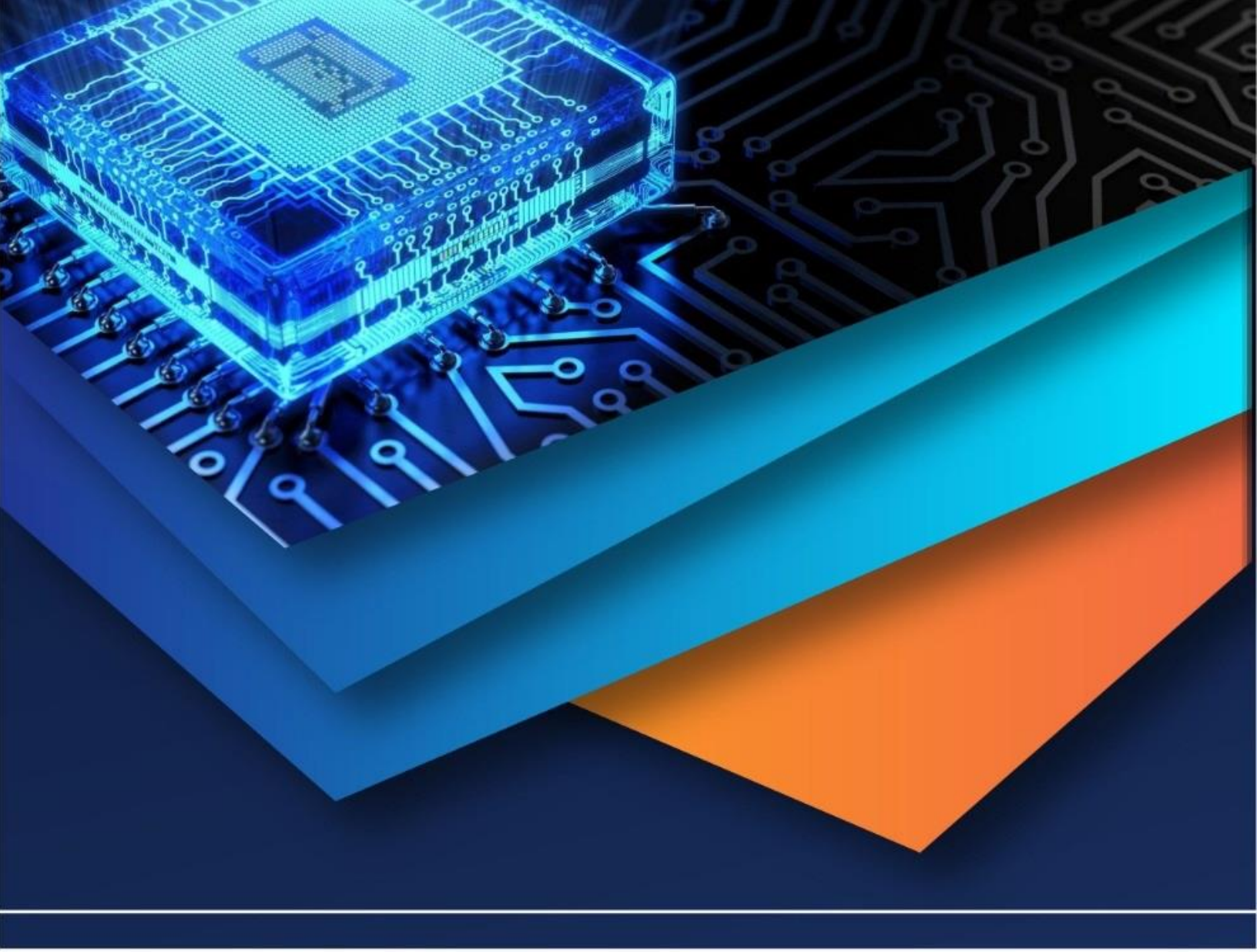

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