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Importance of Substrate and Microbes in Bio Fertilizer Production

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Abstract: ***Agriculture is the main economy in the eastern Uttar Pradesh. Due to availability of biodegradable waste (from the agricultural activity), it can be used as bio fertilizer by applying biological, physical and chemical treatment processes. Bio fertilizers have emerged as a highly potent alternative to chemical fertilizers due to their eco-friendly, easy to apply, non-toxic and cost effective nature. Biofertilizers can be prepared by different methods such as composting, vermin composting and some latest techniques of active microorganism application. Biodegradable waste such as baggase, rice straw, wheat straw which farmers generally burns can be used a substrate in the procress of biofertilizers production. These approach not only prepare organic fertilizers for the farmers but also controls air pollution caused by burning. In this paper include the proper discuusion on material and microorganisms used in process of production. It also described the effect of nitrogenous fertilizers used in the soil for across different section of the world.***

Keywords: biodegradable waste, bio fertilizer, bagasse, air pollution, soil.

I. INTRODUCTION

India's population is around 1.34 billion and continues to rise with the population growth pace of 1.18, as our county's population is growing day by day, it is expected that India will be the world's most populated nation by 2020. Population growth contributes to a reduction in the agricultural land due to the construction of homes, houses and manufacturers, etc., contributing to an rise in output demand in the minimum area.Peoples are in the battle to harvest the most crops from their lands and that's why they started using industrial fertilizer that not only decreases soil productivity but also decreases crop nutrient quality. In eastern India, agriculture is the dominant industry (Hassen et al., 2019). It can be used as a biofertilizer by implementing microbial, physical and chemical treatment processes because of the abundance of biodegradable waste (from the agricultural activity).Biofertilizer production using biodegradable waste is increasing quite rapidly worldwide due to its possible use in many fascinating areas of agriculture and environment since it is one of the key components of integrated nutrient management as it is a cost-effective and safe source to substitute chemical fertilizers for sustainable farming (Vassilev et al., 2015). A bio fertilizer is a material containing living microorganisms that colonize the rhizosphere or the interior of plants when added to seeds or soil and encourage growth by increasing the host plant's supply or availability of primary nutrients. Due to its eco-friendly, easy to apply, non-toxic and cost-effective nature, organic fertilizers have emerged as a highly potent alternative to chemical fertilizers.Bio fertilizer development involves anaerobic treatment of bio waste with the enzymes or microbes in a appropriate climatic environment.Different bacteria are used for specific roles such as rhizobium, azotobactor, azospirillum etc. as nitrogen fixing Bacillus megaterium var.Phosphaticum, Bacillus subtilis Circulans, Pseudomonasstriata for phosphorus fixation in bio fertilizer development (Tsai et al., 2007).Soil is a suitable home with abundance with microorganisms useful for agriculture. Some soil microorganisms have the potential to consume and extract ambient nitrogen from to the plant to be readily accessible. While certain soil microorganism solubilizes most of the soil rendering them accessible to the plants (Chang et al., 2009). Each of these attributes render them essential for biofertilizer use.

II. DIFFERENT SUBSTRATE USED IN BIO FERTILIZER PRODUCTION

A. Bagasse

This is the substance which is the sugar industry's main byproduct.Bagasse 's annual supply is around 30 percent of the crushed sugar cane and about 90 percent of it is used as a fuel to generate the energy bagasse that is also generated in several other sources and is lost in generally burning and other operations.This bagasse is the rich source of organic matter and is very useful in fertilizer production so in our experiments we will use this bagasse as the feed for bio fertilizer development.Nowadays there is a lot of other innovation to use this bagasse for the processing of various kinds of materials such as coal, pulp, cloth, board and feed and nanocellulose etc (Inyang et al., 2010).

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B. Husks

Many of the pulses husk was discarded by citizens by burning in the atmosphere and this triggers the combustion of organic matter as well as the heavy urban air emissions and this emission creates other health issues.Large amounts of husks are burned every year in Punjab and Haryana and they have no choice other than that, if we take the husk from them and extract the fertilizer from it and sell it back to farmers, it would avoid the burning of husk in the atmosphere as well as monitor the usage of chemical fertilizer in the region (Pode et al., 2016).

C. Manure

Manure is organic matter often produced from livestock waste and can be used in cultivation as a bio fertilizer.Manures aim to restore soil productivity by introducing organic matter and minerals, such as phosphorus, found in the soil by microbes, fungi and other species.Over several years, livestock waste such as poultry manure and cow manure has been used as a nutrient over agriculture (Eghball et al., 2002).

D. Algae

Algae are plant or plant-like species producing chlorophyll and are the plants found in the green pigment.The chlorophyll can be removed from the atmosphere through sunlight and water which co2 and will generate glucose, and this cycle is called photosynthesis.This algae is abundant in nitrogen and when it is combined with nitrogen producing microbes and then added in the crop, it provides the soil with a large volume of nitrogen which keeps the microbes healthy for 6 months (Hoek et al., 1995).

E. Bone Meal

Bone meal (or bone manure) is a combination of finely ground meat bones and waste material from the slaughterhouse.This is used for plants as an agricultural nutrient and for livestock as a dietary substitute.Bone meal is primarily used as a source of phosphorus and calcium, as a slow-release fertilizer. Here fig 1 described the different raw materials for bio fertilizers production

Fig 1 : Substrate for bio fertilizer production

III. ROLE OF MICROBES IN BIO FERTILIZER PRODUCTION

There are different types of microbes used in production of bio fertilizer which is rich in different nutrients. This is arranged properly in the table:

A. Rhizobium

This is a form of nodulating micro-organism that symbiotically interacts with the legume plant core. This produces nodules and there it multiplies.It fixesatmospheric nitrogen by remaining inside thannodules. Only diazotrophs that contain the enzyme dinitrogenase can perform the actual process of dinitrogen fixation. Nitrogen is the most essential resource required to sustain the development of plants. Unfortunately, ambient dinitrogen (78% of the air we breathe) is highly fragile owing to triple bonds that can only be dissolved by energy-intensive processes.This involve electrical N2 fastening through lighting where N oxides fall to earth with air. The Haber-Bosch manages the development of agricultural fertilizers and the biological fixation of N2 by bacterial symbionts such as Rhizobium in legumes (Kniazeva et al., 2019).

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B. Azotobacter

It is a free-living bacteria that fixes nitrogen, exists freely in soil and multiples, uses organic matter as a source of carbon (energy) for its development and life.All of these Bio-fertilizers remove the ambient nitrogen by the biological nitrogen fixation cycle to make it conveniently assailable to accessible for planting. Diazotrophs such as rhizobium, azotobacter, and azospirillum are essential in this regard, as they enrich nutrition in N-deficient soils. Diazotrophs such as rhizobium, azotobacter, and azospirillum are essential in this regard, as they enrich nutrition in N-deficient soils. Azotobacter encourages the development of plants as well as the fixation of nitrogen (Boartseva et al., 2017).

C. Azospirillum

It is an associative form of microorganism which can colonize the plant's rootsurface.It lets plants get nutrient 'N' (Nitrogen) from the atmosphere by forming a symbiotic alliance tank.Azospirillum is the best characterized genus of rhizobacteria which promotes plant production.Certain free-living diazotrophs that have been found frequently in contact with plant roots are Acetobacterdiazotrophicus, Herbaspirillumseropedicae, Azoarcusspp and Azotobacter. Four forms of root contact between Azospirillum and the plant are highlighted: natural environment, plant root contact, nitrogen fixation and plant growth hormone biosynthesis.Biofertilizer-(Bacillus megateriumvar. phosphaticum, Bacillus subtilis Bacilluscirculans, Pseudomonas striata) for Phosphorous(P) solubilizingBacteria solubilizing phosphates (BSPs) are useful bacteria that can solubilize inorganic phosphorus from insoluble compounds (Fukami et al., 2018).

The rhizosphere microorganisms' capacity to solubilize P is known to be one of the most significant characteristics correlated with plant phosphate feeding.PSB was added as phosphate biofertilizer to the Agricultural Society.Phosphorus (P) is one of the main important plant macronutrients and is added in the form of phosphate fertilizers to soil.However, a large portion of inorganic soluble phosphate that is applied to the soil as a chemical fertilizer is rapidly immobilized and becomes available to plants.The main purpose of soil phosphorus management at the moment is to optimize crop production and minimize P loss from soil.As soil inoculums, PSBhave attracted the attention of farmers to increase plant growth and yield.Using PSB with rock phosphate will save about 50 percent of phosphaticfertilizer 's crop requirement. Using PSB as inoculants improves plant uptake P.Simple inoculation of PSB seeds offers crop yield responses equal to 30 kg P2O5 /ha, or 50% of the phosphatic fertilizer requirement (Pereg et al., 2016).

D. Potassium fixing Biofertilizer- (Frateuriaaurantia)

K is a bio-fertilizer dependent on Frateuriaaurantia, a specific strain of potash solubilizing, beneficial bacteria.This comes in liquid format (1x 10 ^ 9 bacterial cells / ml).In a number of other processes, potassium is also essential that contributes to growth and development.Potassium is sometimes referred to as the "price factor," owing to its contribution to many of the price-associated characteristics, such as scale, form , color, and even taste (Lidor et al., 2019).

NAME	CROP SUITED	BEFEFITS SEEN
Rhizobium strains	Generally found in pulses and soya beans.	10-35% yield increase, 50-200 kg N/ha
Azospirillum	Non-legumes like oats, maize, barley, sorghum, millet, Sugarcane, rice, wheat etc.	10-20% yield increase
Azotobacter	Soil treatment for non- legume crops including dry land crops Mustard, sunflower, grapes, papaya, water melon, tomato, ladyfinger, Banana, Sugarcane, coconut, spices, fruits, flowers, plantation crops.	10-15% yield increase- adds 20-25 kg N/ha
Phosphate Solubilizers* (*there are 2 bacterial and 2 fungal species in this group)	Applicable in every types of crops	5-30% yield increase
Microhizae (VAM)	Many trees, some crops, and some ornamental plants	30-50% yield increase, enhances uptake of P. Zn, S and Water

Table 1: List of microbes applicable for biofertilizer production (Kniazeva et al., 2019; Boartseva et al., 2017; Pereg et al., 2016; Lidor et al., 2019)

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IV. EFFECT OF NITROGENOUS BIOMASS AND BIOFERTILIZERS ON THE SOIL

Hatch et al. (2000) had measured both the activity of microbes as well as gross rate of soil process using two different samples of grazed soil of permanent pasture which was changed by the help of nitrogen fertilizers or dung. Many studies was done taking rate as a primary factor which shows gross immobilization was less and mineralization of nitrogen was found to be higher on fertilized soils as compare to unfertilized soil. In both the cases content of microbial biomass in the soil was same but the biomass activity was found to be higher in the fertilized soil. The gross mineralization, activity and biomass size was not going to be effected by short term amendments done like addition of nitrogen to the soil or holding of nitrogen from previous one growing seasons. Application of dung slightly effects the mineralization and directly affects the microbial parts of the soil by increasing the composition of carbon. Whereas the application of nitrogenous fertilizers increases the composition of carbon which results the increase in growth of the plants.

Kushwaha et al. (2000) studied the ongoing conditions of dryland agro-ecosystem, were preparation of farming lands more than the requirement causes biological degradation of soil which finally reduces the production. In this study they setup various experiments on variations of organic input by the help of agricultural residue collected from various crops to crops. They constructed a random block design with three different replicates along with six different treatments during the time of harvesting time of rice. Same set of experiments was repeated for barley which was cropped just after harvesting the rice. In these set of experiments the composition of microbial biomass (carbon and nitrogen) was calculated six times which includes seedling, grain forming, as well as maturity stages of rice and barley crops. The range of available nitrogen varies in rice from 7.8 to 16.6 µg g-1 and in barley from 7.8 to 15.6 µg g-1. This valuable data shows the direct effect of agricultural residue which increases the concentration of microbial biomass as well as drops a bright light on nitrogen cycling processes of upcoming crops (Kushwaha et al., 2000).

Witt et al. (2000) conducted an experiment in order to observe different effects of nitrogen and carbon source on flooded soil. The effect was studied on soil organics which can be extracted, microbial biowaste of the soil and many more. Four different root was taken for observation which includes rice straw, prilled urea, rice straw and prillled urea combined and *Sesbaniarostrata*as bio fertilizers. The effect of microbial soil bio waste on the plant was quite small and the application of nitrogen source results a rapid hike in microbial soil biowaste for 10 days after the transplantation and then a decrement for 73 days. These results explain that microbial soil biowaste gone through a conversion to a sink from source of soil nitrogen across the growth period (Witt et al., 2000).

Belanger and Richards (2000) studied the effects of fertilization of nitrogen on the biomass dynamics as well as its accumulation across first regeneration of alfalfa during proceeding years. Along with this the main aim was to determine whether the sample will explain critical content of nitrogen which can set a standard data for proceeding years. The experiment was divided into two different section for two different years in which biomass and accumulation of nitrogen of alfalfa grown was observed at three different rates (80, 40, 0 Kg N/ ha) in a week. The best shoot growth was obtained with 40 kg N/ ha in year 1992 whereas this results was not observed in year 1993. After the defoliation occurs demands of nitrogen gets increased and both reserves root nitrogen as well as inorganic soil nitrogen cannot fulfill the demands. This study concludes that requirements of nitrogen as per biomass of shoot was found to be greater in proceeding year as compare to establishing year which results larger leaves in seeding year (Belanger and Richards, 2000).

Virtanen et al. (2000) performed the study in order to establish a relation between pH of the soil, richness of the species, nutrient applications and biomass of vascular plants with bryophyte biomass. Bryophytes belong to a plant category and classified into three different sections, liverworts which are non vascular land plants, mosses and hornworts. At the lowest value of pH in the performed experiments (3.3-4.5), presence of bryophytes increases from the extent which was likely to be present as well as the mosses disappear virtually. As per the observed study the variation between species richness and bryophytes biomass was curvilinear with respect to pH of the soil. The effect of nitrogen was found to be negative on bulk quantity of *Brachytheciumrutabulum.* Variation in the factors of the soil such as potassium, phosphorus and pH effectively shows a great response on *Eurhynchiumpraelongum* and this species got affected by nitrogen (Virtanen at al., 2000). Malik et al. (2001) performed an experiment for utilization of lignocellulosic biomass for production of nitrogenous bio fertilizer. In this study bagasse, the waste of sugar industry was used as a substrate of the production of biofertilizer. Isolation of nitrogen fixing microbes was done from the samples of the soil collected from the fields. Azotobacterchroococcum was selected because of its nature of surviving and fixing of nitrogen up to great extent as compare to other microbes present in the soil as per the testing done on the bagasse. In the experiment *Azotobacterchroococcum, Azospirilumbrasilense* and *Azotobacterindicus* was used as the microbes for nitrogen fixing from 7 to 28 days. The nitrogen fixed by the help of these microbes was 67.81, 43.20 and 28.00 mg/l. Accroding to the observed results application of Azotobacterchroococcum and bagasse gives the best production of nitrogen (Malik et al., 2001).

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Zhang et al. (2001) observed the effects variations in the quantity of nitrogenous and phosphorous fertilizers on soil microbial nitrogenous biomass, carbon biomass and phosphorous biomass. This experiment was done in the fields during the periods of production of corns. The observed results explains that both the nitrogenius and phosphrous fertilizers did not not plays a significant role on microbial carbon biomass. It was also observed that application of nitrogenous fertilizer about 225 kg/hm2 causes reduction in the microbial nitrogen biomass. When the phosphorous fertilizers was applied in same quantity of 225 kg/hm2 also effects the microbial phosphorous biomass up to some extent (Zhang et al., 2001)

Irisarri et al. (2001) studied the behavior of cyanobacteria in maintaining the fertility of the flooded rice fields. The experiment was performed in the laboratory in order to analyze the ability of cyanobacteria, a nitrogen fixing microbes as a natural organic fertilizers in the rice field.

Determination of both diversity as well as population density of heterocystous cyanobacteria was carried out as it got effected by application urea as a fertilizer. 1.6×10^4 CFU m^{-2} cyanobacteria was found after flooding at the control 8 weeks. *Anabaena* and *Nostoc*was about 90 % of total heterocystous cyanobacteria found after the treatment process. For more clearance as well as understanding of environmental factors, two of the cyanobacterias was isolated and were tested. The test was performed for the testing the tolerance capacity of isolates towards two herbicides and combined nitrogen and in both the isolates 0.2 mM ammonium inhibited nitrogenase activity after 24 h of culture (Irisarri et al., 2001).

Hoque et al. (2001) performed a experiment in order to determine the effects of free air carbon dioxide enrichment and ambient co2 on soil microbial biomass carbon and activity of nitrogen fixation at three different steps. Rice plants were grown and nitrogenous fertilizers was applied in three different level (low, medium and high; 4, 9, 15 g Nm-2) under both condition of carbon dioxide. Soil sample was collected at four different time for analysis of nitrogen fixation as well as microbial biomass carbon. The biological nitrogen fixation in the soil was found greater when treated with free air carbon dioxide enrichment as compare to ambient co2. In case of microbial carbon biomass the amount was slightly greater in case of free air carbon dioxide enrichment as compare to ambient c02.

This study concludes that application of free air carbon dioxide enrichment plays a positive role specially in case of enrichment of microbial nitrogen biomass and microbial carbon biomass as compare to ambient co2 (Hoque et al., 2001)

Shen et al. (2001) had conducted a set of experiments to calculate the variation in both quality and the quantity of nutrition in the soil after 10 to 13 years of continuous maize and wheat crops rotation. Farmyard manure NPK fertilizer and farm manure increases the mineralizable carbon, microbial biomass carbon as well as oxidizable carbon too. Whereas the application of NPK fertilizers increases only microbial biomass carbon and mineralizable carbon and decreases oxidizable carbon. Characterization using 13 carbon nuclear magnetic resonance clearly shows the effects caused by the fertilization on both carboxylic carbon as well as long chain carbon (34- 51%). Carboxylic carbon and carbohydrate carbon was directly related with mineralizable carbon, microbial biomass carbon as well as oxidizable carbon, whereas negatively related with unoxidizable carbon and long chain carbon (Shen et al., 2001).

V. FUTURE PROSPECTIVE AND CONCLUSION

The application biowaste for the production of biofertilizers is one of the best way to control the environmental pollution by reducing the usage of chemical fertilizers as well as by reducing the biodegradable waste. Biological methods for conversion of lignocellulosic biomass to bio fertilizers is now a day most attractive scope of research as it involves government by issuing many supportive energy policies.

This chapter has focused on different raw materials as well as the microorganism which can be used in bio fertilizers production. This chapter also explained the advantage of application of nitrogenous rich bio fertilizers in the soil too by the help of different old studies. This is a developing fields of research and consistent extensive studies are needed towards the complete utilization and quantification of biodegradable waste to bio fertilizer production. The abundance of lignocellulosic biomass and the developing technology in order to utilize the organic matter present for production of biofertilizers by the help of different technique and application of microbes may play a significant role in reduction of cost of biofertilizer production.

VI. ACKNOWLEDGEMENT

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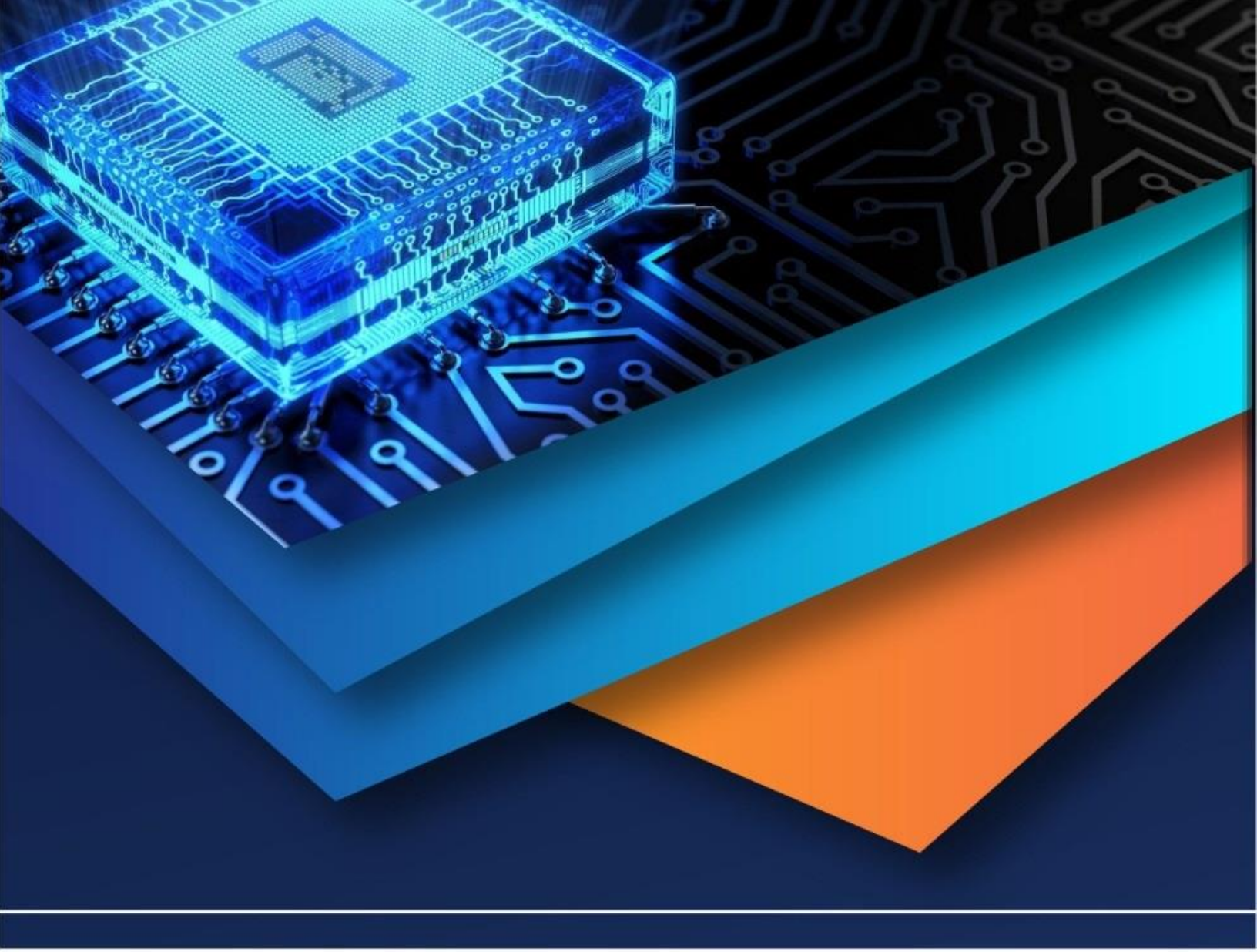

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