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Algae Culturing from Wastewater Samples and Captured Exhaust Gas for Sustainable Fish Cultivation

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Abstract: **Wastewater and exhaust gases from diesel engines are significant environmental pollutants, contributing to nutrient overload and greenhouse emissions. However, these waste products offer a potential solution for nutrient recycling and fish feed production. This study explores the sustainable cultivation of algae using wastewater and captured diesel exhaust gas from diesel generators for fish feed production, addressing environmental and economic challenges in aquaculture. Wastewater from fishponds and food processing industries, along with exhaust gas from diesel generators, were analyzed for their physicochemical and biological properties. Algae species such as Chlorella vulgaris and Cyclotella sp. were successfully cultured in the wastewater enriched with exhaust gas, demonstrating high nutrient content suitable for fish feed. African catfish (Clarias gariepinus) fed the algae-based diet exhibited superior growth (25 cm final length, 0.8 kg weight gain) and lower mortality (12.5%) compared to ca control group fed conventional feed (22 cm, 0.5 kg, 25% mortality). Hematological analysis indicated enhanced immune response in algae-fed fish, though slight liver and kidney stress was observed. The results confirm the feasibility of repurposing wastewater and exhaust gas for algae production, offering a sustainable alternative to conventional fish feed while mitigating environmental pollution. This approach supports resource efficiency and food security, aligning with sustainable goals. Further optimization of the algae culture process could enhance its scalability and minimize metabolic stress in fish.**

Keywords: *Algae; Wastewater; Exhaust Gas Capture; Fish Farming; Sustainability.*

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I. INTRODUCTION

Wastewater encompasses water that has been utilized in domestic, industrial, or agricultural activities, retaining residual contaminants such as organic matter, inorganic chemicals, pathogens, and viruses from prior use. Primary environmental concern lies in the nutrient overload within wastewater, particularly nitrogen and phosphorus compounds, which drive eutrophication in aquatic ecosystems [1].

Exhaust gas from a diesel engine is a complex combination of gases and particulates formed during the combustion of diesel fuel. Nitrogen oxides (NOx), carbon monoxide (CO), and volatile organic compounds (VOCs) are just some of the contaminants that may be found in this mixture. (VOCs). Because of their links to respiratory difficulties, cardiovascular illness, and cancer, NOx and PM are of special concern [2].

The local fish growing technique is hindered by the high cost of fish feed because of importation and raw material purchase. This limits the productivity and profitability of the fish farmers, who are often small-scale producers with limited resources. Readily available product will enhance the fish farming by reducing the input costs and increasing the output quality and quantity. This will contribute to the SDG 2: Zero Hunger by improving the food security and nutrition of the local population, as well as promoting sustainable agriculture that uses natural resources efficiently and responsibly.

Microalgae and cyanobacteria (photosynthetic microorganism) with high growth rates are identified as microorganisms with carbon fixation rates higher than those of terrestrial plant [3]. These photosynthetic microorganisms utilize solar light as their main source of energy and are superior in productivity compared with terrestrial plants and could be cultivated in harsh environmental conditions inappropriate for agricultural activities [4].

This research aims to capture and utilize generator exhaust gas and wastewater for algal culture towards possible fish feed production by; capturing and analyzing the exhaust

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gas from diesel engines, collecting wastewater samples from fishpond and food manufacturing for laboratory analysis (physicochemical and biological, culturing algae from the wastewater samples collected using suitable captured exhaust gas, and cultivating African Catfish in the wastewater with the algae culture using exhaust gas and a controlled system using conventional fish feed from the Port Harcourt metropolis.

II. MATERIALS AND METHODS

➢ *Sample Collection*

Sample collection involved a systematic approach to ensure the acquisition of appropriate and representative samples. The following steps were followed to obtain the necessary samples for the experiment:

- *Exhaust Gas Capture for Laboratory Analysis*

The sample was collected from three different sources in the University environment. The exhaust gas was directed into the balloon using appropriate connectors (see Figure 1 below), ensuring a secure and airtight connection. A 1000-liter capacity biobag was utilized to capture and store more of the exhaust gas for further use (Figure 2).

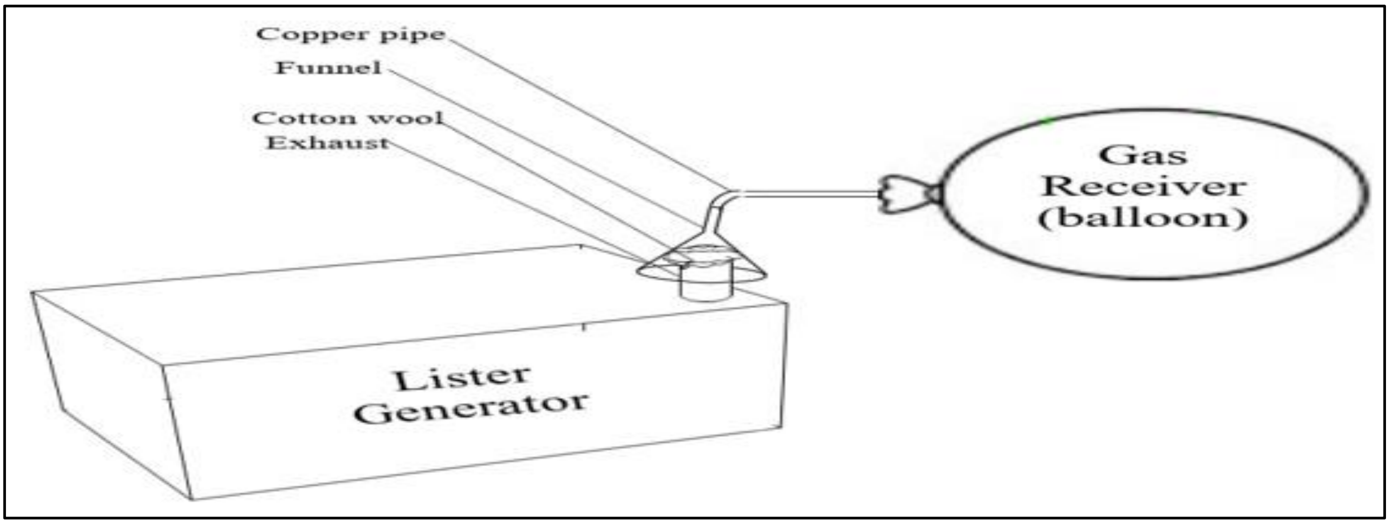

Fig 1 Exhaust Gas Capture for Gas Analysis

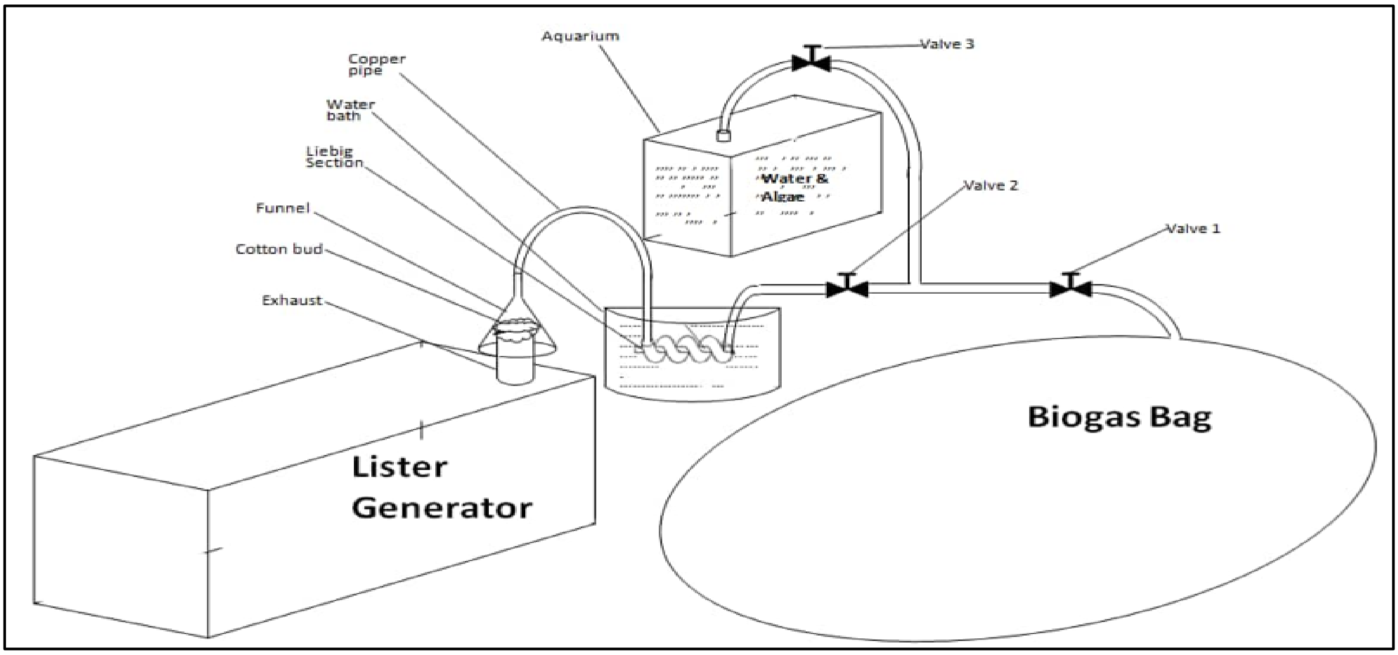

Fig 2 Exhaust Gas Capture Process for Algae Culture

- *Wastewater Sample Collection*

Wastewater was collected from two different sources: farming industry; and food processing and manufacturing industry.

Samples were collected using a sterile sampling approach. Containers of 25 litres gallon water containers and 1.5litre, were used to avoid any potential contamination.

➢ *Wastewater and Exhaust Gas Laboratory Analysis*

- *Exhaust Gas Analysis*

Agilent 6890N Gas Chromatograph with Agilent 5975 Mass Selective Detector were used. Physico-chemical and biological anakysis of wastewater were carried out to identify:

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/25may940

Phosphate - Acid hydrolysable, Total & Organic phosphate, following APHAAWWA-WPCF (1980). The determination was made by Vanadomolybdo phosphoric acid method. Amount of phosphorus/L was calculated by using following formula:-

$$\text{Phosphate mg/L} = \frac{\text{mg P x 1000}}{\text{ml sample}} \quad (1)$$

Sulphate – Determined using Turbidimetric Method following APHA-AWWA-WPCF (1980). The quantity of sulphate was calculated in mg/L by using calibration curve.

$$\text{Sulphate mg/L} = \text{mgSO}_4{}^{2-} \times 1000 \quad (2)$$

PH – measured with a pH meter.

Turbidity - Determined using a standardized Hanna H198703 Turbidimeter.

Total dissolved solids (TDS) – Determined by gravimetric method. the total dissolved solids content of the water was calculated:

$$\text{Total dissolved solids}\left(\frac{\text{mg}}{\text{l}}\right) = \frac{[(\text{W2}-\text{W1})\text{ mg x 1000}]}{\text{ml of filtrate used}} \quad (3)$$

Nitrite – Determined using Strickland and Parsons 1968 method. The nitrite concentration is calculated by:

$$\mu\text{M NO2} = \text{Corrected absorbance} \times \text{F} \quad (4)$$

$$\text{F} = (20\text{mmol/kg})/(\text{Es} - \text{Eb}) \quad (5)$$

Where;
20 µmol/kg = conc of standard

Es = mean absorbance of the standards

Eb = mean absorbance of the blank

Nitrate – Determined using Brucine Sulphate method.

Salinity – Determined by evaporation.

$$\text{Salinity} = \frac{\text{weight of salt}}{\text{weight of water}} \times 1000 \quad (6)$$

Faecal Coliform – The colonies were cultured on M-FC agar for 24 hrs. The colonies were then counted as colony forming unit per 100ml.

Salmonella – Cultured in 50 ml of Rappaport-Vassiliadis broth (RVB). Colonies were then counted as colony forming unit per 100ml.

Total Coliforms - The colonies were cultured on M-FC agar for 24 hrs. The colonies were then counted as colony forming unit per 100ml.

Escherichia coli – The colonies were cultured in the MI agar and TSA plate. The colonies were then counted as colony forming unit per 100ml.

- *Atomic Absorption Spectrometry (AAS)*

➢ *Algae Culture.*

In this phase of the research, the experimental setup for cultivating algae is depicted in Figure 2 and 3 (provided below).

Fig 3 Algae Culture Process

The exhaust gas, a critical component, undergoes a cooling process to lower its temperature before passing through a gas filter equipped with cotton wool (see Figure 4 below), aimed at eliminating soot from the gas stream. Subsequently, the treated gas is directed into a 1000L biogas bag for efficient storage. The wastewater, housed in an aquarium, is then infused with the gas from the bag. This comprehensive process extends over one month for each wastewater sample.

Regular monitoring was conducted, with weekly readings taken for key parameters including electrical conductivity, salinity, pH, and turbidity, for every wastewater sample involved in the experimental procedure. The primary objective of this initial phase is to identify potential algae species that thrive in the wastewater, utilizing exhaust gas.

At the conclusion of the one-month period, the wastewater containing the cultivated algae species undergoes analysis in the Microbiology Laboratory of the University of Port Harcourt.

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Fig 4 Filter for Sooth and Moisture from Exhaust Gas Stream

- *Algae Species Identification*

Three samples, collected from distinct positions within the wastewater system (both fishpond WW and food processing WW), were submitted to the Department of Microbiology at the University of Port Harcourt for comprehensive analysis aimed at identifying the algae species present or developed during the culture process. This analytical approach was instrumental in assessing the abundance of each algae species, thereby providing insights into the suitability of the wastewater for fish farming

➢ *Fish Cultivation*

- *Selection of Fish :*

Sixteen fingerling cat fishes (Clarias gariepinus) of similar size and weight (1.6 kg each) and age (3 weeks old) were selected. The fingerlings were divided into groups, Group A (experimental) and Group B (control), each containing eight fishes.

- *Feeding Prootcol :*

Group A (Experimental group) was fed the CO_2-enriched algae three times a week, specifically at 11:00 AM on designated feeding days.

Group B (Control group) was fed a handful of commercially available fish feed daily.

- *Experimental Conditions:*

Both groups were maintained under controlled environmental conditions:

Temperature: 25°C ± 1°C

PH: 7.0 ± 0.2

- *Post-Experiment Analysis:*

After completing the feeding regimen, both groups of fish were taken to the laboratory for further analysis.

III. RESULTS AND DISCUSSION

➢ *Gas Analysis*

Comparative analysis of Gas Components

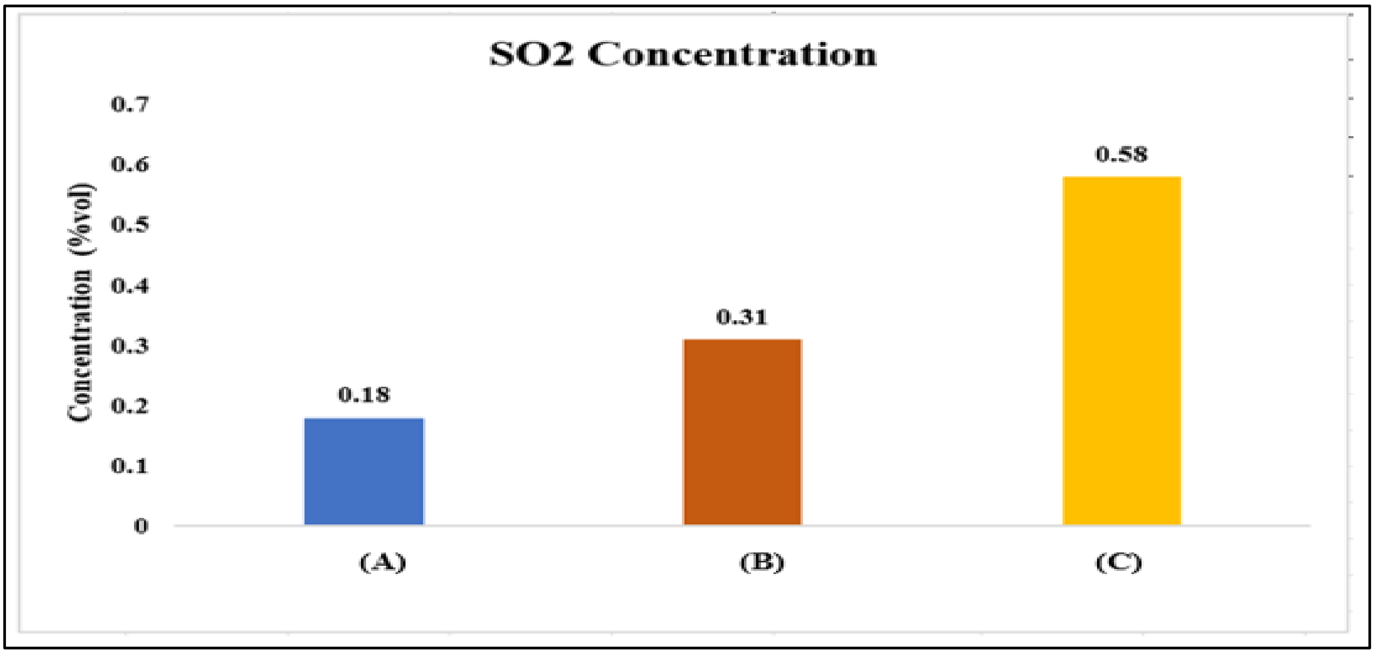

Fig 5 Comparison of SO2 in Samples A, B, and C

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Sulphur dioxide (SO2) is a gas produced primarily by the combustion of fuels containing sulphur impurities, such as diesel fuel with higher sulphur content.

The variations in SO_2 concentration among the samples could be attributed to differences in the sulphur content of the diesel fuel used.

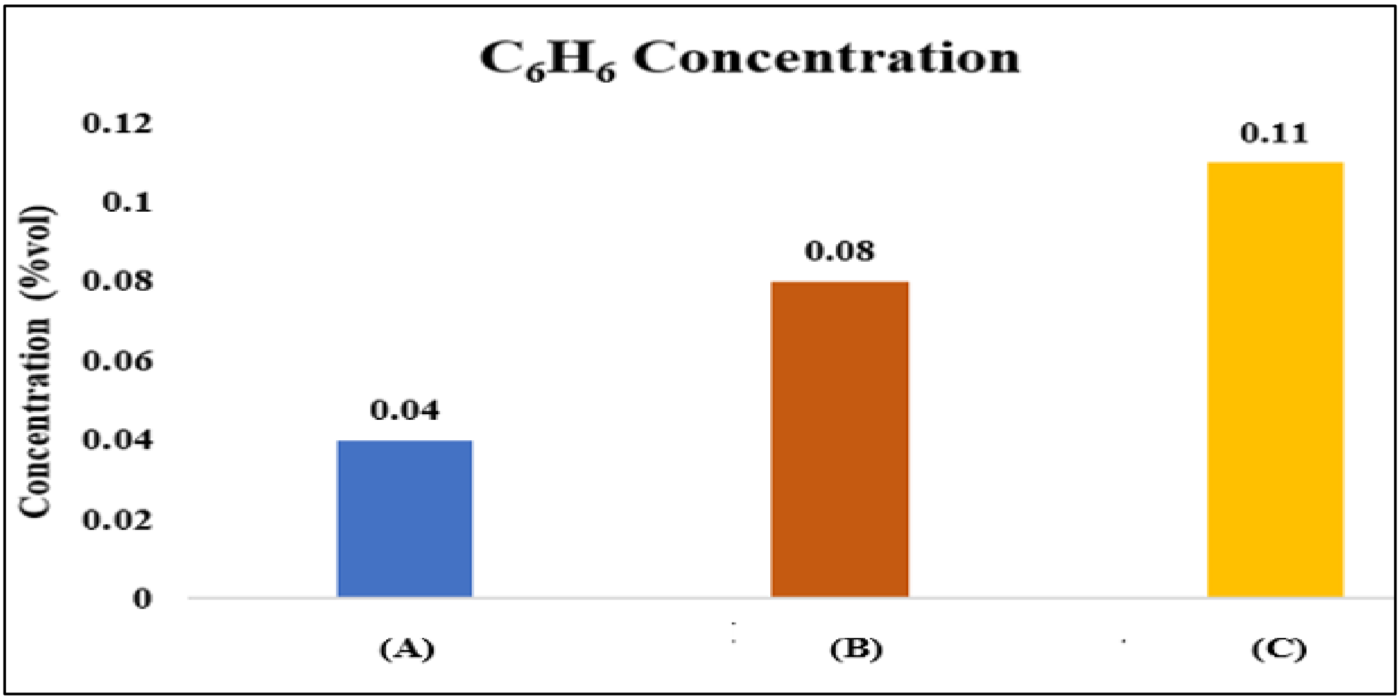

Fig 6 Comparison of C6H6 in Samples A, B, and C

Benzene is not a common component of diesel engine exhaust gas. Its presence in the exhaust may be due to trace amounts of benzene present in the diesel fuel itself, or it could be from other sources unrelated to the diesel generator.

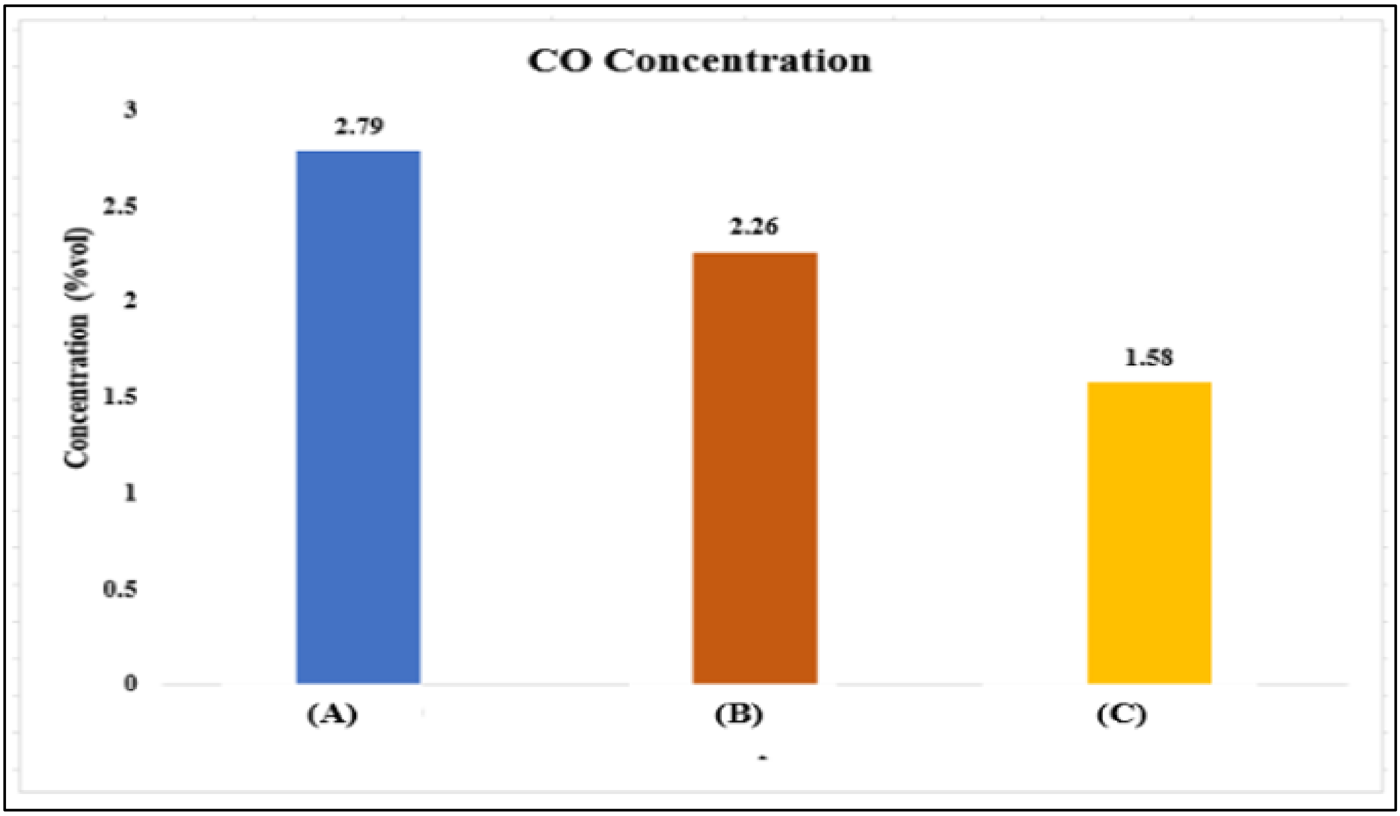

Fig 7 Comparison of CO in Samples

Carbon monoxide (CO) is a byproduct of incomplete combustion in the diesel engine. It forms when there is insufficient oxygen for the complete oxidation of carbon in the fuel. High levels of CO emissions can be particularly dangerous in enclosed spaces and can cause carbon monoxide poisoning, which can be fatal.

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/25may940

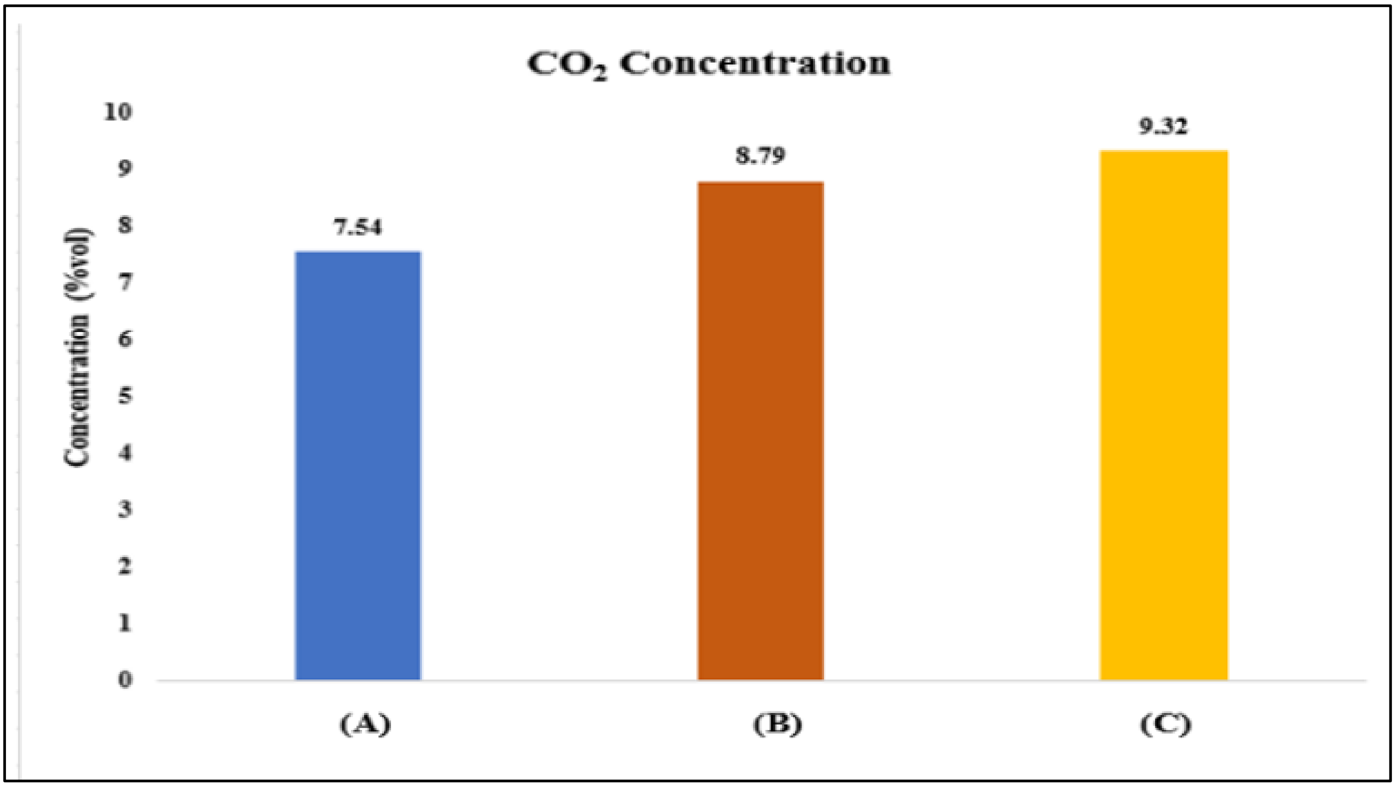

Fig 8 Comparison of CO2 in Samples

Carbon dioxide (CO_2) is the main product of complete combustion of carbon-containing fuels, including diesel. When carbon in the diesel fuel is fully oxidized with oxygen from the air, CO_2 is formed. Carbon dioxide is a greenhouse gas responsible for global warming and climate change.

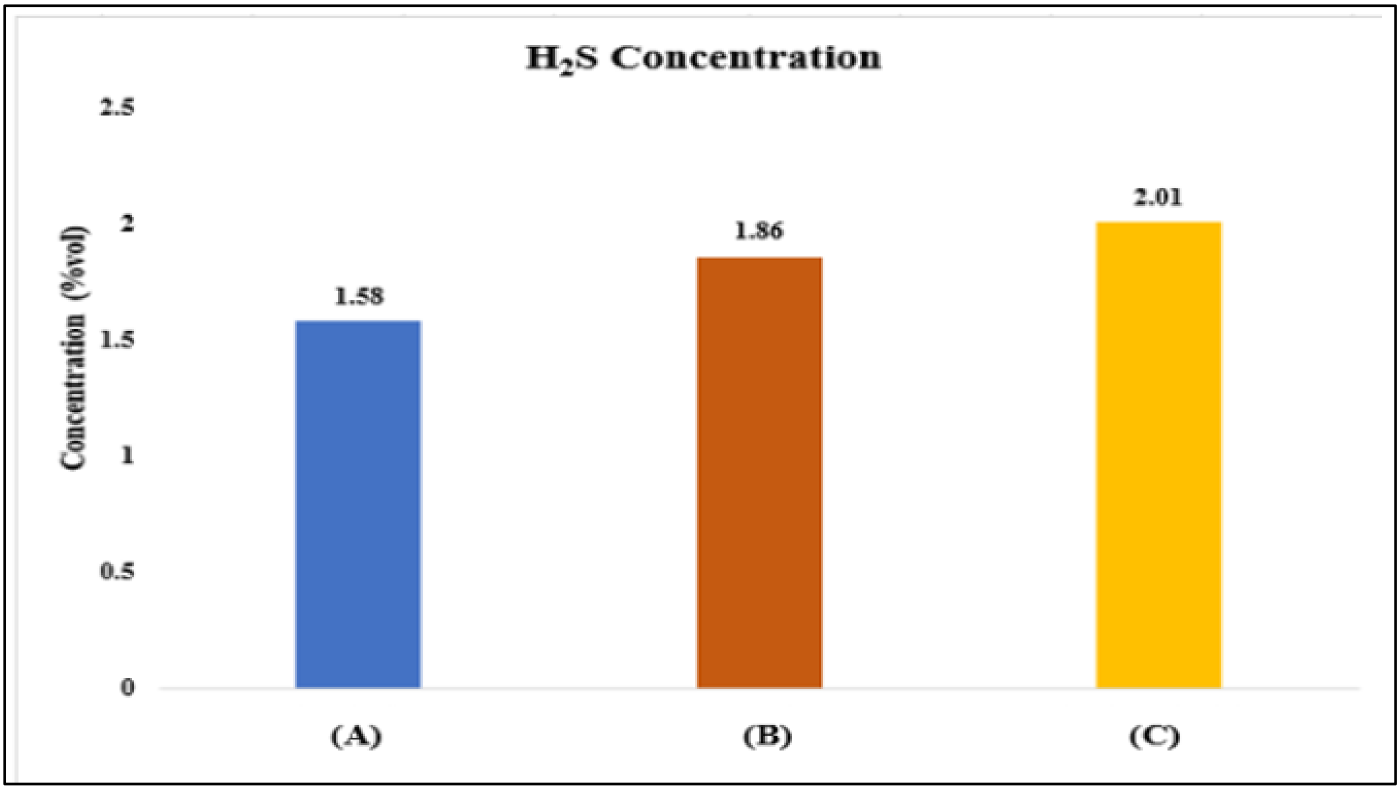

Fig 9 Comparison of H_2S in Samples

Hydrogen sulphide (H_2S) is not a typical component of diesel engine exhaust gas. Its presence in the samples may indicate the combustion of diesel fuel with a higher sulphur content or could be influenced by other sources unrelated to the diesel generator. Hydrogen sulphide is a toxic gas with a distinctive "rotten egg" odour. It can cause respiratory irritation and other health issues in humans.

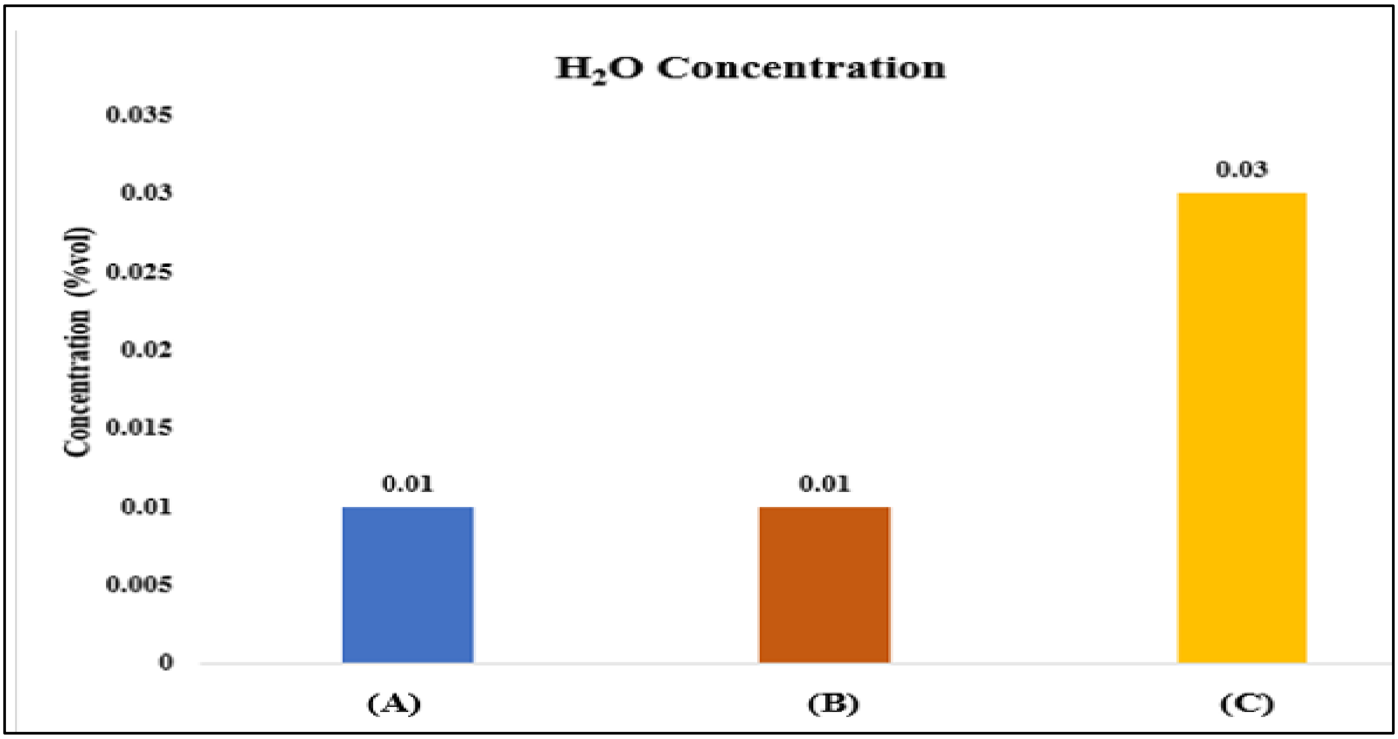

Fig 10 Comparison of H2O in Samples

Water vapor (H_2O) is produced as a byproduct of the combustion process in diesel engines when hydrocarbons in the fuel are burned with oxygen from the air. Water vapor is a natural component of the atmosphere, and its presence in exhaust gases is not a significant environmental concern.

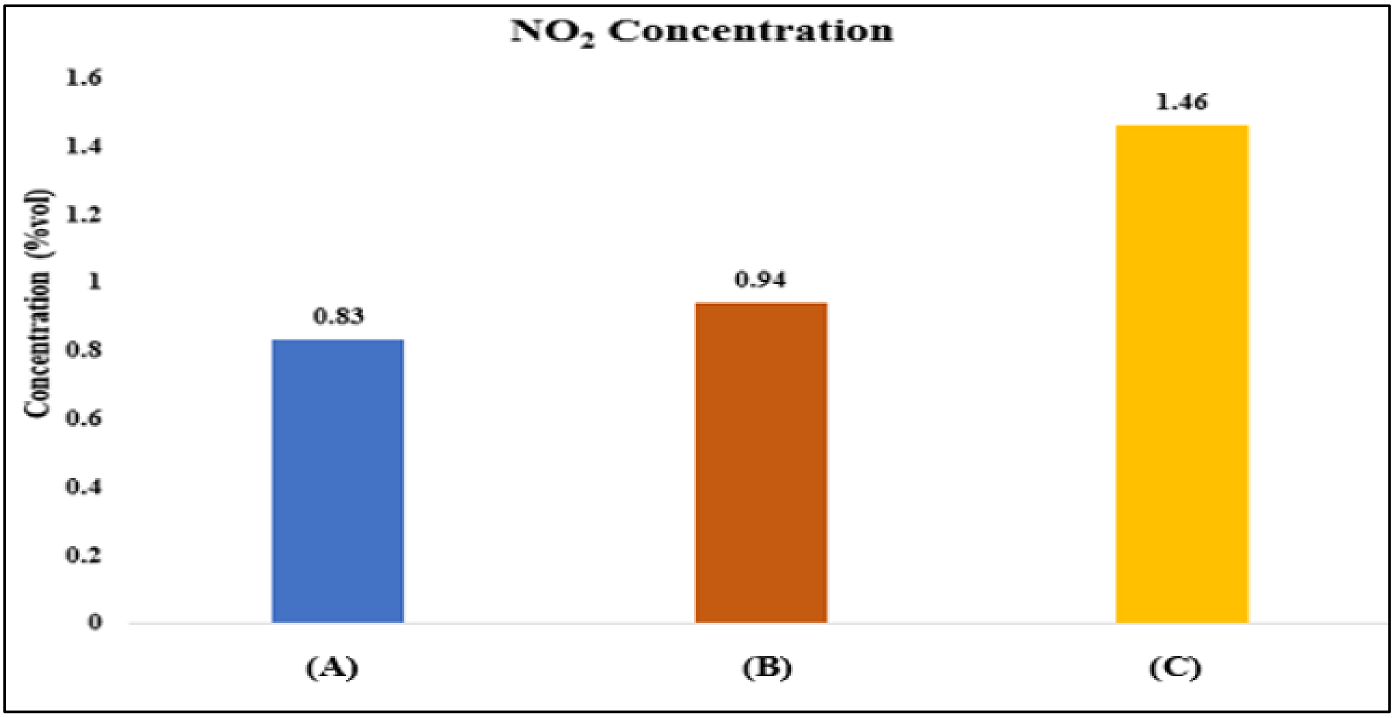

Fig 11 Comparison of NO2 in Samples

Nitrogen dioxide (NO_2) is formed when nitrogen oxides (NOx) react with atmospheric oxygen during high-temperature combustion processes. Nitrogen dioxide is a harmful air pollutant that contributes to the formation of ground-level ozone and particulate matter, which can degrade air quality and negatively impact human health.

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/25may940

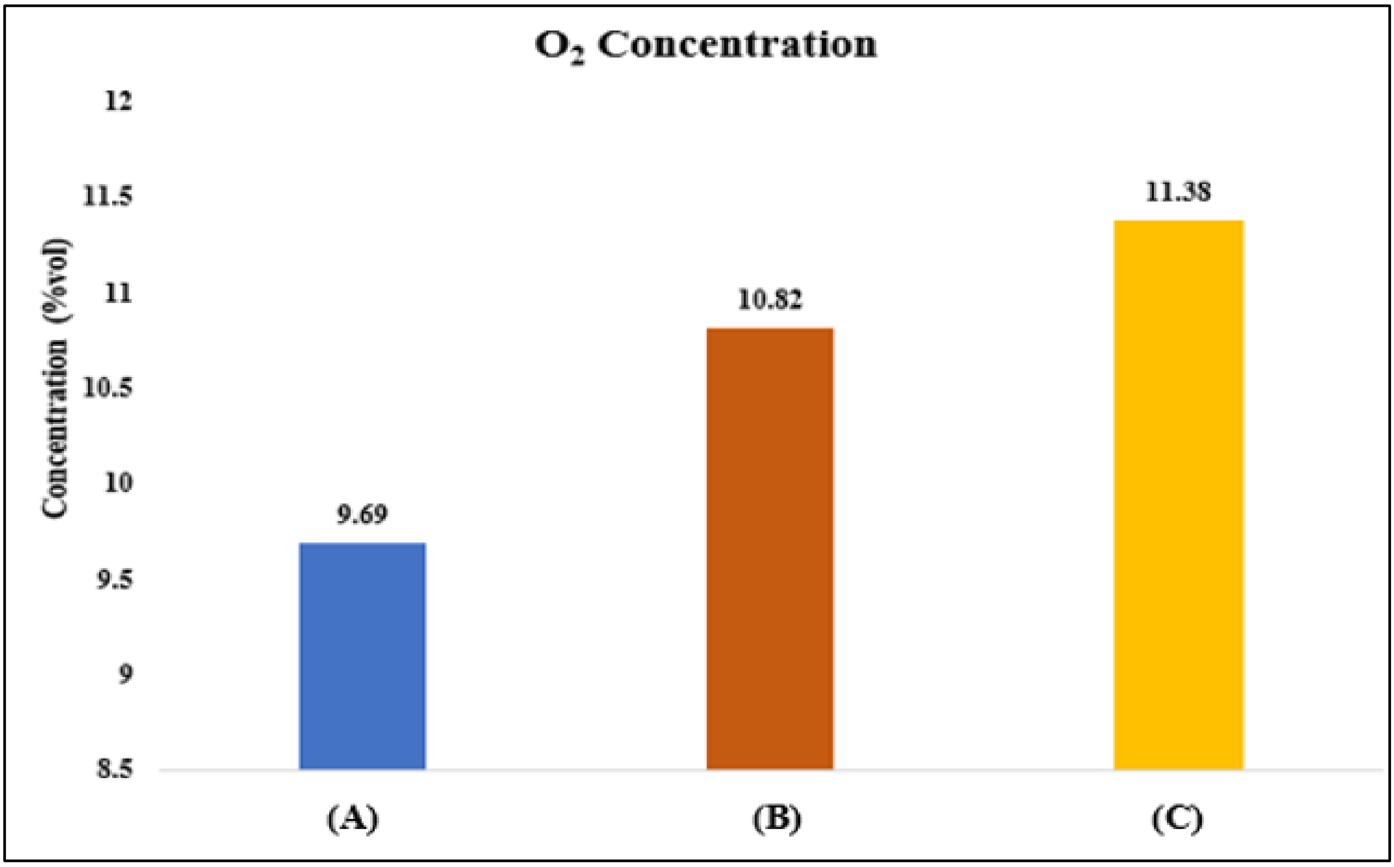

Fig 12 Comparison of O2 in Samples

Oxygen (O_2) is required for the combustion process in diesel engines. The presence of oxygen in the exhaust gas is an essential component for the combustion reaction to take place.

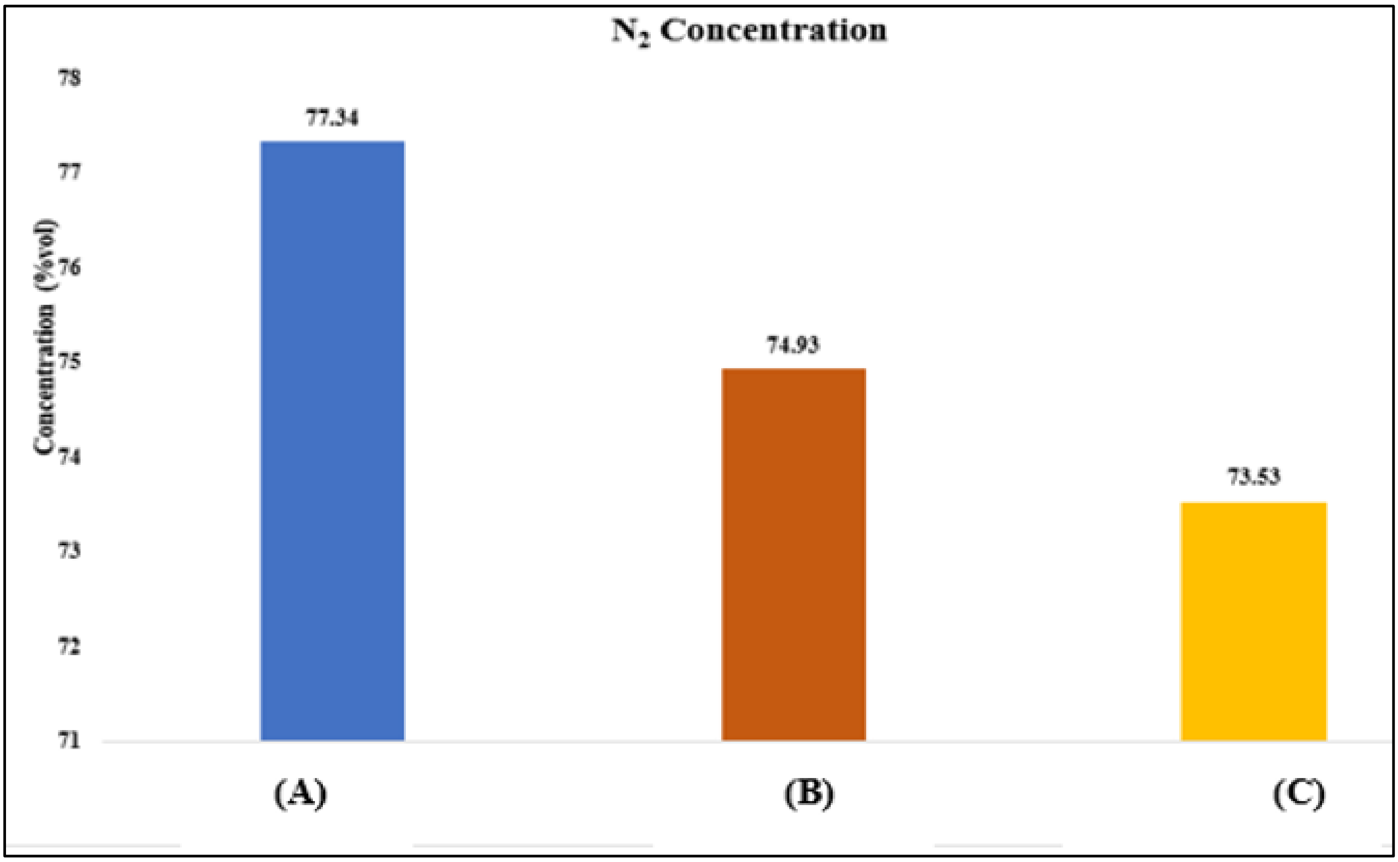

Fig 13 Comparison of N2 in Samples

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Nitrogen (N2) is the major component of the Earth's atmosphere and makes up the largest portion of the exhaust gases from combustion processes. Nitrogen gas (N2) is relatively inert and does not have direct harmful effects on the environment. However, during high-temperature combustion in the engine, some nitrogen reacts with oxygen to form nitrogen oxides (NOx), which are air pollutants.

➢ *Wastewater Analysis*

Table 1 Wastewater Analysis for Fish Pond

S/N	Sample	Wastewater (pond)	WHO Standard
1	pH	6.94	6.50 – 8.50
2	TSS (mg/l)	3.78	500
3	TDS (mg/l)	530.62	500
4	Turbidity (NTU)	13.59	<10
5	Nitrate (mg/l)	27.13	50
6	Nitrite (mg/l)	11.86	3
7	Phosphate (mg/l)	0.135	0.03 – 2.0
8	Sulphate (mg/l)	52.19	500
9	Salinity (ppt)	13.48	25.35
10	Lead (Pb) (mg/l)	3.63151	0.01
11	Chromium Cr (mg/l)	11.7587	0.05
12	Calcium Ca (mg/l)	142.38	250
13	Magnesium Mg (mg/l)	57.04	250
14	Copper Cu (mg/l)	8.31306	20
15	Iron Fe (mg/l)	0.52784	0.3
16	Arsenic As (mg/l)	1.27413	0.01 0.05
17	Mercury Hg (mg/l)	0.04392	0.001
18	Nickel Ni (mg/l)	4.82101	0.02
19	T. Coliform (Cfu/ml)	2.74E+04	Nil
20	Faecal coliform (Cfu/ml)	1.83E+03	Nil
21	E. coli (Cfu/ml)	1.91E+03	Nil
22	Salmonella (Cfu/ml)	1.33E+03	Nil

Table 2 Wastewater Analysis for Food Manufacturing

S/N	Sample	Food Processing WW	WHO Standard
1	pH	6.28	6.50-8.50
2	TSS (mg/l)	25.17	500
3	TDS (mg/l)	270.42	500
4	Turbidity (NTU)	3.86	<10
5	Nitrate (mg/l)	64.52	50
6	Nitrite (mg/l)	10.73	3
7	Phosphate (mg/l)	1.546	0.03-2.0
8	Sulphate (mg/l)	430.17	500
9	Salinity (ppt)	19.64	25-35
10	Lead (Pb) (mg/l)	5.21841	0.01
11	Chromium Cr (mg/l)	13.50385	0.05
12	Calcium Ca (mg/l)	170.48392	250
13	Magnesium Mg (mg/l)	115.67431	250
14	Copper Cu (mg/l)	8.91053	2
15	Iron Fe (mg/l)	5.73522	0.3
16	Arsenic As (mg/l)	1.10548	0.01-0.05
17	Mercury Hg (mg/l)	0.00371	0.001
18	Nickel Ni (mg/l)	2.18539	0.02
19	T. Coliform (Cfu/ml)	1.13E+2	Nil
20	Faecal coliform (Cfu/ml)	10	Nil
21	E. coli (Cfu/ml)	54	Nil
22	Salmonella (Cfu/ml)	35	Nil

➢ *Nutrient Levels*

Both sources have elevated nitrate and phosphate levels, which could promote algae growth. However, excessive nutrients may lead to algal blooms, affecting water quality and oxygen levels for fish.

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/25may940

➢ *Metal Contaminants*

The presence of metals such as lead, chromium, and copper in both sources could have toxic effects on algae and fish. Regular monitoring and mitigation measures are essential.

➢ *Microbial Content*

High microbial content in fish pond wastewater may pose a risk to fish health. Food manufacturing wastewater has lower microbial content, but the presence of coliforms suggests the need for proper treatment.

➢ *PH and Turbidity*

Both sources have pH within an acceptable range. Turbidity in fish pond wastewater may impact light penetration for algae photosynthesis, while higher turbidity in food manufacturing wastewater indicates the need for filtration.

➢ *Overall Impact*

Both sources may provide nutrients for algae growth, but the high microbial content and metal concentrations in fish pond wastewater may pose challenges

➢ *Weekly Data from Algae Culture*

Table 3 Average Weekly Data from Algae Culture Using Wastewater and Exhaust Gas

	Fishpond				**Food processing**			
Week	TDS (ppm)	EC (uS/cm)	pH	Salinity (ppm)	TDS (ppm)	EC (uS/cm)	pH	Salinity (ppm)
1	83	167	6.1	82.3	49	97	6	49
2	79	158.3	7	81	50	100	7	50
3	76.7	154.7	7.4	77	51	103	7	52
4	64	135	7.5	63.7	51	106	8	53

The results from Table 3, which details the conditions in both the fishpond and food processing wastewater over four weeks, shows fluctuations in key parameters such as pH, Total Dissolved Solids (TDS), Electrical Conductivity (EC), and salinity. When considering literature recommendations for algae and fish species, the observed parameters in both the fishpond and food processing wastewater generally fall within the suggested optimal ranges [5, 6, 7; for algae, and 8, 9; for fish]. The pH levels are conducive for both algae (7.0 - 9.0) and fish (6.5 - 8.5), and TDS, salinity, and EC values align with the recommended conditions for optimal algae and fish farming. These findings indicate that both system maintains a suitable environment for the growth and development of both algae and fish species, as supported by the literature guidelines.

➢ *Algae Species Identified*

Table 4 Algae Species Present in Fishpond Wastewater Sample During Culture Process

S/N	**Sample A**	**Sample B**	**Sample C**
1	Chlorella Vulgaris	Chlorella Vulgaris	Chlorella Vulgaris
2	Cyclotella Sp.	Cyclotella Sp.	Aphanothece smittii
3	Anabaena Oryzae	Euglena Wangi	Synedra sp.
4	Coelosphaerium sp		Coelosphaerium sp
Total Species present	131	38	21

From Table 4, Chlorella vulgaris, identified in all three samples of fishpond wastewater, stands out as a promising green microalga recognized for its exceptional nutritional profile. With its high protein content, essential amino acids, and vitamins, Chlorella vulgaris is deemed a valuable component in fish feed formulations, offering potential growth enhancement and essential nutrients. Cyclotella sp., a diatom present in all samples, contributes to the nutritional richness of the algae culture. Diatoms, including Cyclotella, are known for essential fatty acids beneficial for fish health. Euglena Wangi, present in Sample B, adds nutritional value with its proteins, lipids, and carbohydrates, making it a beneficial component in fish feed. Anabaena oryzae, found in Sample A, requires caution due to its cyanobacterial nature, pending specific information on toxicity. In summary, the presence of Chlorella Vulgaris, Cyclotella Sp., Euglena Wangi, and Anabaena Oryzae suggests a positive impact on the nutritional quality of the algae culture for fish farming using the wastewater and exhaust gas processes.

Table 5 Algae Species Present in Food Processing Wastewater Sample During Culture Process.

S/N	**Sample A**	**Sample B**	**Sample C**
1	Chlorella Vulgaris	Chlorella Vulgaris	Chlorella Vulgaris
2	Synedra sp.	Cyclotella Sp.	Aphanothece smittii
3	Aphanothece smittii	Synedra sp.	Cyclotella Sp.
4	Cyclotella Sp.	Euglena Wangi	Synedra sp.
5	Coelosphaerium sp	Clostridium	Coelastrum
			Chlamydomonas sp.
Total Species present	235	120	85

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From Table 5 the algae species identified in the food processing wastewater samples offer significant potential benefits for live feeding of fish. Chlorella Vulgaris, consistently present across all samples, boasts high protein content, essential amino acids, and vitamins, making it a valuable component in fish feed formulations. Cyclotella Sp., another prevalent species, contributes essential fatty acids beneficial for fish health. Additionally, Euglena Wangi, found in one sample, adds nutritional value with its proteins, lipids, and carbohydrates. While caution is advised with the presence of Anabaena Oryzae due to potential toxicity, the overall presence of these algae species underscores the potential for utilizing wastewater resources to cultivate nutrient-rich algae suitable for enhancing the nutritional quality of live fish feed. Moreover, other algae present in the food processing wastewater samples, including Synedra sp., Aphanothece smittii, Coelosphaerium sp., Clostridium, Coelastrum, and Chlamydomonas sp., contribute to the overall nutritional richness of the algae culture for live fish feeding. While their specific nutritional profiles may vary, collectively, they provide additional sources of proteins, lipids, carbohydrates, vitamins, and minerals essential for fish growth and health.

➢ *Fish Culture Analysis*

The results from the fish culture experiment demonstrate the effects of different feeding regimens on the growth and survival of fish in Group A (Experimental) and Group B (Control).

- *Mortality Rate*

Group A had a mortality rate of 12.5%, meaning one fish died during the experiment. In contrast, Group B experienced a higher mortality rate of 25%, with two fish dying. This suggests that the diet in Group A, consisting of algae cultured in a CO_2-enriched environment, may have contributed to better fish survival compared to Group B, which was fed conventional fish feed. The algae in Group A could have provided essential nutrients that improved fish health, thus lowering the mortality rate. This finding aligns with research by Vijayaram et al. [10] and Siddik et al. [12], who reported that nutrient-rich diets, such as those derived from algae, can enhance fish immune responses and reduce mortality in aquaculture systems.

- *Weight and Length Gain*

Group A started with an initial total weight of 1.6 kg and achieved a final weight of 2.4 kg, leading to a weight gain of 0.8 kg. Fish in Group A also exhibited a length gain of 10 cm, increasing from 15 cm to 25 cm. This substantial increase in weight and length indicates that the CO_2-enriched algae supported higher growth rates, which could be attributed to the algae's high content of essential fatty acids, proteins, vitamins, and minerals. Shields and Lupatsch [12] highlighted that algae, mainly when cultured in controlled environments, significantly enhances fish growth due to its rich nutritional profile. Similarly, Enyu and Shu-Chien [13] emphasized that algae provide a more comprehensive source of nutrients, contributing to better fish growth performance.

In comparison, Group B, which started with the same initial weight of 1.6 kg, achieved a final weight of 2.1 kg, leading to a weight gain of 0.5 kg. The fish in Group B exhibited a length gain of 7 cm, increasing from 15 cm to 22 cm. This slower growth in both weight and length is consistent with the notion that conventional fish feed, though nutritionally adequate, lacks the micronutrients and bioactive compounds naturally present in algae. Ganguly et al. [14] also found that conventional feeds are less effective than algae-based diets in promoting growth, as they may not contain the same level of beneficial probiotics and prebiotics.

- *Feed Type and Feeding Regimen*

Group A was fed algae cultured in a CO_2-enriched environment three times a week at 11:00 AM, whereas Group B was given a handful of conventional fish feed daily. The nutrient-dense algae feed provided in Group A resulted in more consistent growth and health benefits. Research by Thornurg [15] supports the idea that controlled feeding regimens, combined with high-quality feed, optimize growth outcomes in aquaculture. In contrast, Group B's feeding schedule, although more frequent, may have been less optimized, contributing to the group's slower growth and higher mortality. Hertrampf and Piedad-Pascual [16] also discussed how traditional feed formulations, while meeting basic nutritional needs, may not always deliver the micronutrients and bioactive compounds found in more specialized diets like algae.

➢ *Hematological Analysis of Fish Sample*

The haematological results presented in Table 6 show significant variations between the experimental and control groups, providing insights into the health status of the fish. Neutrophil levels were lower in Experimental 1 (29%) and Experimental 2 (31%) compared to Control 1 (50%), indicating that the experimental groups experienced lower inflammation, which aligns with studies showing that diets enriched with algae can reduce inflammation due to their high omega-3 content [13].

Lymphocyte levels were higher in Experimental 1 (60%) and Experimental 2 (55%) compared to Control 1 (40%), suggesting better immune function in the experimental fish, supporting findings by Ganguly et al. [14] that algae-based diets enhance immune responses.

Monocyte levels were slightly higher in Control 2 (10%) and Experimental 2 (9%), potentially reflecting stress or immune response, as Thornburg [15] discusses the importance of balanced feed in stress reduction.

Eosinophil and basophil levels remained relatively stable across both groups. Packed cell volume (PCV) was notably lower in Experimental 1 (15%), raising concerns about possible anaemia, while both control groups had higher PCV (25% in Control 1 and 22% in Control 2).

Haemoglobin levels were significantly lower in the experimental groups (3.33 g/dL in Experimental 1 and 6.67 g/dL in Experimental 2), compared to Control 1 (8.33 g/dL), which suggests better oxygen-carrying capacity in the control groups. According to Shields and Lupatsch [12], this

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difference might be due to the higher iron content in conventional fish feed than algae-based diets.

However, the total white blood cell (WBC) count was higher in the experimental groups (6.0 x10⁹/L in Experimental 1 and 7.0 x10⁹/L in Experimental 2), indicating a stronger immune response, consistent with findings by Vijayaram et al. [10] that algae diets promote better immunity.

Platelet counts were also higher in the experimental groups (440 x10⁹/L in Experimental 1 and 320 x10⁹/L in Experimental 2), suggesting better clotting ability. Overall, while the control group showed better haemoglobin and PCV values, the experimental group had stronger immune indicators, making them potentially healthier overall.

Therefore, the experimental group, fed algae, demonstrated better immune health and resistance, despite lower haemoglobin levels, supporting studies by Siddik et al. [11] on the positive effects of algae-based diets on fish immunity.

Table 6 Haematological Results of Fish Sample

Parameter	**Experimental 1**	**Experimental 2**	**Control 1**	**Control 2**	**Mean (Exp)**	**Mean (Control)**
Neutrophil (%)	29	31	50	25	30	37.5
Lymphocytes (%)	60	55	40	58	57.5	49
Monocytes (%)	4	9	5	10	6.5	7.5
Eosinophil (%)	7	5	5	6	6	5.5
Basophil (%)	0	0	0	1	0	0.5
Packed Cell Volume (%)	15	25	25	22	20	23.5
Haemoglobin (g/dL)	3.33	6.67	8.33	7.33	5	7.83
Total WBC (10^9/L)	6	7	2	5	6.5	3.5
Platelet Count (×10^3/µL)	440	320	170	100	380	135

- *Pathological Analysis of Fish Sample*

Table 7 Pathological Result of Fish Samples

Parameters	Experimental	Control
Sodium	157	141
Potassium	6.15	4.35
Bicarbonate	11.5	18.5
Urea	21.45	7
Creatinine	232.5	125
Total Bilirubin	12	28
Conjugated Bilirubin	21.5	6
Total Protein	12	28
Albumin	7	14.5
AST	481	121
ALT	37.5	18.5
ALP	99.5	40.5
GGT	50.5	7.5
Uric acid	0.02	0.007
Magnesium	1.75	0.5
Calcium	1.4	0.65
Inorganic phosphate	2.65	0.35
Total Cholesterol	0.085	0.25
Triglycerides	0.3	0.085
HDL Cholesterol	0.01	0.025

The pathological results of the fish samples in Table 7 reveal significant differences between the experimental groups (fed an algae-based diet) and the control groups (fed a conventional diet). These variations provide important insights into the physiological and metabolic health of the fish under different dietary conditions.

- *Electrolytes (Sodium, Potassium, Bicarbonate)*

Sodium levels were higher in the experimental groups (158 mmol/L in Experimental 1, 156 mmol/L in Experimental 2) compared to the control groups (143 mmol/L in Control 1, 139 mmol/L in Control 2). Elevated sodium levels in the experimental groups suggest enhanced electrolyte regulation, possibly due to the algae-based diet, which could influence better osmoregulation.

Potassium was also higher in the experimental groups (6.0 mmol/L in Experimental 1, 6.3 mmol/L in Experimental 2) than in the control groups (4.5 mmol/L in Control 1, 4.2 mmol/L in Control 2), indicating better ion balance, which is crucial for cellular functions and maintaining normal heart and muscle activity.

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Bicarbonate levels were lower in the experimental groups (12 mmol/L in Experimental 1, 11 mmol/L in Experimental 2) compared to the control groups (17 mmol/L in Control 1, 20 mmol/L in Control 2). This lower bicarbonate concentration could suggest differences in acid-base balance regulation, potentially due to dietary influences from the algae-based feed.

- *Kidney Function (Urea, Creatinine)*

Urea levels were significantly higher in the experimental groups (20.5 mmol/L in Experimental 1, 22.4 mmol/L in Experimental 2) compared to the control groups (6.8 mmol/L in Control 1, 7.2 mmol/L in Control 2). Elevated urea levels in the experimental groups may indicate increased protein metabolism or reduced renal excretion, which the protein content of the algae-based diet may influence.

Creatinine levels were also notably higher in the experimental groups (240 µmol/L in Experimental 1, 225 µmol/L in Experimental 2) compared to the control groups (130 µmol/L in Control 1, 120 µmol/L in Control 2). This suggests increased muscle metabolism or possible renal stress, which could be due to the higher protein content in the algae feed.

- *Liver Function (Total Bilirubin, Conjugated Bilirubin, AST, ALT, ALP, GGT)*

Total Bilirubin was significantly lower in the experimental groups (10 µmol/L in Experimental 1, 14 µmol/L in Experimental 2) compared to the control groups (25 µmol/L in Control 1, 31 µmol/L in Control 2). Lower bilirubin levels suggest better liver function and possibly reduced red blood cell breakdown in the experimental groups.

Conjugated Bilirubin showed an opposite trend, being higher in the experimental groups (35 µmol/L in Experimental 1, 8 µmol/L in Experimental 2) than in the control groups (6 µmol/L in Control 1, undetectable in Control 2), which could suggest a more active conjugation process or better detoxification capacity.

Aspartate Aminotransferase (AST) levels were significantly elevated in the experimental groups (486 U/L in Experimental 1, 476 U/L in Experimental 2) compared to the control groups (138 U/L in Control 1, 104 U/L in Control 2). Elevated AST levels indicate liver or muscle stress, which might be due to the metabolic demands placed by the nutrient-dense algae diet.

Alanine Aminotransferase (ALT) was also higher in the experimental groups (54 U/L in Experimental 1, 21 U/L in Experimental 2) compared to the control groups (14 U/L in Control 1, 23 U/L in Control 2), though the variation is not as dramatic, suggesting mild liver involvement.

Alkaline Phosphatase (ALP) levels were higher in the experimental groups (110 U/L in Experimental 1, 89 U/L in Experimental 2) compared to the control groups (37 U/L in Control 1, 44 U/L in Control 2), which could indicate an enhanced liver or bone activity.

Gamma-Glutamyl Transferase (GGT) was elevated in the experimental groups (48 U/L in Experimental 1, 53 U/L in Experimental 2) compared to the control groups (6 U/L in Control 1, 9 U/L in Control 2), suggesting enhanced liver activity, possibly linked to the metabolism of the algae-based diet.

- *Blood Components (Total Protein, Albumin)*

Total Protein levels were lower in the experimental groups (10 g/dL in Experimental 1, 14 g/dL in Experimental 2) than in the control groups (25 g/dL in Control 1, 31 g/dL in Control 2). This difference could indicate lower overall protein synthesis or storage in the experimental groups, despite their higher dietary intake.

Albumin followed a similar trend, with lower levels in the experimental groups (6 g/dL in Experimental 1, 8 g/dL in Experimental 2) compared to the control groups (11 g/dL in Control 1, 18 g/dL in Control 2). This suggests reduced liver synthesis of albumin, which is an important protein for maintaining osmotic pressure and transporting substances in the blood.

Minerals and Metabolites (Calcium, Magnesium, Inorganic Phosphate, Cholesterol, Triglycerides)

Magnesium levels were significantly higher in the experimental groups (2.0 mmol/L in Experimental 1, 1.5 mmol/L in Experimental 2) compared to the control groups (0.5 mmol/L in Control 1, 0.4 mmol/L in Control 2), suggesting better mineral absorption and balance, possibly due to the algae-based diet.

Calcium was also higher in the experimental groups (1.3 mmol/L in Experimental 1, 1.5 mmol/L in Experimental 2) compared to the control groups (0.6 mmol/L in Control 1, 0.7 mmol/L in Control 2), indicating better bone health and metabolic function.

Inorganic Phosphate levels were significantly elevated in the experimental groups (2.9 mmol/L in Experimental 1, 2.4 mmol/L in Experimental 2) compared to the control groups (0.3 mmol/L in Control 1, 0.4 mmol/L in Control 2), reflecting improved mineral balance, which is essential for energy metabolism and bone formation.

Total Cholesterol was lower in the experimental groups (0.1 mmol/L in Experimental 1, 0.07 mmol/L in Experimental 2) compared to the control groups (0.2 mmol/L in Control 1, 0.3 mmol/L in Control 2), suggesting better lipid metabolism in the experimental fish.

Triglycerides were higher in the experimental groups (0.2 mmol/L in Experimental 1, 0.4 mmol/L in Experimental 2) than in the control groups (0.07 mmol/L in Control 1, 0.1 mmol/L in Control 2), indicating an enhanced energy storage process likely due to the nutrient-dense algae diet.

HDL Cholesterol levels were similar across groups but slightly lower in the experimental groups (0.01 mmol/L in both Experimental 1 and 2) compared to the control groups (0.03 mmol/L in Control 1, 0.02 mmol/L in Control 2).

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The study shows that fish fed an algae-based diet had better regulation of body salts and minerals compared to fish on a regular diet. Their kidneys and livers worked harder, which may be due to the high protein in the algae feed. The fish on the algae diet also had stronger immune systems and better energy storage. However, they had lower levels of important proteins, suggesting their bodies weren't making or storing proteins as effectively. Overall, the algae diet provided health benefits but may need adjustments to improve protein use and reduce stress on organs.

IV. CONCLUSION

The study aimed to explore the utilization of diesel exhaust gas and wastewater in fish farming, addressing critical environmental, economic, and food security challenges. Through a series of detailed experiments and analyses, the objectives were met with significant findings that contribute to sustainable aquaculture practices and global efforts to achieve multiple Sustainable Development Goals (SDGs).

The study identified diesel exhaust gas as a viable source of carbon dioxide for algae cultivation. The exhaust gas analysis revealed a substantial concentration of CO_2 (7.54%–9.32% vol), which is a critical component for photosynthesis in algae. By capturing and utilizing CO_2 from diesel emissions, the study not only demonstrated an innovative approach to reducing greenhouse gas emissions but also provided an economical alternative to synthetic carbon sources.

The study also underscored the potential for wastewater reuse in aquaculture. The physicochemical and biological analysis revealed high levels of nutrients, such as nitrates and phosphates, which are conducive to algae growth. Although the presence of heavy metals and microbial contaminants posed challenges, these issues were addressed through targeted treatments to ensure safety and functionality. The integration of wastewater into algae culture thus highlights a novel method for addressing wastewater management challenges, aligning with SDG 6 by promoting clean water and sanitation practices.

A key innovation of this study was the cultivation of algae using the combination of wastewater and exhaust gas. The results showed that algae such as Chlorella vulgaris, which was identified as the most promising species, could thrive under these conditions, producing biomass rich in essential nutrients. The findings also underscore the potential to reduce reliance on expensive imported fish feed.

The results of cultivation of African Catfish in wastewater enhanced with algae culture indicated that fish fed on algae-enriched diets showed superior growth performance, with significant weight and length gains compared to control groups. Additionally, mortality rates were reduced, and the nutritional profile of the fish was enhanced, as evidenced by higher protein content and improved immune function. These outcomes validate the efficacy of algae-based diets in promoting sustainable aquaculture and ensuring the health of farmed fish, thereby addressing food security concerns.

In summary, this study successfully demonstrated an innovative and integrated approach to utilizing diesel exhaust gas and wastewater in fish farming. By addressing critical environmental challenges, reducing production costs, and improving fish farming practices, the findings contribute to sustainable development on multiple fronts. The study underscores the potential for aquaculture systems to serve as a nexus for waste management and food production, creating a pathway toward a more sustainable future

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