

Physicochemical Quality and Mycodegradation of Commercial Paint Effluents from Factories in Ado-Ekiti, Southwestern Region, Nigeria

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Abstract

Bioremediation is a useful method for restoring contaminated soils because of its cost effectiveness and environmental friendliness. However, the process is slow in soils with low pH. This study sought to explore the isolation and bioremediation of oil-based paint in aqueous medium using indigenous fungal isolates from paint contaminated soil from Ado Ekiti. The fungal isolates were initially screened for bioremediation potential in mineral salts medium containing oil-based paint (conc. 300 ppm, w/v) under shake flask conditions. Results of the physicochemical analysis of the soil showed: pH, 5.60-6.25; organic carbon, 2.28-4.70%; temperature, 27 -35 °C; BOD, 182-219 mg/L; COD, 292-719 mg/L; sulphate, 30-42 mg/L; nitrate, 2.25-8.0 mg/L. The load of heterotrophic fungi ranged from 1.21×10^4 to 17.0×10^4 cfu/g while paint-utilizing fungal counts in the samples ranged from 0.27×10^3 to 3.5×10^3 cfu/g. The isolated fungi mainly belonged to six (6) fungal genera, namely; *Penicillium* spp (43.75%), *Fusarium* spp (18.75%), *Rhizopus oryzae* (12.5%), *Aspergillus niger* (12.5%). Among them, *Penicillium notatum* showed the greatest paint degradation ability from day 7 to day 21 while *Penicillium citrium* showed the least among the fungal isolates on day 7. The results suggest that *Penicillium* isolates in this study could be exploited in bioremediation of paint effluents and contaminated soil.

Keywords: Biodegradation, Fungi, Contamination, Isolate, Paint effluent

INTRODUCTION

The rising population of humans has in no doubt resulted in increase in development of industries to accommodate their various needs. The major consequence of this is the problem of waste management to prevent environmental pollution and their attendant effects. One of the main sources of pollution worldwide is the paint industry. Paint industries pollute the environment through effluent discharge, gas emission and waste disposal in the form of organic and inorganic chemicals (Aniyikaiye *et al.*, 2019).

Paint is a complex solvent mixture of pigments that add colour to surfaces of materials or equipment. According to Phulpoto *et al.* (2016) paints are made by mixing binders/additives (which adhere paints to surfaces) with pigments (which give the paint a colour and prevent corrosion) and solvents to make the paint spreadable. Paints are of different types. But they can be generally grouped into two based on the thinning reagent. Those thinned with mineral turpentine or other organic solvents are known as oil-based or solvent-based paints, while the ones that are thinned with water are referred to as emulsion paints or water-based vinyl or acrylic paints (Odokuma *et al.* 2013).

Though, the paint industry has brought revolution to the economy globally, some draw backs have been identified to be associated with the effluent generated during the production processes. According to Chukwujike *et al.* (2015), paint effluents contain high amounts of

heavy metals and organic pollutants; therefore the removal of these toxic pollutants from effluent before discharging to the environment and from raw water before public use is essential for the protection of health and environment. Industrial effluents not only contain nutrients that enhance the growth of crop plants but also have other toxic materials, which can pose serious environmental and health hazards (Chukwujike *et al.*, 2015).

Several methods have been selected to manage the pollutants present in industrial effluents, amongst which are chemical precipitations, conventional coagulation, reverse osmosis, ion-exchange, solvent extraction, membrane filtration, chemical precipitation, electro dialysis and lime coagulation, oxidation and reduction method (Chukwujike *et al.*, 2015), but the attendant high capital intensiveness coupled with reported environmental side effects of these physicochemical techniques warranted the search for a better alternative (Gulzar *et al.*, 2017; Shah, 2018).

Bioremediation of wastewater has been considered as a better alternative for their treatment (Ravikumar *et al.*, 2012; Phulpoto *et al.*, 2016; Alaguprathana and Poonkothai, 2017). According to Faryal and Hameed (2005), bioremediation involves the use of natural biota and their processes for pollution reduction to non-hazardous substances. Microbial communities are of basically significant in bioremediation of metal contaminated soil and water, because microbes possess the capacity to transform the structure and mobility of compounds and metal through reduction, accumulation, mobilization and immobilization (Faryal and Hameed, 2005). Thus, bioremediation is currently gaining serious attention in recent years. This is because it has been reported by environmental managers to completely mineralize, detoxify, and transform pollutants into environmentally friendly products (Gulzar *et al.*, 2017; Shah, 2018).

In recent times, bioremediation researchers have isolated a variety of microorganisms from paint effluents having the capability of degrading and remediating paints and paint-related wastes (Obidi *et al.*, 2009; Ravikumar *et al.*, 2012; Okunye *et al.*, 2013; Rosado *et al.*, 2013; Ishfaq *et al.*, 2015;). According to Phulpoto *et al.* (2016), most of these hazardous pollutant-polluted sites could serve as novel sources of choice for the isolation of microorganisms with biodegradation and bioremediation capabilities. Most studies by previous researcher focused on isolation of bacteria from paint wastewater, with little emphasis on fungi. However, studies have shown that fungi can also play key roles in biodegradation and deterioration of paints and coated surfaces (Ravikumar *et al.*, 2012; Okunye *et al.*, 2013; Rahim and Dawar, 2015).

Some fungi such as *Mucor*, *Aspergillus*, and *Penicillium* species have been found to possess the enzymatic machinery for the biodegradation of both soluble and insoluble organic compounds in waste water (Faryal and Hameed, 2005). This work was therefore undertaken to isolate paint-degrading fungi from paint contaminated sites and thus enlarge the arsenal of microorganisms used in the bioremediation of paint effluent contaminated environments.

MATERIALS AND METHODS

Sampling Location

The sampling locations are paint effluents contaminated sites around the production unit of two famous factories (A and B) both located in Ado Ekiti, the State capital of Ekiti, South Western part of Nigeria. Geographically, Ado-Ekiti is situated between latitude 7.667° N and longitude 5.250° E and bounded in the north by Kwara State and Kogi State while Osun State occupies the west and Ondo State lies in the south and extends to the eastern part (Fig 1). The population of the indigenes is about 2,384,212 and the inhabitants of the state are mainly farmers, artisans, traders, civil and public servants (Salau *et al.*, 2016). The study area is shown in figure 1.

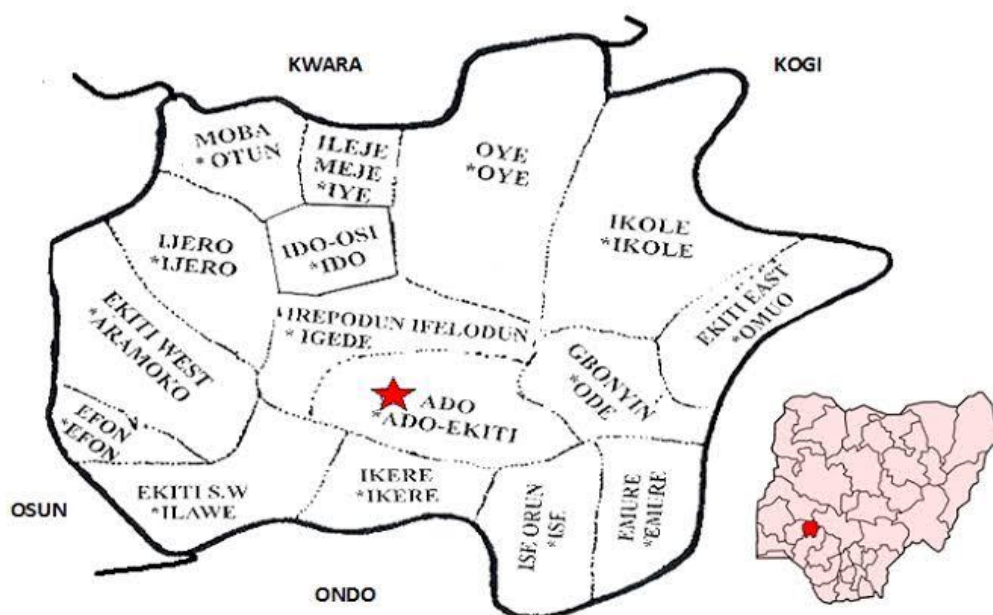


Figure 1: The map of Nigeria (pink) with location of Ekiti State in red. The star in Ekiti State indicates the location of the State Capital, Ado Ekiti (Salau *et al.*, 2016).

Sample Collection

Top soil samples from paint effluents contaminated sites within the vicinity of the production unit of the factories within Ado Ekiti, South West Nigeria, were collected in three replicates using soil auger into sterile beaker wrapped with aluminium foils. Also, fresh paint effluents were sampled from both industries using 1 liter sterile plastic containers from the main discharge units. The samples were transported in ice packed containers to Microbiology Laboratory of Ekiti State, University, Ado-Ekiti, for analyses within 24 hours of collection.

Media preparation and sterilization

All the glassware were washed and rinsed with 70% ethanol, wrapped with paper and sterilized in a hot air oven at 160°C for 2hrs. The media used for the isolation of the fungi was potato dextrose agar (Hi media). To prepare the media, thirty-nine grams (39g) of potato dextrose agar was weighed on a weighing balance and was mixed with 1000ml of distilled water in a conical flask. The conical flask was placed on a heating mantle for homogenization. After homogenization, it was then sterilized in an autoclave at 121°C for 15 min. Thereafter, 4ml of streptomycin was added aseptically (after the medium has cooled to the molten PDA to inhibit the growth of bacteria).

Physicochemical and Heavy Metal Analysis of paint Effluents

All samples were analyzed for some heavy metals (zinc, cadmium, and nickel) and physicochemical parameters using the methods of Apha, 2001; 2005 and Dagona, 2007. The major parameters analyzed included temperature, chemical oxygen demand, dissolved oxygen, biochemical oxygen demand, turbidity, odour, colour, total suspended solids, pH, conductivity, and total dissolved solids.

pH

The pH was determined by placing a pH probe (Hanna instrument C-99- USA) into the sample in a 250 ml conical flask and allowed to equilibrate for 3 minutes and pH meter was read and recorded accordingly.

Temperature

The temperature of water and effluent was also determined on the field by lowering a mercury thermometer (Hanna instrument C-99- USA) into the sample and allowing it to equilibrate for 4 minutes and the reading was taken to the nearest degree Celsius (°C).

Electrical conductivity (EC)

The electrical conductivity was determined by placing a conductivity probe (Hanna instrument C-99- USA) into the sample in a 250 ml conical flask and allowing it to equilibrate for about 3 minutes and the electrical conductance in micro second per centimeter ($\mu\text{s}/\text{cm}$) was recorded.

Dissolved Oxygen (DO)

Dissolved oxygen of the effluent samples was determined using Jenway Model 9070 (Hanna instrument C-99-USA) waterproof DO-meter. The protective cap of the DO meter was removed from the probe. Membrane module was taken and held in the vertical position. The probe was calibrated prior to measurement with the appropriate traceable calibration solution of 5% sodium sulphate in accordance with the manufacturer's instruction. The probe was immersed into the effluent samples to be analyzed and the readings were recorded at the point of sample collection.

Determination of Chemical Oxygen Demand (COD)

Fifty (50 ml) of sample was dispensed into a refluxing flask and several boiling stones were added. Then 0.1 g HgSO_4 was added to the solution and 5 ml of concentrated H_2SO_4 was also added to the solution. To ensure that HgSO_4 dissolved completely, the solution was swirled slowly while adding Sulphuric acid, then 0.1 g of Ag_2SO_4 was added to this solution and finally Potassium dichromate was added. Thorough mixing of the solution was done by swirling the flask in a water bath to prevent any volatile substances that may have escaped from the liquid state. The flask was then attached to a condenser and further cooling was carried out and 20 ml of sulphuric acid was added to the solution in the flask continuing cooling and swirling to mix the solution.

The solution was refluxed for 1 hour. A blank run (using 50 ml distilled water instead of sample) was simultaneously conducted with the same procedure after cooling; the solution was then transferred to an Erlenmeyer flask. The reflux flask was rinsed thrice, pouring the rinsing water to the Erlenmeyer flask. The solution was diluted to about 300 ml and about 8 drops of phenanthroline ferrous sulphate was added to the solution as an indicator. The solution was titrated against ammonium iron (II) sulfate (Mohr's salt) and the titre volume required for the colour change from blue-green to reddish blue was noted. The procedure was repeated for the blank run. Below was the formula used to calculate the COD:

$$\text{COD} = 8000 \times (\text{V}_{\text{bl}} - \text{V}_{\text{s}}) \times \left[\frac{M}{\text{Original vol. of sample taken in ml}} \right]$$

Where, V_{bl} = Titre volume for the blank

V_{s} = Titre volume for the sample

M = Molarity of Mohr's solution.

Determination of Biochemical Oxygen Demand (BOD₅)

Biochemical Oxygen Demand (BOD-5) was determined using DO HI9146 (Winkler) method of DO determination, Microprocessor Dissolved Oxygen Meter. The amount of sample to be analyzed was measured, clean calibrated thermometer was placed into the sample; temperature was stabilized at $20^\circ\text{C} \pm 1^\circ\text{C}$ in the refrigerator. DO instrument was turned on for 30-60 minutes. After aeration, 1 ml each of the potassium phosphate, magnesium sulphate, calcium

chloride, was diluted according to manufacturer's instruction. Dilution was placed at constant temperature to maintain the initial temperature until sample dilutions and analyses began. The initial and final (after 5 days \pm 4 hours) DO concentration of the sample was measured as D1 of each sample and each dilution blank. Temperature was checked using air incubator with laboratory thermometer to ensure that the temperature has been maintained. At the end of 5 days \pm 4 hours, BOD bottle was removed from incubator and the final DO concentration (D2) was measured. The DO1 uptake (DO2 days – DO5 days) in the dilution water should not be greater than 0.2 mg/l and preferably not more than 0.1 mg/l. For each test bottle meeting the 2.0-mg/L minimum DO depletion and the 1.0-mg/L residual DO. BOD₅ was calculated by using the formula below:

$$BOD5 (mg/l) = \frac{D1-D2}{P}$$

Where, D1= DO diluted sample immediately after preparation (in mg/l)

D2= DO diluted sample after 5 days of incubation at 20 °C \pm 1°C (in mg/l)

P= decimal volumetric fraction of sample used.

Determination of Total Suspended Solids (TSS)

Before sampling, glass fibre filters were prepared first by soaking them in distilled water, drying them at 103°C and weighing and recording their weight. Sample bottles were dried, and weighed glass fibre filters were poured into a filtering flask – wrinkled side up. Sample bottle was shaken first, and then water was poured on the pump. The amount of water needed to filter may change according to water conditions. One hundred ml of sample was filtered with paper with porosity 0.8 mm. Filtered quantity was recorded with volume of water filtered. Filter paper was dried from 103 °C to 105 °C, and was allowed to dry at room temperature, and weighed. It was dried again and re-weighed. This was repeated until the filter reached a constant weight. Final end weight was recorded. This increase in weight representing TSS was calculated by using the equation,

$$TSS = \frac{A-B \times 100}{C}$$

Where, A = End weight of the filter

B = Initial weight of the filter

C = Volume of water filtered

Total dissolved solids (TDS)

Total dissolved solid (TDS) was determined by evaporating the wastewater samples to dryness. In this method, 50 ml of sample was transferred to a weighed evaporating dish, and evaporated to dryness by heating for 1-2 hours at 180 °C to a constant weight. A total dissolved solid was calculated as follows:

$$mg/l \text{ of TDS} = \frac{mg \text{ residue}}{mL \text{ sample}} \times 1000$$

Determination of heavy metals in effluent Samples

Determination of Cu, Zn, Mn, Fe, Cr, Cd, Ni and Pb was carried out directly in each final solution using Standard methods as described by Dagona (2007). Each of the metals was analysed by using Atomic Adsorption Spectrophotometer (AAS -model-GBC-932 plus Chem Tech – USA).

The wastewater samples were digested as follows: 100 millilitre of the sample was transferred into a beaker and 5 ml concentrated HNO₃ was added. The beaker with the content was placed on a hot plate and evaporated down to about 20 ml. The beaker was then cooled and another 5ml concentrated HNO₃ was also added. The beaker was covered with watch glass and returned to the hot plate. The heating was continued, and then small portion of HNO₃ was added until

the solution appeared light coloured and clear. The beaker wall and watch glass were washed with distilled water and the sample was filtered to remove any insoluble materials that could clog the atomizer. The volume was adjusted to 100cm³ with distilled water, the result was read in mg/L.

Isolation of Organisms from Sample

The method of Oduola *et al.* (2018) was adopted. Briefly, one gram of the paint effluent contaminated soil sample was added into 9ml of sterile distilled water and the tube was shaken gently to ensure thorough mixing. After mixing, a serial dilution was then carried out by transferring 1ml from the first test-tube into the second test-tube that contains 9ml of distilled water. The second tube was mixed gently and 1ml was taken from the second tube into the third test-tube, and so on till the fifth test-tube. One milliliter (1ml) each from the third test-tube was pipetted using a new sterile pipette into two empty sterile Petri-dishes, one for nutrient agar and the other for potato dextrose agar. Also, 1ml each was pipetted from the fifth test-tube and poured into the sterile Petri-dishes to be used.

After the samples have been poured into the Petri-dishes, about 15ml of the sterile PDA medium was poured into the plates. After the plates had set, they then were incubated at 25°C for 3-5 days. Suspected colonies of *Penicillium* were then characterized using macroscopic colonial appearance and lactophenol cotton blue stain with reference to standard pictorial fungal atlas (Dorge, *et al.*, 2000; Adebayo-Tayo *et al.*, 2012; Onuorah *et al.*, 2015).

Characterization and identification of *Penicillium* isolates

The fungal isolates were characterized using cultural and morphological features such as colony growth pattern, conidial morphology, and pigmentation as described earlier (Oyeleke and Manga, 2008; Mailafia *et al.*, 2017). The identification of the isolated fungi was done using cotton blue in lactophenol stain. The identification was achieved by placing a drop of the stain on clean slide with the aid of a mounting needle where a small portion of the aerial mycelia from the representative fungi cultures was removed and placed in a drop of lactophenol. The mycelium was well spread on the slide with the needle.

A cover slip was gently placed with little pressure to eliminate air bubbles. The slide was then mounted and viewed under the light microscope with ×10 and ×40 objective lenses. The morphological characteristics and appearance of the fungal organisms seen were identified in accordance to previous standard pictorial fungal images (Dorge, *et al.*, 2000; Adebayo-Tayo *et al.*, 2012; Onuorah *et al.*, 2015).

Biodegradation of Paint Effluent Using *Penicillium* Isolates

The procedure described by Okoduwa *et al.* (2017) was adopted. The surface of 3 day old cultures of *Penicillium* sp. was flooded with normal saline. From the suspension, 5 ml of inoculum was collected and used to inoculate different concentrations of the effluent [(100 ml of effluent) as 100 % effluent, (75 ml of effluent plus 25 ml of tap water) as 75 % effluent and (50 ml of effluent plus 50 ml of tap water) as 50 % effluent]. These were contained in different 250 ml conical flask and properly labelled. A control flask without the fungal spore was also set up. They were kept in an orbital shaker for three weeks and maintained at 25 ± 2 °C. The experiments were set up in triplicate and at 100 %, 75% and 50% concentrations of the effluent. The pH was recorded every 48 hours.

Other parameters monitored weekly were BOD, DO, TDS, TSS, and COD to determine the extent of the biodegradation using previously described techniques (APHA, 2001; APHA, 2005; Dagona, 2007).

RESULTS AND DISCUSSION

The results of the physicochemical analysis of paint effluent-contaminated soil collected from both sampling locations (A and B paint warehouses) are shown in Table 1. Generally, the average ranges of the physicochemical parameters were; pH (5.60-6.25), organic carbon (2.28-4.70%), temperature (27 -35 °C), BOD (182-219 mg/L), COD (292-719 mg/L), sulphate (30-42 mg/L), nitrate (2.25-8.0 mg/L). When compared with the World Health Organization (WHO) and National Environmental Standards and Regulations Enforcement Agency (NESREA) standards, all the tested parameters were found to be within the maximum limits except for BOD and COD, which were significantly above the maximum levels of 30 and 80mg/L respectively. The colours of the paint contaminated soils did not differ significantly from the uncontaminated control samples. They were generally milky, and or light/dark brown depending on the sampling sites.

Table 1: Physicochemical Properties of soil from paint effluents contaminated sites

Soil sample	Location	Colour	pH	Organic Carbon (%)	Temp (°C)	BOD (mg/L)	COD (mg/L)	SO ₄ ²⁻ mg/L	NO ₃ mg/L
1	Control (soil free from paint)	Milky	6.25	2.28	27	182	292	30	2.25
2	Soil from paint warehouse A	Milky	6.20	4.27	35	200	402	35	3.6
3	10 m away from paint warehouse A	Milky	5.60	4.10	32	189	641	42	5.2
4	15 m away paint warehouse A	Light brown	5.70	4.70	32	219	530	32	3.3
5	20 m away from paint warehouse A	Dark brown	5.70	3.71	33	209	482	34	6.2
6	Soil sample from warehouse B	Dark brown	6.12	3.56	34	214	719	34	8.0
7	10 m away from warehouse B	Dark brown	6.10	4.01	29	198	529	33	4.3
8	15 m away from warehouse B	Light brown	5.93	4.12	30	201	482	30	5.4
9	Average		5.95	3.84	33	201.5	505.88	33.75	4.78
	*WHO limit, 2006	-	6.5-8.5		20-32	-	-	250	10
	*NESREA limit, 2009	-	6.0-9.0		<40	30	80	500	20

*WHO: World Health Organization, 2006 NESREA: National Environmental Standards and Regulatory Enforcement Agency, 2006

The quantity of total heterotrophic and paint degrading fungi was determined in soil samples collected from the two warehouses (Coded A and B) (Tables 2). The total heterotrophic fungal counts and paint utilizing fungal counts varied significantly with distances of sampling location from the contaminated sites. The population of the fungi was not directly proportional to the increase or decrease in distances from each warehouse. For total heterotrophic fungi (THF), the counts at 10 m away from location A had the highest number (5.32×10^4 cfu/g), while for location B, the highest count (17.00×10^4 cfu/g) was also recorded 10 m away from warehouse. However, the loads of fungi in and around the contaminated sites were significantly higher than those of the control (1.21×10^4 cfu/g). Generally, the density of actual paint hydrocarbon-utilizing fungi was relatively lower than the heterotrophic groups.

The morphological and cultural characteristic identification tests of fungal isolates from paints effluents in Ado-Ekiti are shown in Table 3. The results indicated that sixteen (16) fungal isolates belonged to six (6) different genera and were identified as *Penicillium* spp., *Fusarium* spp., *Geotrichum* spp., *Rhizopus oryzae*, *Aspergillus niger* and *Mucor* spp. However the frequency distribution (%) of the isolated organisms (Table 4) showed that *Penicillium* spp. had

the highest occurrence (43.75%), followed by *Fusarium* spp (18.75%), *Rhizopus oryzae* (12.5%), *Aspergillus niger* (12.5%) and both *Geotrichum* spp and *Mucor* spp had 6.25%.

Table 2: Mean densities of total heterotrophic fungi (THF) and Total paint utilizing fungi (TPUF) isolated from the paint effluent contaminated sites

S/N	Samples	THF (CFU/g) × 10 ⁴	TPUF (CFU/g) × 10 ³
1	Control (soil free from paint)	1.21	0.27
2	Soil from paint warehouse A	1.45	2.45
3	10 m away from paint warehouse A	5.32	1.60
4	15 m away paint warehouse A	4.21	0.76
5	20 m away from paint warehouse A	2.21	0.60
6	Soil sample from warehouse B	2.50	3.50
7	10 meter away from warehouse B	17.00	1.80
8	15 meter away from warehouse B	2.30	2.90

Table 3: Morphological characteristics of fungi isolated from paint effluents contaminated sites

S/N	Isolates	Growth	Front view	Back view	Nature of hyphae
F1	<i>Penicillium citrinum</i>	Rapid	Dark green	Pale yellow	Septate
F2	<i>Fusarium</i> sp.	Abundant	Pale brown	dark zonation	Septate
F3	<i>Penicillium chrysogenum</i>	Rapid	Blue green	Red with yellow	Non-Septate
F4	<i>Penicillium frequentans</i>	Rapid	Green with yellow	Red with yellow	Septate
F5	<i>Penicillium oxalicum</i>	Rapid	Dark green	Cream-yellow	Arial and smooth
F6	<i>Geotrichum</i> sp.	Rapid	White	White	Septate
F7	<i>Penicillium oxalicum</i>	Rapid	Dark green	Cream-yellow	Arial and smooth
F8	<i>Rhizopus oryzae</i>	Abundant	Brown	Dark brown	Non-septate
F9	<i>Aspergillus niger</i>	Rapid	Grey black	Dark brown	Non-septate
F10	<i>Aspergillus niger</i>	Rapid	Grey black	Dark brown	Non-septate
F11	<i>Penicillium notatum</i>	Rapid	Olive green	Off white	Smooth
F12	<i>Rhizopus oryzae</i>	Abundant	Brown	Dark brown	Non-septate
F13	<i>Fusarium</i> sp.	Abundant	Pale brown	dark zonation	septate
F14	<i>Penicillium funiculosum</i>	Rapid	White with brown	Creamy yellow/red	Arial
F15	<i>Fusarium chlamydosporum</i>	Abundant	Deep pink	Carmin red	Aerial
F16	<i>Mucor</i> sp.	Rapid	White	Whitish	Ellipsoidal

Table 4: Frequency distribution of fungi isolated from paint effluents contaminated sites

S/N	Isolate	Frequency	Percentage (%)
1	<i>Penicillium</i> spp	7	43.75
2	<i>Fusarium</i> spp	3	18.75
3	<i>Geotrichum</i> spp	1	6.25
4	<i>Rhizopus oryzae</i>	2	12.50
5	<i>Aspergillus niger</i>	2	12.50
6	<i>Mucor</i> spp	1	6.25
	Total	16	100%

Preliminary biodegradation study showed that significant degradation was observed with the *Penicillium* species and thus were further assayed to determine the most efficient species. The quantities of paint degraded by a known weight of *Penicillium* species over a period of twenty

one (21) days at room temperature are as shown in Figure 1. From the result, *Penicillium notatum* showed the greatest paint degradation potential from day 7 to day 21 while *Penicillium citrium* recorded the least among the isolates on day 7. Furthermore, *Penicillium oxalicum* was the least at day 14 but increased markedly after day 21. Moreover, at day 21 *Penicillium notatum* displayed the greatest paint degradation capacity and *Penicillium chrysogenum* showed the least among fungal isolates.

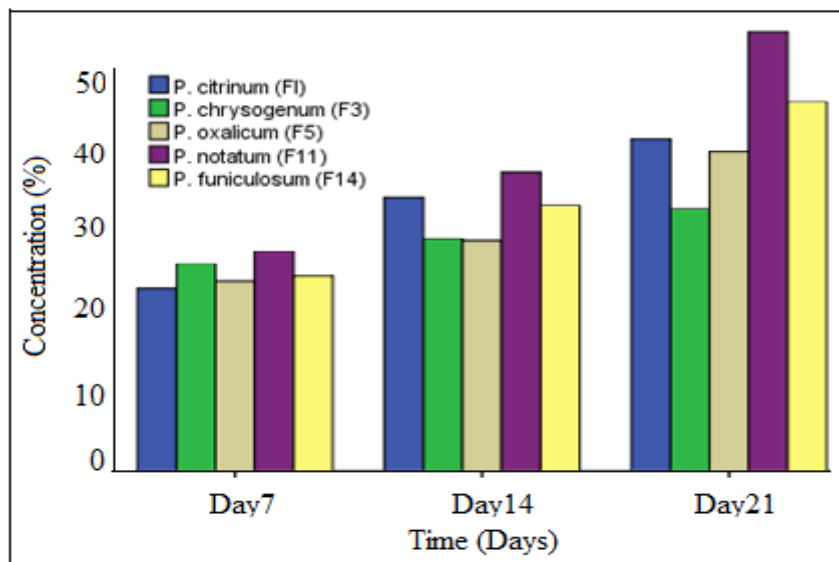


Figure 1: Paint degradation by fungal isolates

Environmental pollution has continued to produce negative impacts on the ecosystem. As a consequence environmentalists are heavily saddled in recent times with the responsibility of finding a lasting solution to this mayhem. The study examined some physicochemical and the biodegradation of paint effluents using fungal isolates from paint contaminated soil, with a view to expanding the arrays of alternative solutions available for addressing this global issue. The results obtained from the physicochemical analysis showed that the pH of paint-effluent impacted soils were slightly acidic, though within the standard limit of 6-9 (NESREA, 2009). Hence, in the study location, there was a slight increase in the acidity of the environment, thereby favoring the activities of fungi, whose optimal biochemical activities fall within such range. Similarly, the average temperature of the samples was within the limits set by NESREA but slightly above that of WHO, which is between 20-32°C.

Previous studies attributed the slight differences to heat exchange generated from machinery during and after production processes and discharges (Onuegbu *et al.*, 2013). This study also revealed that BOD and COD of the effluents were above the standard limits. Both BOD and COD are useful indicators of levels of pollution of water by organic compounds, but COD is less specific, because it measures both chemically and biologically oxidized compounds (Sawyer *et al.*, 2003). COD determines the oxygen equivalent needed to chemically oxidise the organic material in wastewater, hence the ratio of COD: BOD is an important guide to monitor the concentration of organic matter found in wastewaters (Akpore and Muchie, 2011). The BOD of wastewater effluents is an effective measure of the immediate impact on the oxygen levels of the receiving water (Sawyer *et al.*, 2003). The high level of BOD and COD obtained in this study therefore suggest that the effluents from the paints industries are highly polluted by organic compounds.

According to Aniyikaiye *et al.* (2019), the presence of pure acrylic and styrene acrylic binders, cellulose thickener and organic pigment components of paint must have elevated the BOD and COD of the wastewater. The implication of this outcome is that greater amount of organic matter or nutrients are readily available for aerobic bacteria leading to dissolved oxygen depletion and ultimately, death of aquatic organisms. The BOD/COD obtained in this study were relatively lower than values reported for industrial wastewater from Lagos (162.8 - 840.6), Morocco (828 mg/L), Ethiopia (600 mg/mL) and India (535.8 mg/mL) (Ram *et al.*, 2011, Aboulhassan *et al.*, 2014 ; Tesfalem *et al.*, 2017; Aniyikaiye *et al.*, 2019). Suphate and Nitrate were significantly below the maximum limits, hence, might not impact adversely on the balance of the ecosystem.

The total heterotrophic fungal counts and paint utilizing counts varied significantly with distances of sampling location from the contaminated sites. For THF, the maximum counts were obtained at a distance of 10 m away from locations A and B. This finding suggests that favorable conditions for fungal growth around the paint wastewater contaminated site are obtainable at a distance of 10 m away. At this region, probably the right level of the organic and inorganic components of the effluents needed for microbial growth is readily available for microbial utilization. According to Dey *et al.* (2004) and Phulpoto *et al.* (2016), majority of microorganisms possess the potential to utilize some components of the water-based paint effluents as substrate for active growth and development. Similarly, previous reports found that paint effluents contain several biodegradable organic and inorganic nutrients which can impact positively on the bacterial community of the receiving soil environments (Noorjahan, 2016).

Additionally, the rapid change in fungal community structure coupled with the fast rate of oil degradation, suggest the presence of a pre-adapted oil-degrading microbial community and sufficient supply of nutrients (Hamamura *et al.*, 2008). Hence, the finding from this study is in agreement with the reports of previous research. Furthermore, the findings from the decrease in fungal population with respect to the distances of collections of the soil samples suggest that that the growth of fungi in water-based paints is dependent on concentrations of the biodegradable components within the surrounding.

The level of heterotrophic fungi recorded in this study for the paint contaminated soils is in agreement with previous reports (Ishfaq *et al.*, 2015) and the relatively reduced populations of the fungal paint utilizers could be attributed to the toxic effect of certain components of paint at relatively high concentrations (Rahim *et al.*, 2015; Phulpoto *et al.*, 2016). The possible reason for the rise in paint degradation could be attributed to the increase in cell number during the degradation process demonstrating the ability of utilizing these paints as potential the energy source for the fungi. This result agrees with the work reported by Khan and Rizvi (2011) and Abioye *et al.*, (2012) who isolated microorganisms from oil contaminated soil. Previous report had found some of the fungal isolates as potential hydrocarbon utilizers (April *et al.*, 2000; Chaudhry *et al.*, 2012).

In this study, the quantities of paint degraded revealed that *Penicillium notatum* demonstrated the greatest paint degradation ability from day 7 to day 21, while *Penicillium citrium* showed the least among the bacterial isolates on day 7. One of the major indicators of microbial utilization of organic matter is an increase in the population of the microbes in the growth medium. Previous study had shown that the growth of microorganisms in organic pollutants is often indicated by an increase in turbidity and decrease in pH (Nwinyi *et al.*, 2011). The reduction in the population of the biodegrading microorganism could be attributed to the accumulation of toxic metabolites from th metabolized substrated as pointed out in earlier report

(Vidhya and Thatheyus, 2013). The capacity of the fungi, *Penicillium* sp. To break down the paint hydrocarbon coupled with the rise in their biomass suggest the paint is a good source of energy and carbon source. This finding is in consonance with earlier submission on the capacity of *Penicillium* to effectively utilize aromatic hydrocarbons (Leitao *et al.*, 2007). The isolation of the *Penicillium* sp.

From contaminated soil corroborates previous studies that active hydrocarbon degraders could be source from polluted soil environments (Hofrichter and Scheibner, 1993; Hammel 1995; Leitao *et al.*, 2007). This could probably be the reasons why most interior painted surfaces are frequently colonized by moulds, with yeasts growing in areas with excessive moisture (RaviKumar *et al.*, 2012). Hence, it was concluded that fungi, including *Penicillium* spp. were amongst the main contaminants and were able to break down and penetrate the paint film using their special enzymatic machinery (Berk *et al.*, 1957; Bravery, 1988; Smith, 1978; RaviKumar *et al.*, 2012). The use of microorganisms, such as fungi, for the removal of paints from industrial effluents offers considerable advantages; this process is relatively inexpensive, running costs are low and the end products are completely mineralized with no toxicity.

CONCLUSION AND RECOMMENDATION

This study has investigated some physicochemical and the biodegradation of paint effluents using fungal isolates from paint contaminated soil. *Penicillium* spp. was shown to possess the highest frequency of occurrence in paint contaminated site, because they were more resistant to the oil-based paints at elevated concentrations. This could be one of the reasons why *Penicillium* species predominated other fungi species inhabiting the oil-based- paint-polluted environment. The most efficient *Penicillium* spp. could be used in bioremediation of paint contaminated soil. Based on the findings of this study. It is recommended that *Penicillium notatum* which exhibited the highest biodegradation ability in this study be mass-produced and the mechanisms of action studied for possible engineering of the organism for better performance.

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