

## Enhanced Microbial Removal of Residual Oil in Palm Oil Mill Effluent in the Presence of Organic Supplement

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**Abstract:** The residual oil in palm oil mill effluent (POME) poses a problem in the environment because palm oil is not easily degraded or utilized by microorganisms. A study was carried out to investigate the ability of three bacteria, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus subtilis* isolated from soil contaminated with palm oil mill effluent to utilize the residual oil in POME in the presence of bunch ash as an organic supplement. Results of the study showed that *Pseudomonas aeruginosa* gave the highest percentage reduction (79.5%) in fatty acid concentration in the palm oil mill effluent. *Bacillus subtilis* gave 72.6% reduction while *Staphylococcus aureus* gave 65.1% reduction in fatty acid content of the effluent. Effluent samples without the organic supplement recorded very low percentage reduction in fatty acid with *Pseudomonas aeruginosa* (0.5%), *Bacillus subtilis* (0.4%) and *Staphylococcus aureus* (0.2%). The results of this study suggest that the biological removal of residual oil in palm oil mill effluent and bioremediation of POME contaminated soil can be enhanced using a natural supplement such as bunch ash.

**Key words:** Organic Supplement • Enhanced • Microbial Removal • Residual Oil • POME

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### INTRODUCTION

Palm oil production is a great economic activity in areas where palm trees grow readily. The large production of palm oil in these areas has given rise to the provision of palm oil mills. Palm oil mill is a product of modern technology and an approach to the problem of poor utilization of natural resources. Large quantities of water are used in the extraction of palm oil from the mesocarp of oil palm fruits and much of this water constitutes palm oil mill effluent [1]. Palm oil mill effluent contains high organic matter including suspended solids and residual oil.

In most developing countries, untreated palm oil mill effluents are discharged to the environment from palm oil mills. Raw palm oil mill effluent (POME) is an objectionable industrial effluent capable of polluting streams, rivers or surrounding land [2]. When POME is discharged on soil, there are changes in soil structure, which in turn affect the arrangement of soil aggregates, thereby affecting the movement of microorganisms in

the soil [3]. It also affects the soil texture which determines the water retention ability and the frequencies of cation exchange in the soil [4]. These changes make the soil which was previously fertile before exposure to the effluent pollution to lose its productive potential. The high organic matter content of POME results in dissolved oxygen depletion in aquatic environment which endangers the life of aquatic organisms [5, 6].

Although various innovative treatment technologies and processes have been developed for palm oil mill effluent [7, 8], the processes are expensive to maintain, difficult to operate and require expertise [9]. Moreover, the residual oil in POME poses a problem in the maintenance of the systems and contributes to environmental pollution because palm oil is not easily degraded or utilized by microorganisms. Hence, there is the need for alternative and more efficient means of managing palm oil mill effluent that is easy and cheap to handle which will enhance microbial removal of residual oil in the effluent.

The aim of this research work is to investigate the ability of three bacteria namely *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Staphylococcus aureus* isolated from soil contaminated with palm oil mill effluent, to utilize the residual oil in POME in the presence of bunch ash as a natural supplement.

## MATERIALS AND METHODS

**Collection of Materials:** The palm oil mill effluent used for this study was collected from a palm oil mill located in Ohaji/Egbema Local Government Area of Imo State, Nigeria. The effluent was collected in five liter plastic containers from effluent storage tank. Dried and defruited palm bunch used to prepare bunch ash was collected from the palm oil mill. The bunch ash was obtained by burning 2kg of dried and defruited oil palm bunches into ash in the presence of air. The ash sample formed was collected and stored in a sterile container.

**Isolation and Characterization of the Organisms:** The microorganisms *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Staphylococcus aureus* used for this study were isolated from soil contaminated with palm oil mill effluent in Ohaji/Egbema, Imo State, Nigeria. The organisms were properly identified using various biochemical tests and their cultural and morphological characteristics in accordance with prescribed microbiological standard and procedures [10].

**Determination of Utilizing Ability of the Isolates for Residual Oil in POME:** Three pairs of 250ml glass cylinders containing 200ml untreated palm oil mill effluent were used for the experiment. Three of the glass

cylinders containing the effluent were inoculated with different organisms but without the organic supplement. This served as the control test. The remaining three cylinders containing the effluent were added 10g of bunch ash as organic supplement and inoculated with different organisms. The set up was mounted on a shaker incubator at ambient temperature for 15 days. The residual oil was determined as the concentration of fatty acid in the effluent. The determinations were done at 3 day interval for 15 days.

**Determination of Fatty Acid Concentration in the Effluent:** The traditional method for the determination of free fatty acids in palm oil through titration against potassium hydroxide in hot 2-propanol solution by using phenolphthalein as indicator [11] was adopted.

## RESULTS AND DISCUSSION

The presence of bunch ash as an organic supplement enhanced microbial utilization of fatty acids in palm oil mill effluent (Figs. 1 to 3). The three organisms, *Staphylococcus aureus*, *Bacillus subtilis* and *Pseudomonas aeruginosa* in the presence of the organic supplement remarkably reduced the fatty acid content of palm oil mill effluent from its original concentration of 6.494mg/L to 2.267mg/L, 1.778mg/L and 1.333mg/L respectively (Table 1). There was poor utilization of the fatty acids in the absence of the organic supplement. The residual fatty acid concentrations recorded with *Staphylococcus aureus*, *Bacillus subtilis* and *Pseudomonas aeruginosa* in the absence of the organic supplement were 6.480mg/L, 6.466mg/L and 6.458mg/L respectively (Table 2). *P. aeruginosa* gave the highest

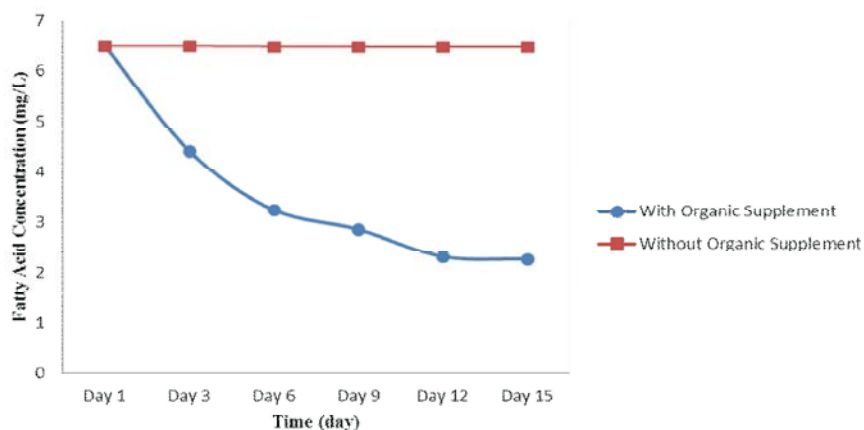


Fig. 1: Rate of utilization of fatty acid content in POME by *S. aureus*

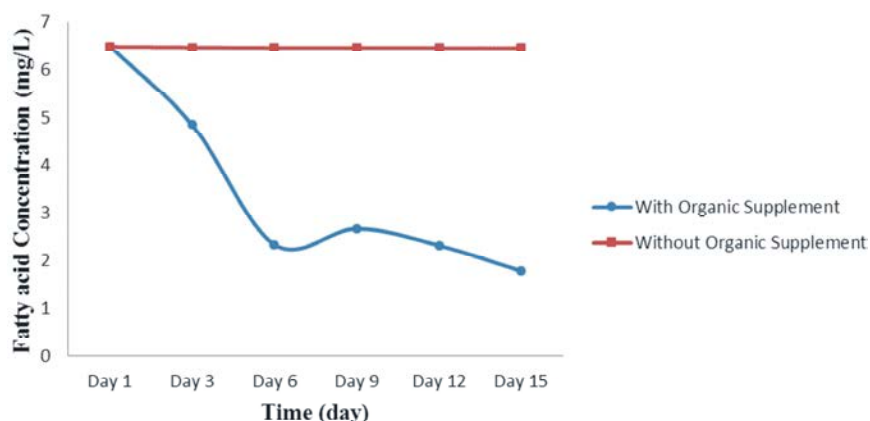


Fig. 2: Rate of utilization of fatty acid in POME by *B. subtilis*

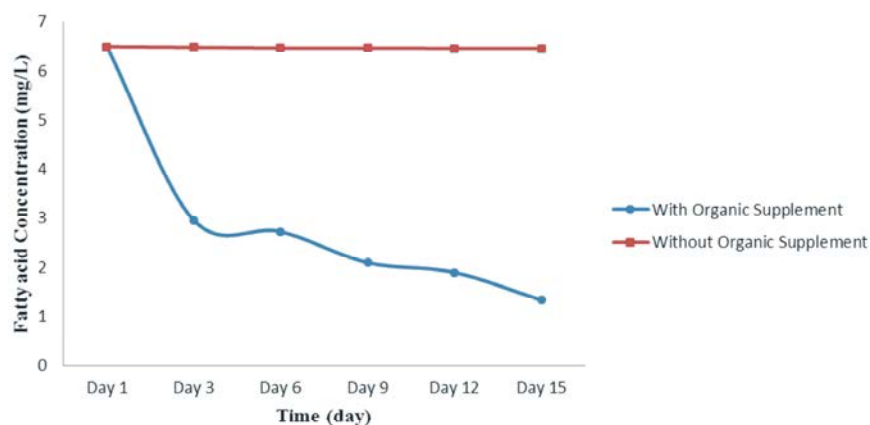


Fig. 3: Rate of utilization of fatty acid in POME by *P. aeruginosa*

Table 1: Residual concentration of fatty acid in POME containing organic supplement after 15-day incubation with various organisms at ambient temperature

Organism used	Fatty acid concentration (mg/L)					
	Day1	Day 3	Day 6	Day 9	Day 12	Day 15
<i>S. aureus</i>	6.494	4.416	3.247	2.857	2.316	2.267
<i>B. subtilis</i>	6.494	4.857	2.338	2.667	2.316	1.778
<i>P. aeruginosa</i>	6.494	2.961	2.727	2.095	1.895	1.333

Table 2: Residual concentration of fatty acid in POME without organic supplement after 15-day incubation with various organisms at ambient temperature

Organism used	Fatty acid concentration (mg/L)					
	Day1	Day 3	Day 6	Day 9	Day 12	Day 15
<i>S. aureus</i>	6.494	6.493	6.486	6.481	6.481	6.480
<i>B. subtilis</i>	6.494	6.482	6.472	6.470	6.468	6.466
<i>P. aeruginosa</i>	6.494	6.480	6.466	6.463	6.460	6.458

Table 3: Percentage reduction in fatty acid content in POME by the various organisms

Organism Used	Percentage reduction without organic supplement (%)	Percentage reduction with organic supplement (%)
<i>S. aureus</i>	0.2	65.1
<i>B. subtilis</i>	0.4	72.6
<i>P. aeruginosa</i>	0.5	79.5

percentage reduction of 79.5% in fatty acid content of the effluent while *B. subtilis* and *S. aureus* gave 72.6 and 65.3% reduction respectively (Table 3). Effluent samples without organic supplement had very low percentage reduction in fatty acid with *P. aeruginosa* giving 0.5%, *B. subtilis*, 0.4% and *S. aureus*, 0.2%.

Organic and inorganic supplements have been demonstrated to enhance bioremediation processes [12, 13]. Positive effect of oil palm bunch ash on the bioremediation of oil polluted soil have been widely demonstrated [14-16]. These reports and the observations made in this present study could be attributed to the process of biostimulation. This involves the modification of the environment to stimulate existing bacteria capable of bioremediation [17]. This can be achieved by the addition of various forms of limiting nutrients and electron acceptors, such as phosphorus, nitrogen, oxygen or carbon which are available in quantities low enough to increase microbial activity [18-20]. It has been suggested that with good concentrations of nitrate, phosphate and potassium present in bunch ash, it may be used in enhancing the degrading potentials of hydrocarbon degraders since the mentioned parameters are applied in bioremediation [21].

Apart from its stimulating effect on the microorganisms, the emulsifying action of bunch ash on the residual oil in POME also may be responsible for the enhanced removal of the oil in the effluent by the microorganisms. Surface-active agents (surfactants) increase the oil-water interfacial area by emulsification. Surfactants have affinity for both oil and water so that oil tends to spread and be easily dispersed into small globules commonly called emulsion [22]. In the same vein organic ash such as bunch ash and green plantain peel ash react with palm oil to produce an organic mixture which allows the oil to be emulsified or dispersed [23]. Hence bunch ash acts as dispersant when added to palm oil mill effluent. As dispersant, it is expected to enhance microbial degradation of residual oil in palm oil mill effluent.

### CONCLUSIONS

The stimulating effect of organic supplements on microbial activity in palm oil mill effluent was highly manifested in this study. The presence of bunch ash as an organic supplement enhanced microbial removal of residual oil in POME. This research work could serve as a useful strategy in bioremediation process and biological treatment of palm oil mill effluent.

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