Quest Journals
Journal of Research in Environmental and Earth Sciences
Volume 9 ~ Issue 10 (2023) pp: 01-11
ISSN(Online) :2348-2532
www.questjournals.org

Research Paper

Remediation Potentials of Sweet Orange and Cassava Peels on Physicochemical Properties and Microbial Counts of Crude Oil- Contaminated Soils

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Abstract
The study assessed the possible remediation of various agricultural wastes, including sweet orange and cassava peels, on the physicochemical parameters and microbiological counts of crude oil polluted soils. The soils from Rumuekpe in Emuoha Local Government Area, Rivers State, Nigeria were tested for the concentrations of the indicators before and after pollution. The polluted specimens were subsequently treated with organic substrates for 6 weeks, with parameter values made at 0week, 2weeks, 4weeks, and 6weeks. Descriptive statistics were employed for the data analysis. The study's findings suggested that sweet orange and cassava peels have a high potential for bioremediation of crude oil damaged soils. After 4 weeks of treatment with orange peels and 2 weeks of treatment with cassava peels, soil pH was returned to pre-contamination values. Total organic carbon (TOC) levels in treated soil samples were considerably lower ($p<0.05$) than in controls. Total petroleum hydrocarbon (THP) decreased gradually throughout the course of the study, however total hydrocarbon consuming bacteria rose gradually in the nutrient-treated plots as compared to the controls. The study indicated that the substrate had reasonable potential for reducing total organic carbon, total petroleum hydrocarbon, and poly aromatic hydrocarbons, with all three parameters moving to levels lower than the control level. These agricultural wastes also demonstrated enormous potential for raising the population of Total Hydrocarbon Utilizing Bacteria (THUB), which has an influence on the breakdown of petroleum hydrocarbons. Both substrates appear to be more effective after 4 weeks of treatment; nonetheless, they had little effect on heavy metal concentrations in crude oil polluted soils. The pH of polluted soils became more alkaline and hence more appropriate for agricultural reasons after cassava and orange peel treatments. As a result, it was advised that cassava and orange peel not be tossed as waste rubbish but can be useful in the treatment of soils polluted by crude oil.

Keywords: *Sweet orange, Cassava, Hydrocarbon, Soil properties, Pollution, Bacteria*

R*eceived 01 Oct, 2023;* ***R****evised 09 Oct., 2023;* ***A****ccepted 11 Oct., 2023 © The author(s) 2023.*
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I. Introduction

Raw petroleum and other petroleum-related materials pose a threat to the survival of marine and terrestrial life forms, including humans who live in such polluted environments. This is due to the unrefined petroleum products' predicted poisonousness, mutagenicity, and cancer-causing properties when present in large concentrations (Adebusoye et al. 2007). Man has been searching for eco-friendly solutions to cope with recovering dirty situations, notably unrefined petroleum polluted soil, due to the potential concerns associated with unrefined petroleum pollution (Chikere and Azubuike, 2013). Unprocessed petroleum is known to possess an amazing mixture of naturally occurring hydrocarbons (aliphatic, alicyclic, and sweet-smelling), which may be processed into diesel, gas, heating oil, fly fuel, lamp oil, and several other products (petrochemicals) of commercial importance (Iyagba and Offor, 2014). These hazards can alter population components, disrupt trophic cooperation, and disrupt the structure of regular networks within a living system (Bejarano and Michel, 2010).

The epidemic of natural pollution in Nigeria has recently reached a scary magnitude, especially in the Niger-Delta region, the largest delta in Africa and the third largest in actuality where the majority of the country's raw petroleum is discovered (Essien et al, 2015). Since commercial research into gasoline began in Nigeria in 1958 (Essien et al. 2015), gasoline has steadily grown to form the foundation of the country's economy. Whatever the case, the search for oil has led to the degradation of land and waterways. The fishing rivers and streams have essentially dried up, and the agricultural land has become less productive (Dabbs, 1996; Okpokwasili and Odokuma, 1990). In the Nigerian Niger Delta region, there have also been a few instances of widespread unrest brought on by natural devastation from oil exploration (Inoni et al. 2006). Given the widespread modern use of oil-based goods, petroleum hydrocarbons are the most well recognised natural toxins in the environment.

Long-term efforts to clean up polluted areas have increased in oil-rich and economically advantageous countries (Song, et al., 2017). Many approaches, including physical, substance-based, and natural ones, have been adopted. However, some of these are expensive and others have harmed the environment, particularly soil health and human jobs (Morales-Bautista et al. 2016; Hussain et al. 2018). For instance, burning (synthetic technique) produces ozone depleting compounds to the environment, causing a serious atmospheric deviation, whereas excavating (real methodology) has methods and transport limits (Arthur and Casey, 2014)1. Utilizing natural compost is a more environmentally friendly solution since it releases nutrients more gradually and acts as a soil conditioner (Khudur, et al. 2018; Hussain et al. 2018).

Compared to its counterparts (physical and chemical remediation approaches), bioremediation treatments are safe, cost-effective, and eco-friendly (Dados et al. 2015). Additionally, bioremediation preserves the surface and properties of the soil (Adams and Guzman-Osorio, 2008). Despite the many advantages of bioremediation, its use depends on a number of complex factors, which may be divided into two main groups: the toxin's nature (level of contamination, total, and oxidation condition of raw petroleum), and the environment (Agarry and Jimoda, 2015). It is crucial that appropriate supplement concentrations, especially nitrogen and phosphorus, are maintained in the ideal ratio in any polluted climate to balance the unevenness caused by the high carbon content of raw petroleum during contamination, which could impede the growth and activities of hydrocarbon clastic microbes (Bamforth and Singleton, 2015; Ayotamuno et al. 2006)

Composts that have been developed have been used as biostimulants for enhanced bioremediation of oil hydrocarbon-contaminated areas for a long time. However, because to its excessive use, there have been negative side effects such eutrophication, blue child syndrome, and barometric pollution (Geddes et al. 2015). In addition, due of its widespread usage as essential agricultural knowledge, manufactured composts are exorbitantly costly in non-industrial countries like Nigeria (Danjuma et al. 2012). In order to improve bioremediation, these challenges and the need for ecological manageability have led analysts to search for natural substrates as alternatives to produced composts. Currently, nitrogen-rich natural substrates are used as biostimulants, and they have shown to be effective in accelerating bioremediation. Corn build-ups, sugarcane bagasse, banana skins, sweet potato strips, sawdust, wasted fermenting grains, rice husks, coconut shells, cow dung, pig fertiliser, and bird faeces are some of the nitrogen-containing natural substrates that have been used as biostimulants (Agarry et al. 2010; Agarry and Jimoda, 2013)

Natural compost maintains an adequate inventory of soil natural matter with high microbial burdens and improves soil physical and material characteristics (Khudur et al. 2018). This facilitates the faster degradation of hydrocarbon contaminants (Vidonish, et al. 2016). In their study of rye grass, Kaimi et al. (2006) provided evidence that adding fertiliser excrement to the soil accelerates the removal of petroleum hydrocarbons (PHCs), and Obasi et al. (2013) provided specifics on the removal of 60–65 percent of the hydrocarbon from soils treated with excrement and urban biowaste fertiliser. This study, which was distinct from those mentioned above and focused on the biostimulation of phytoremediators, used orange strips as a cost-effective, widely available resource (Okunwaye*et al.*, 2019).

Palm kernel husk is one major product that is generated from the processing of fresh palm fruits from which, palm oil is extracted. Oil palm processing mills are built and used by indigenous communities, most especially in Rivers, Imo, Akwa-Ibom, and Delta State, etc., to process there palm fruits. It had been observed by researchers that over twenty-two percent of the harvested palm fruits processed, ends up as palm kernel husk (Lim and Zaharah, 2000)

Nigeria is blessed with domestic birds and livestock’s such as fowl, ram, sheep, cow, goat, and etc. These livestock’s produces waste dungs and are abound in the cattle markets (i.e., slaughter houses), which are avenue for such dung’s, and are considered waste. These wastes are considered useless to the ordinary man. In any case, research has shown that such squanders are valuable material to change the dirt physical and synthetic properties and furthermore, to deliver supplements for a more extended timeframe. A few of previous works made use of organic wastes and this is being demonstrated in the literature. Therefore, the present study is assessing the potentials of some organic wastes on physicochemical properties and of crude oil- contaminated soils.

II. Materials and Methods

The study was carried out in Rumuekpe Community, Emohua LGA, Rivers State, Nigeria. The geographical coordinate of the study area is located geographically within 4°58′25.9″N and 6°31′45.2″E (Figure 1).

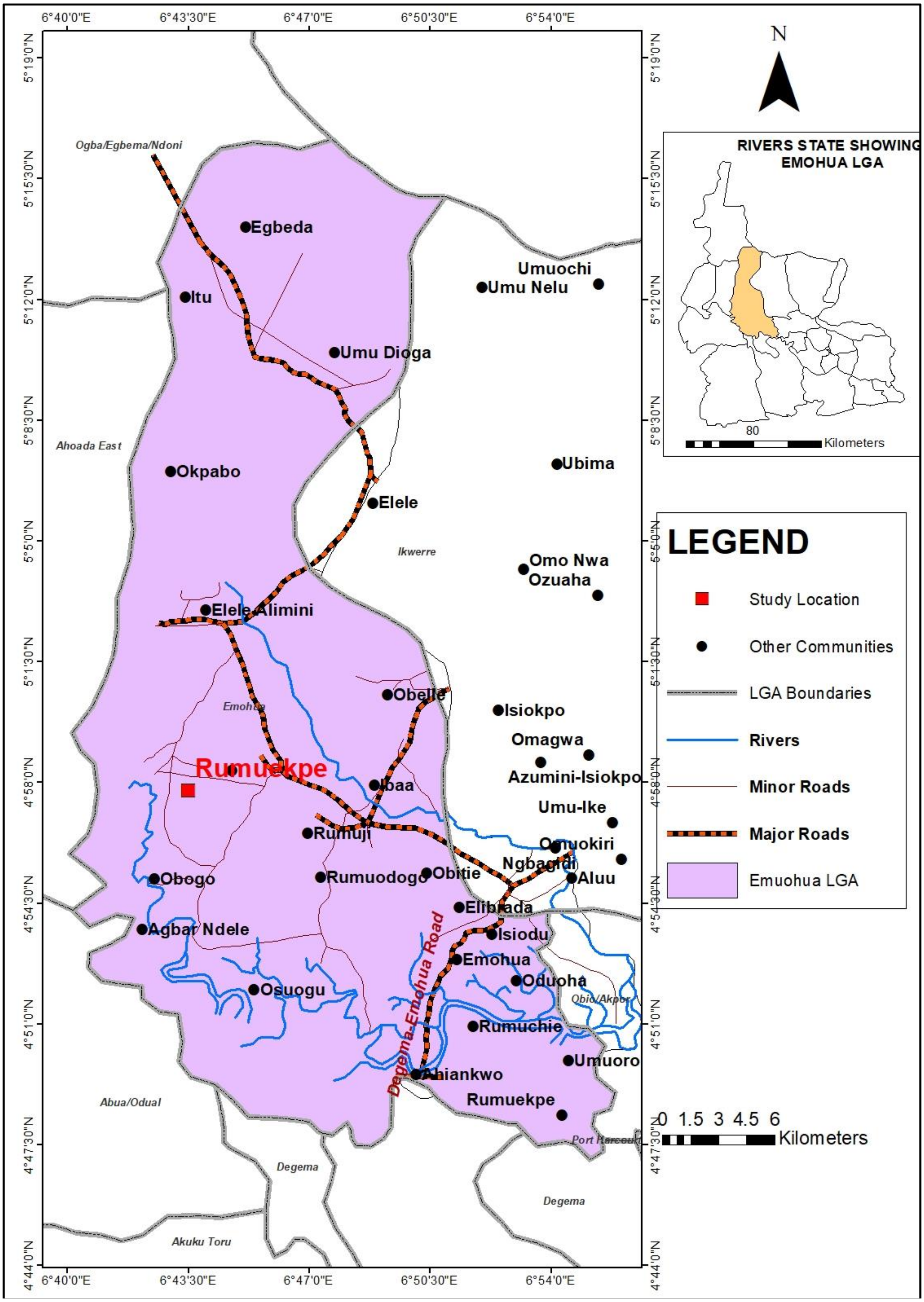


Figure 1. Emohua LGA showing the Study Location

Preparation of Peel Extracts

The fresh peels of cassava tuber and orange fruit were collected from farmers at Ubima town and orange sellers at Rumuola market in Ikwerre and ObioAkpor local government areas of River State respectively.The peels were washed thoroughly with deionized water to remove sand and other impurities, sun-dried for 2 weeks and ground to powder using an electric blender. The powdered peels of each of the plants were extracted with the use of Soxhlet extractor (Plate 1). 5kg of each powdered sample was extracted with 1 liter of 99.8% methanol in the Soxhlet extractor. Using an oven at $40^{o}C$ the resulting extracts were evaporated to near dryness to obtain a paste residue that is free of methanol. The extracts were then weighed and stored in sample bottles and stored in refrigerator.

Sample Collection

The crude impacted and unimpacted soils for the study were taken from top soil (0 - 30cm depth) from Rumuekpe in Rivers State's Emuohua local government area. To cover the depth of penetration of the pollutant, soil from 0 to 30cm deep was collected. The soil samples were gathered and arranged in wooden boxes 40cm by 40cm in size. The boards allowed for management of the soil's depth and exposed surface area, as well as temperature, nutrient content, and oxygen availability (Mattewson& Grubbs, 1988).Cassava peels were gathered from farmers in Ubima town, Ikwerre LGA, and peelings from fruit vendors in Rumuola, ObioAkpor LGA in Rivers State.

Experimental Design

Exactly 2kg of soil was introduced into each of eleven experimental boxes, built with wooden board, with the following treatment options:

Experimental Box V (1 box): Non contaminated soil to serve as Control 1

Experimental Box W (1 box): Contaminated soil without treatment to serve as Control 2

Experimental Box X (3 boxes): Contaminated soil with cassava peels treatment (CPT)- Three boxes with different measurements of treatments- X_1- 100, X_2- 200, X_3- 300.

Experimental Box Y (3 boxes): Contaminated soil with orange peels treatment (OPT)- Three boxes with different measurement of treatments- Y_1- 100, Y_2- 200, Y_3- 300.

Experimental Box Z (3 boxes): Contaminated soil with mixed peels treatment (MPT)- Three boxes with different measurement of treatments- Z_1- 100, Z_2- 200, Z_3- 300.

These experimental boxes were moistened and properly homogenized before sampling the boxes at an interval of 14days for a total duration of 42days.

Determination of Physico-chemical Parameters in Samples

Standard methods were employed for determining physical and chemical parameters in the peeled samples of cassava and orange in both polluted and unpolluted soils. These included pH, moisture content, electrical conductivity, total organic carbon, total nitrogen, available phosphorus, and total petroleum hydrocarbon, PAH and THUB (Adesodun and Mbagwu, 2008). Descriptive statistics which involved the use of mean values and standard deviation to describe the results. The results were presented in tables and graphs.

III. Results and Discussions

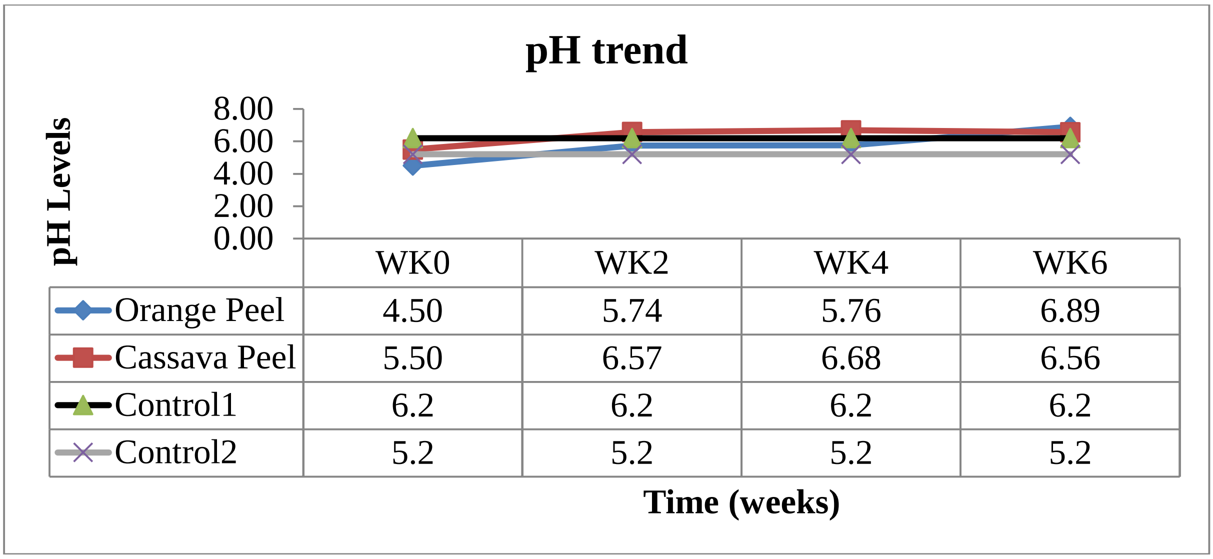
The study's findings are displayed in Table 1 and Figures 1 through 16, which indicate the concentrations of physicochemical elements in uncontaminated, crude oil-contaminated and organic waste-treated soils. The soil had a pH range of 4.50 to 6.89 and a temperature range of 24.90 to 27.37°C. Moisture content ranged from 17.7 to 22.27%, total organic carbon ranged from 0.77 to 2.30 ppm, and total potassium ranged from 0.01 to 20.23 ppm. Electrical conductivity ranged from 0 to 1970 S/cm; Total Nitrogen ranged from 0.02 – 36.40 ppm; PAH ranged from 0.00 – 2.77 mg/kg; TPH ranged from 0.00 – 5752 mg/kg; THB ranged from 0.00 - 6.94 x 10^6 cfu/g; THUB ranged from 0.00 – 72333 cfu/g.

Table 1. Levels of Soil Parameters in Uncontaminated, Contaminated and Treated Soils

Soil	US	CS	Treatment with OP				Treatment with CP			
Parameter	(Control1)	(Control2)	0wks	2wks	4wks	6wks	0wks	2wks	4wks	6wks
pH	6.2^{a}	5.2^{a}	4.50^{a}	5.74^{ab}	5.76^{ab}	6.89^{b}	5.50^{ab}	6.57^{ab}	6.68^{b}	6.56^{ab}
Temp (^{0}C)	25.6^{a}	24.9^{a}	25.30^{a}	25.10^{a}	26.07^{a}	27.37^{a}	25.30^{a}	25.13^{a}	26.13^{a}	27.63^{a}
EC(µS/cm)	60^{a}	128^{a}	1970^{b}	77.33^{a}	74.00^{a}	56.67^{a}	0.00^{c}	61.33^{a}	59.67^{a}	59.00^{a}
Moisture	18.7^{a}	17.7^{a}	18.50^{a}	19.77^{a}	20.20^{a}	20.34^{a}	18.30^{a}	21.87^{a}	21.30^{a}	22.27^{a}
TOC	0.78^{a}	2.01^{b}	2.30^{b}	1.50^{b}	1.04^{ab}	1.93^{b}	2.00^{b}	1.04^{ab}	0.77^{a}	1.87^{b}
Total K	0.045^{a}	0.01^{a}	20.23^{b}	0.05^{a}	0.08^{a}	0.03^{a}	12.43^{b}	0.07^{a}	0.09^{a}	0.04^{a}
Total N	0.06^{a}	0.02^{a}	36.40^{b}	0.06^{a}	0.07^{a}	0.06^{a}	2.15	0.08^{a}	0.08^{a}	0.08^{a}
PAH	0^{a}	2.77^{b}	0.00^{a}	0.86^{ab}	0.00^{a}	0.00^{a}	0.00^{a}	0.49^{a}	0.00^{a}	0.00^{a}
TPH	15.73^{a}	5752^{b}	0.00^{c}	2742.3^{b}	2347.7^{b}	3978.0^{b}	0.00^{c}	2357.0^{b}	2173.3^{b}	3459.67^{b}
THB	$3.30 \times 10^{5\ a}$	$6.32 \times 10^{6\ b}$	0.00^{c}	2.97 x	5.91 x	2.07 x	0.00^{c}	6.73 x	6.94 x	2.03 x

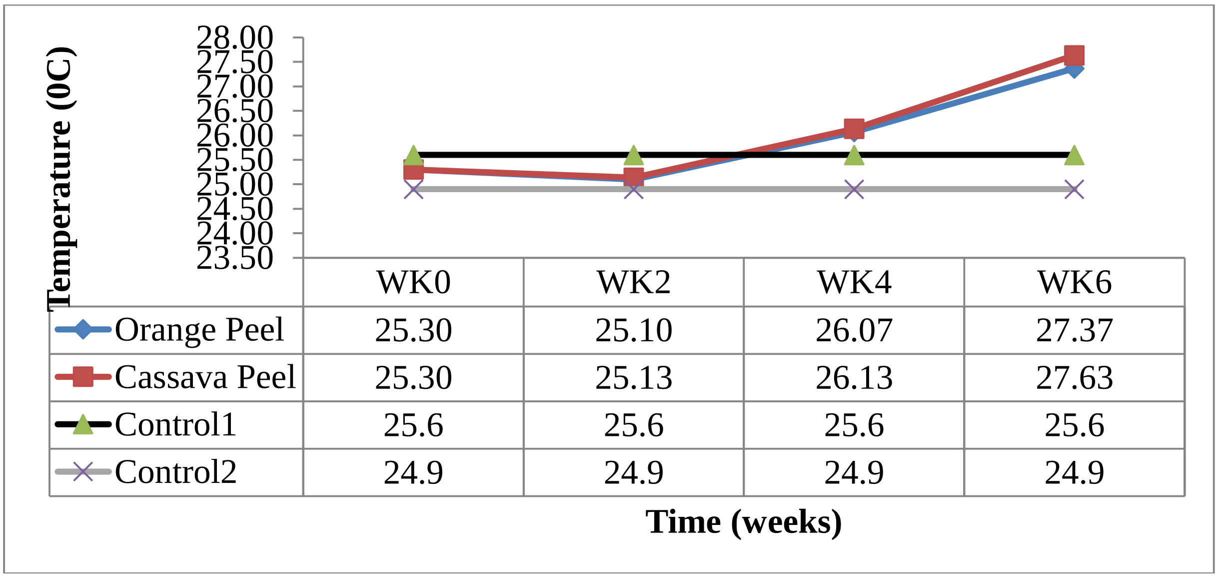
THUB	3000 ^{a}	9000 ^{b}	0.00 ^{d}	$10^{6\,b}$ 39200^{bc}	$10^{6\,b}$ 60000^{c}	$10^{6\,b}$ 16700 ^{b}	0.00^{d}	$10^{6\ b}$ 59900^{c}	$10^{6\,c}$ 72333 ^{c}	$10^{6\ b}$ 62033 ^{c}

Key: Values on the same row that do not share a letter are significantly different at 0.05 significant level



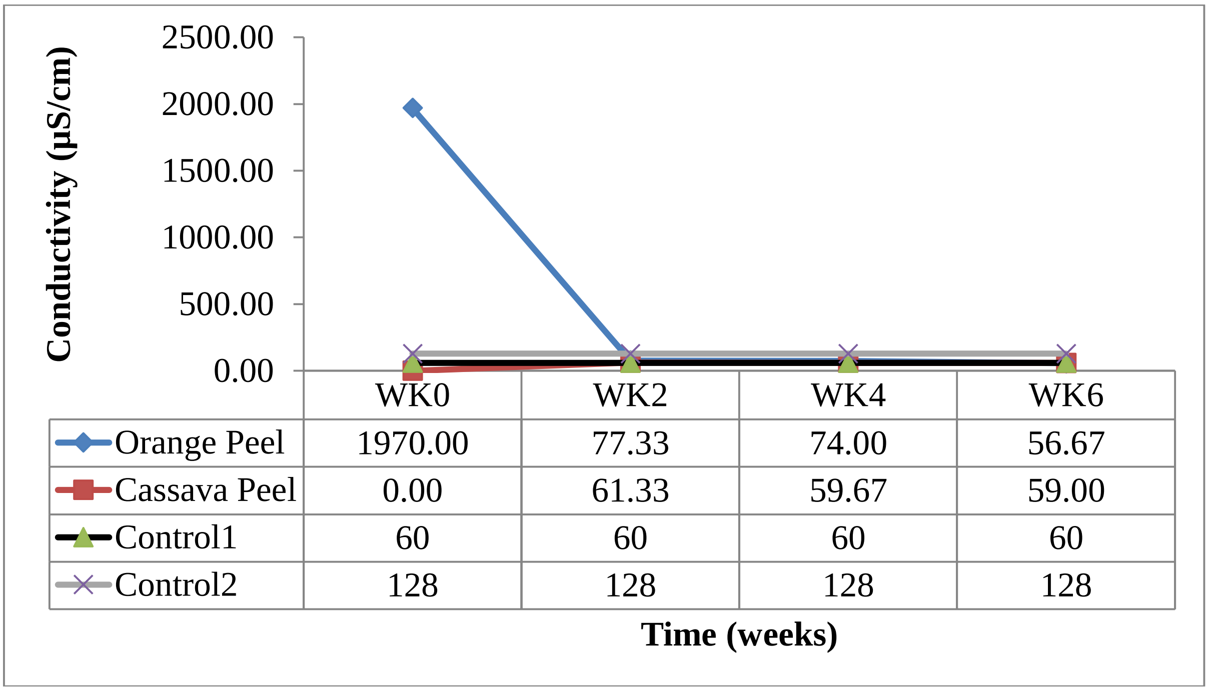
	WK0	WK2	WK4	WK6
Orange Peel	4.50	5.74	5.76	6.89
Cassava Peel	5.50	6.57	6.68	6.56
Control1	6.2	6.2	6.2	6.2
Control2	5.2	5.2	5.2	5.2

Fig 1: Substrate Treatment and Trends in Soil pH level overtime



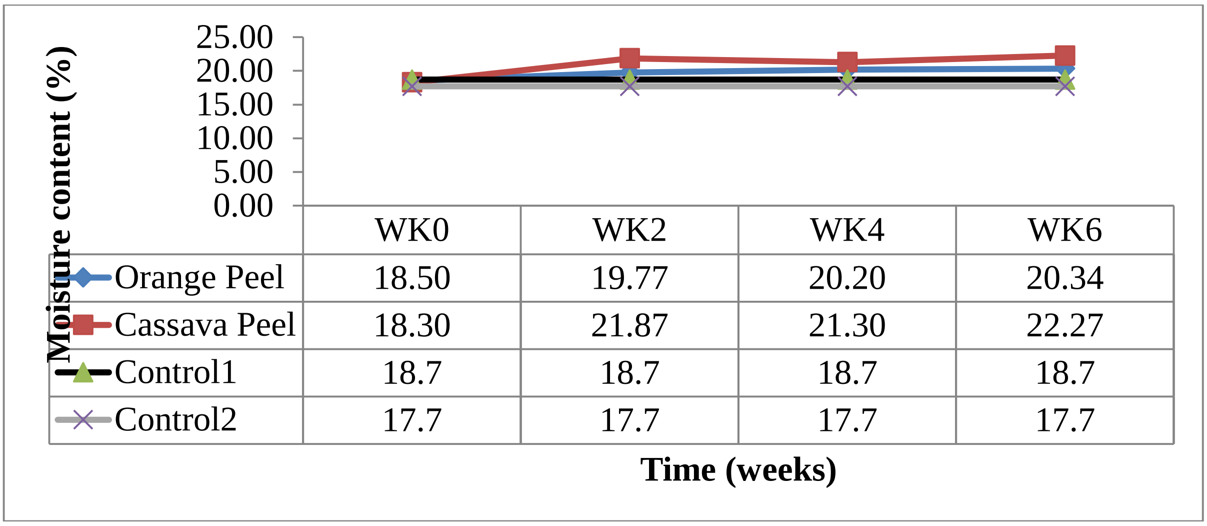
	WK0	WK2	WK4	WK6
Orange Peel	25.30	25.10	26.07	27.37
Cassava Peel	25.30	25.13	26.13	27.63
Control1	25.6	25.6	25.6	25.6
Control2	24.9	24.9	24.9	24.9

Fig 2: Substrate Treatment and Trends in Soil Temperature Level over Time



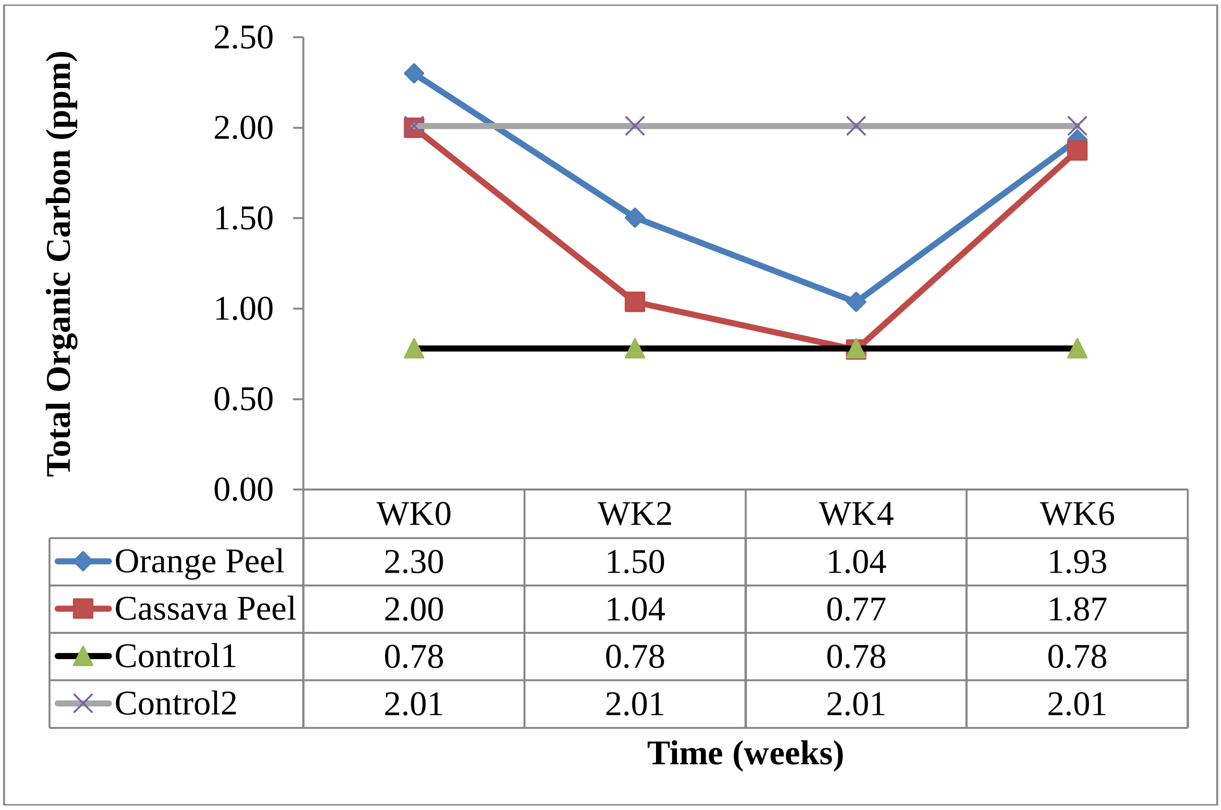
	WK0	WK2	WK4	WK6
Orange Peel	1970.00	77.33	74.00	56.67
Cassava Peel	0.00	61.33	59.67	59.00
Control1	60	60	60	60
Control2	128	128	128	128

Fig 3: Substrate Treatment and Trends in Soil Electrical Conductivity over time



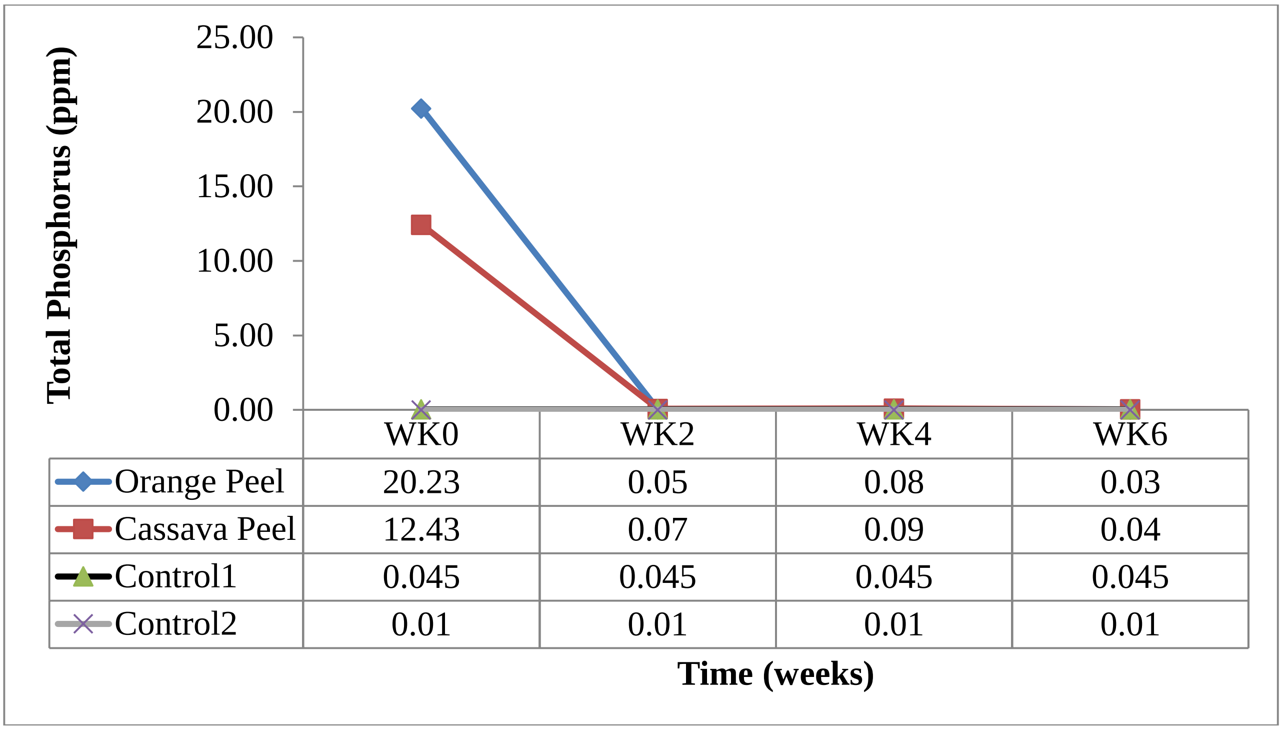
	WK0	WK2	WK4	WK6
Orange Peel	18.50	19.77	20.20	20.34
Cassava Peel	18.30	21.87	21.30	22.27
Control1	18.7	18.7	18.7	18.7
Control2	17.7	17.7	17.7	17.7

Fig 4.4: Substrate Treatment and Trends in Soil Moisture Content over time



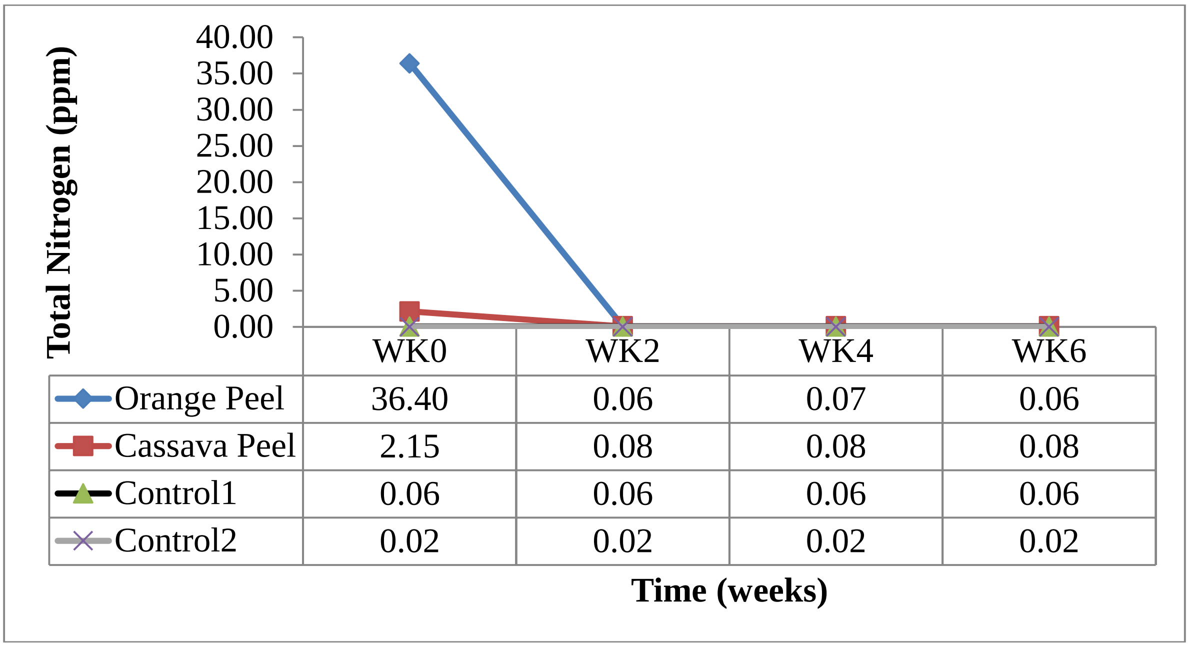
	WK0	WK2	WK4	WK6
Orange Peel	2.30	1.50	1.04	1.93
Cassava Peel	2.00	1.04	0.77	1.87
Control1	0.78	0.78	0.78	0.78
Control2	2.01	2.01	2.01	2.01

Fig 5: Substrate Treatment and Trends in Soil Total Organic Carbon Content over time



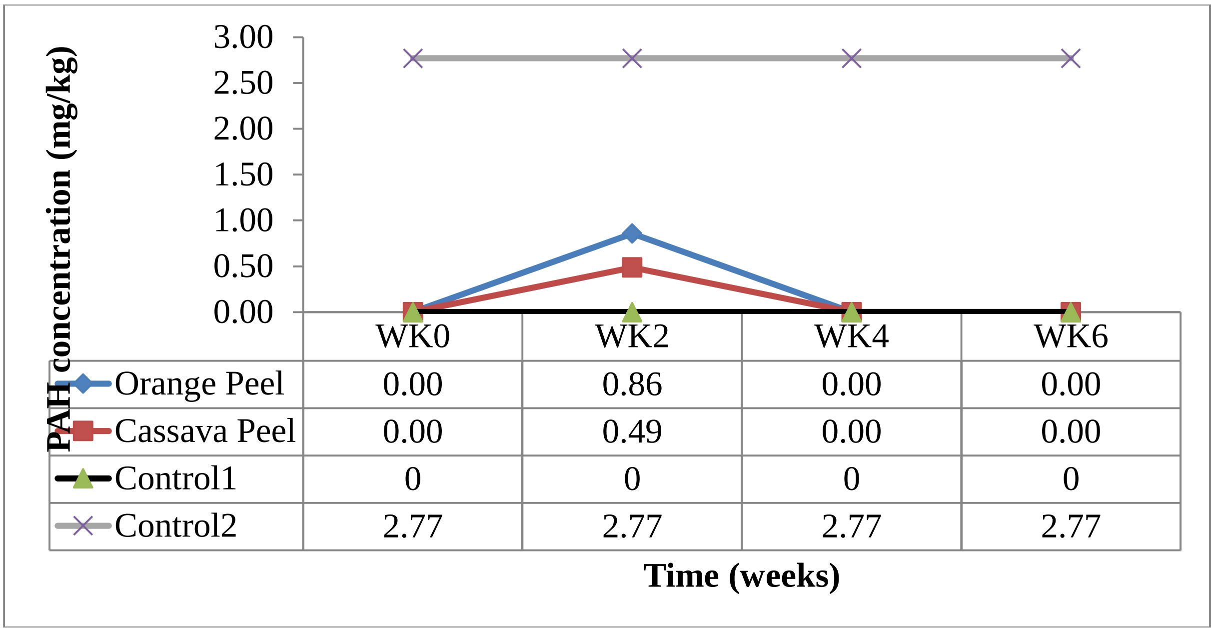
	WK0	WK2	WK4	WK6
Orange Peel	20.23	0.05	0.08	0.03
Cassava Peel	12.43	0.07	0.09	0.04
Control1	0.045	0.045	0.045	0.045
Control2	0.01	0.01	0.01	0.01

Fig 6: Substrate Treatment and Trends in the Levels of Total Phosphorus over time



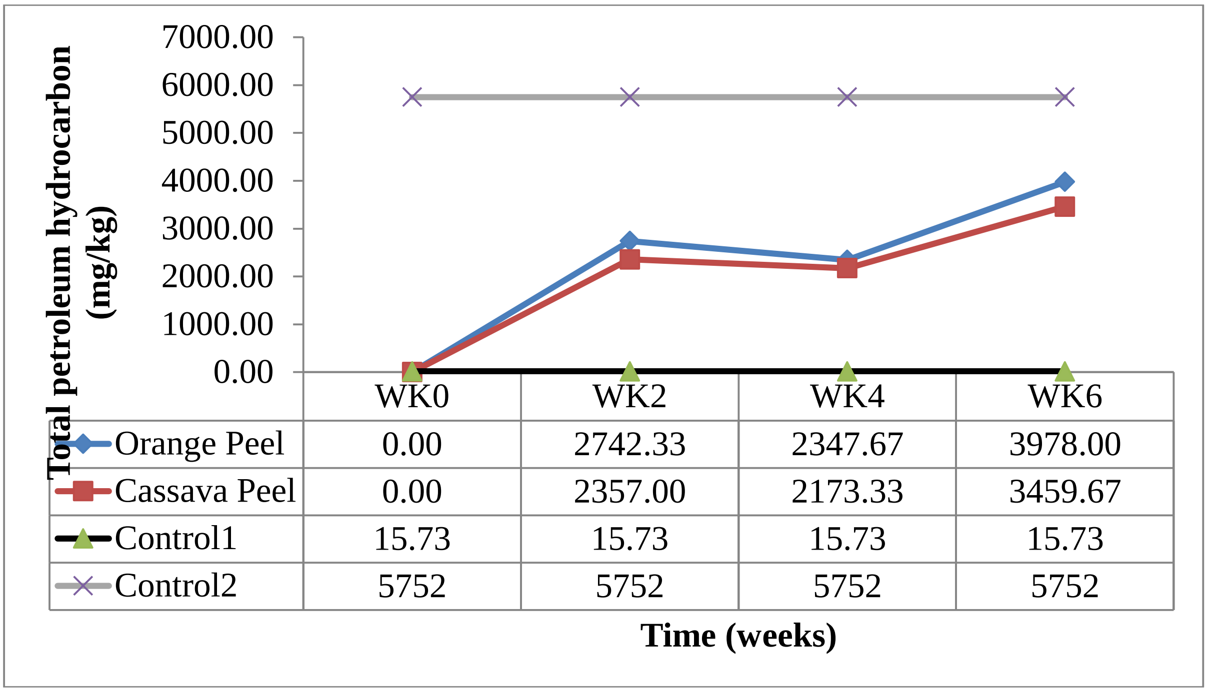
	WK0	WK2	WK4	WK6
Orange Peel	36.40	0.06	0.07	0.06
Cassava Peel	2.15	0.08	0.08	0.08
Control1	0.06	0.06	0.06	0.06
Control2	0.02	0.02	0.02	0.02

Fig 7: Substrate Treatment and Trends in the Levels of Total Nitrogen over time



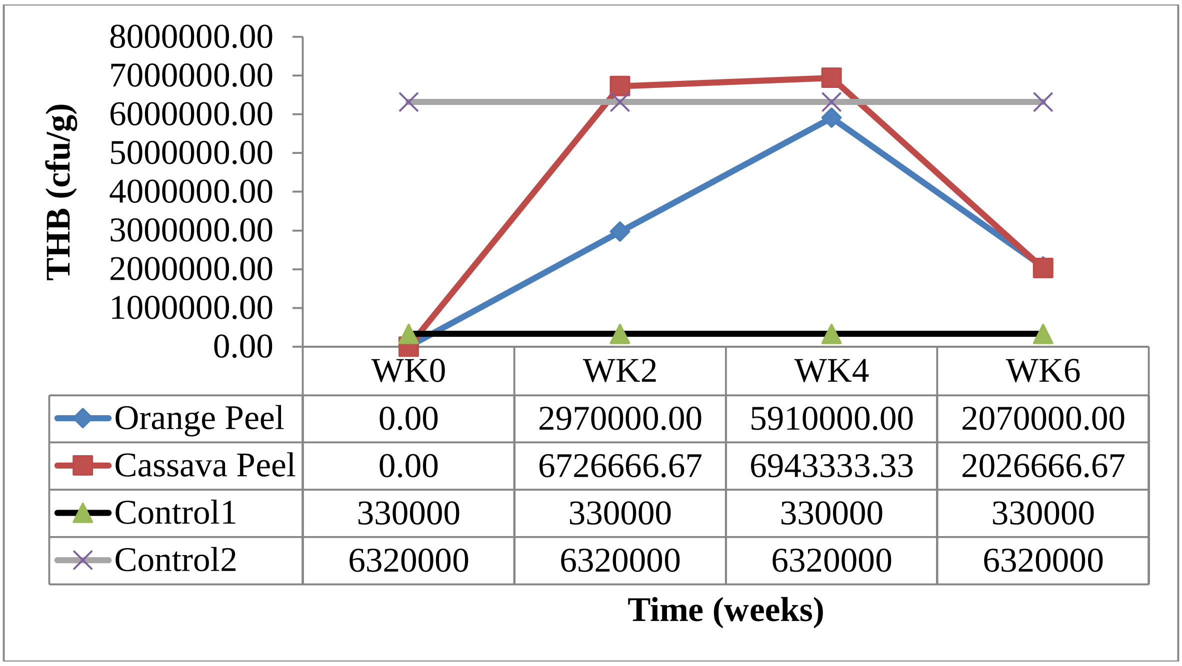
	WK0	WK2	WK4	WK6
Orange Peel	0.00	0.86	0.00	0.00
Cassava Peel	0.00	0.49	0.00	0.00
Control1	0	0	0	0
Control2	2.77	2.77	2.77	2.77

Fig 8: Substrate Treatment and Trends in PAH Concentration over time



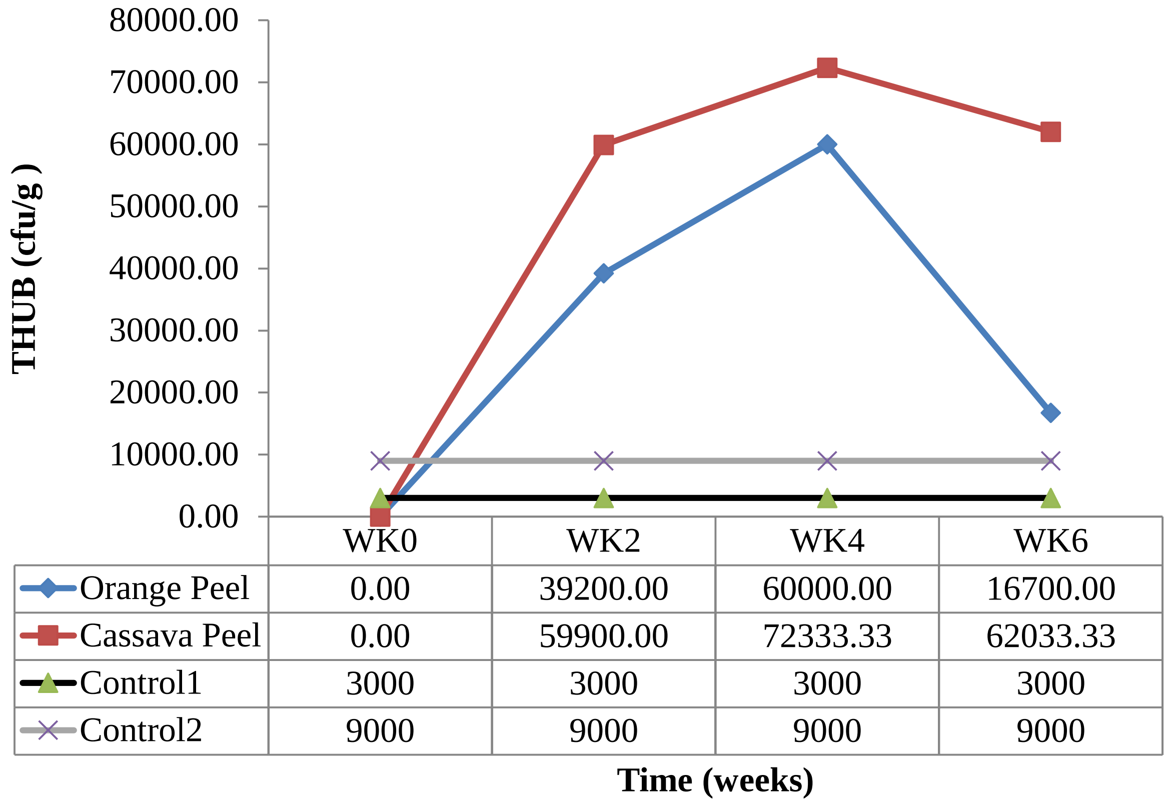
	WK0	WK2	WK4	WK6
Orange Peel	0.00	2742.33	2347.67	3978.00
Cassava Peel	0.00	2357.00	2173.33	3459.67
Control1	15.73	15.73	15.73	15.73
Control2	5752	5752	5752	5752

Fig 9: Substrate Treatment and Trends in Total Petroleum Hydrocarbon Levels with time



	WK0	WK2	WK4	WK6
Orange Peel	0.00	2970000.00	5910000.00	2070000.00
Cassava Peel	0.00	6726666.67	6943333.33	2026666.67
Control1	330000	330000	330000	330000
Control2	6320000	6320000	6320000	6320000

Fig 10: Substrate Treatment and Trends in THB Levels with time



	WK0	WK2	WK4	WK6
Orange Peel	0.00	39200.00	60000.00	16700.00
Cassava Peel	0.00	59900.00	72333.33	62033.33
Control1	3000	3000	3000	3000
Control2	9000	9000	9000	9000

Fig 11: Substrate Treatment and Trends in THUB Levels with time

IV. Discussion of Findings

The study's findings showed that after being contaminated with crude oil, the soil became more acidic. The soils of the Nigerian region of the Niger Delta, where Egbema is located, are unique in that they have minimal soil acidity (Ogbonna, et al. 2019). After being treated with organic waste, the pH of the soil varied between being below and over the control range. The soil's pH level decreased to 4.50 after being treated with orange peel, making with higher acidic levels seen in the polluted soils (Fig 1). However, increased pH level was observed over time after treatment with orange peel. The soil pH reached pre-contamination levels after 4 weeks and attained a maximum level of 6.89 after 6 weeks of treatment. With cassava peel, the soil pH level rose steadily from the levels observed after crude oil contamination and reached pre-contamination levels within two weeks of treatment. In table 1 it is observed that significantly different ($p<0.05$) peak levels of soil pH was observed at week 6 after orange peel treatment and at week 4 after treatment with cassava peel.Following crude oil contamination, the soil's electrical conductivity was marginally greater, but after bioremediation using cassava peel, it returned to its pre-contaminated values. When orange peels were used as the substrate, the EC increased to a peak of 1970 S/cm at week 0 before falling to pre-contamination values at weeks 2 to 6. The reduction in electrical conductivity of the soil after remediation treatments suggested that the site was undergoing remediation or soil reclamation.

Throughout the cleanup phase, the total organic carbon decreased. When the pollutant was first introduced into the soil, the organic carbon content increased significantly, but as biodegradation developed using cassava peel, the parameter decreased to pre-contamination values (Fig 5). The total Org C content of the soil was considerably ($p<0.05$) lower after four weeks of treatment with this substrate than in the contaminated soil, but it was not statistically ($p>0.05$) different from the levels in uncontaminated soils. Utilizing orange peel had a minimal impact on total organic carbon, but it did have bioremediation qualities because the parameter decreased after four weeks of treatment.Similar to total phosphorous, total nitrogen levels decreased significantly over time after initially increasing with the introduction of the substrates. Bacteria that break down petroleum quickly use up soil nitrogen in highly polluted soils. According to Fitzpatrick (2016), who has a similar perspective, in addition to biological absorption, nitrogen can be quickly lost from soils by ammonia leaching and denitrification in moist soils. It is possible that the study area's high rainfall totals and periodic water applications caused the soil to get moist and quickly lose nitrogen from the soil.

V. Conclusion and Recommendations

It can be concluded that the substrate showed a reasonable capacity for the reduction of total organic carbon, total petroleum hydrocarbon, and poly aromatic hydrocarbons after 6 weeks of treatment, trending all three measures to values below the control. The pH of polluted soils improved with both the cassava and orange peel treatments, becoming more alkaline and overall more conducive to agricultural use.These agricultural wastes also demonstrated enormous potential for boosting the population of Total Hydrocarbon Utilizing Bacteria (THUB), which affects how petroleum hydrocarbons degrade. The study recommended that agricultural waste such as cassava and orange peel should not be discarded as mere waste trash but should be employed for use as treatments in crude oil contaminated soils.

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