



Biodegradation of Polyethylene Sheet by *Acinetobacter calcoaceticus* with Thermostable Enzyme Activity

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Abstract: Polyethylene is a massively produced, and utilized plastic polymer with numerous research on the microbial degradation however, studies on enzyme activities in the degradation process are scarce. This study aims to produce and characterize manganese peroxidase (MnP), lignin peroxidase (LiP), and esterase from *Acinetobacter calcoaceticus* isolated from a local dumpsite in Ondo State, Southwest Nigeria in a polyethylene-based medium (PBM). Furthermore, the biodegradation efficiency of *A. calcoaceticus* on an unmodified polyethylene sheet was evaluated via a Fourier transform infrared spectrophotometer (FTIR). The result showed *A. calcoaceticus* produced optimum activities of MnP (7.92 U/mg), LiP (8.26 U/mg), and esterase (5.70 U/mg) on the tenth day of cultivation. The physicochemical studies showed optimal activity for MnP at 60 °C and pH 7, for LiP at 60 °C and pH 5, and for esterase at 50 °C and pH 9. The activities of LiP and esterase were inhibited by Cu²⁺, and Hg²⁺ and enhanced by Mg²⁺ and Ca²⁺ while MnP activity was only elevated by Mn²⁺. Notably, EDTA chelated the activities of all the enzymes. There were shifting's at different wavelengths, and aromatic OH functional group was seen in the biodegraded PE FTIR spectra. The results shows *A. calcoaceticus* has the ability to produce thermostable MnP, LiP, and esterase activities and hold great prospects that can be utilized for industrial and domestic remediation of PE wastes.

1. Introduction

Polyethylene (PE) is a thermoplastic made of ethane monomer (Osarumwense *et al.*, 2020). It is used to manufacture shopping bags, nylons, cups, plates, and several industrial packaging materials (Liu *et al.*, 2019). Polyethylene is an extensively used plastic polymer with a vast rising rate of production, however, despite the numerous domestic and industrial advantages, improperly disposed PE wastes persist and adversely impact both the aquatic and terrestrial environments (Eze *et al.*, 2024). Conventional ways of handling the PE wastes includes recycling, landfilling, and incineration menace but alternate environmentally friendly methods are essential due to their cost, (Chamas *et al.*, 2020). Biodegradation has then been adopted as an accepted and effective environmentally friendly technique for eradicating water and land pollution caused by PE (Amobonye *et al.*, 2021).

Biodegradation involves the application of biological emissaries such as plants, bacteria, and fungi) which are readily available to remediate PE wastes and release CO₂, H₂O, and CH₄ as end

products (Wei *et al.*, 2020). During PE biodegradation, abiotic factors (ultra-violet (UV) radiation or heating) act as bio-deteriorating agents on the chemical and physical properties of PE, after which the microbes produce enzymes to break the polyethylene polymer into subunits that can be ingested into the microbial cells (Maurya *et al.*, 2020). The ingested PE-monomers are finally metabolized to generate adenosine triphosphate (ATP) for cellular activities, while CO₂ and H₂O are released as environmentally friendly waste products (Pathak and Navneet 2017), (Salinas *et al.*, 2023). Studies has unveiled bacterial and fungal strains to biodegrade PE, these include *Bacillus* (Verma and Gupta, 2019), *Streptomyces* (Danso *et al.*, 2019), *Pseudomonas* (Hou *et al.*, 2022), *Aspergillus* (Gajendiran *et al.*, 2017), and *Penicillium* (Taghavi *et al.*, 2021). However, research on microbes with unique growth, enzymatic activities, and biodegradative potentials on polyethylene are still necessary especially unmodified polyethylene (Salinas *et al.*, 2023), (Zuriash *et al.*, 2023).

Acinetobacter calcoaceticus is a gram-negative bacteria that has been revealed to dominate among bacterial populations with hydrocarbon-degrading potentials and has been reported to have the ability to produce different enzymes (Nwaguma *et al.*, 2016). *Acinetobacter* spp has also been used to degrade different polymers hence it was peak used for this study (Yan *et al.*, 2022).

Esterase is an hydrolytic enzyme produced by microbes to catalyzes the cleavage of bonds found in esters which are intermediate product of PE bio-degradation (Koshti *et al.*, 2018) while lignin and manganese peroxidase are oxidative enzymes with bio-degradative activities on recalcitrant polymers with high cause of environmental pollution (e.g. plastic, textile effluents, and crude oil) (Sunil *et al.*, 2020), (Yan *et al.*, 2022). Although *Acinetobacter calcoaceticus* has been reported to degrade different polymers, study on the biodegradative and enzymatic activities on unmodified PE are scarce. The physicochemical properties of the enzymes released are also limited. Hence, this study aimed to characterize MnP, LiP and esterase produced by *Acinetobacter calcoaceticus* in a polyethylene base medium, and determine the biodegradation efficiency of *Acinetobacter calcoaceticus* on an unmodified polyethylene sheet.

2. Materials and Methods

2.1 Polyethylene preparation

Trashed water sachet nylons (100 pieces) were obtained from waste bins at different cafeteria in Elizade University. The water sachet nylons were confirmed as polyethylene by the Chemistry Unit, Physical and Chemical sciences Department, Elizade University. The PE sheets were minced into small particle sizes and decontaminated by soaking in distilled water and 90% ethanol for 30 min. (Azeko *et al.*, 2015).

2.2 Culture Preparation and Production of MnP, LiP and esterase

Acinetobacter calcoaceticus maintained on nutrient agar slant at 4°C was obtained from our laboratory for this study. *Acinetobacter calcoaceticus* culture was primed in 200 mL sterile nutrient broth containing peptone (2.0 % w/v), NaCl (2.0 % w/v), beef extract (0.60 % w/v) and yeast extract (0.60 % w/v) in a shaking incubator at pH 7.0, 37°C, and 180 rpm for 12 h . Four hundred (400 mL) PE-based medium (PBM) was prepared by introducing 0.05g of polyethylene particles as the sole carbon source into mineral salt medium containing NH₄NO₃ (0.04 % w/v), KH₂PO₄ (0.04 % w/v), KH₂PO₄·12H₂O (0.04 % w/v), NaCl (0.16 % w/v), KCl (0.16 % w/v), CaCl₂·2H₂O (0.02 % w/v), MgSO₄ (0.04 % w/v) and FeSO₄·7H₂O (0.0004 % w/v). Ten percent (v/v) *Acinetobacter* culture was also added into the PBM and incubated at 180 rpm, 37 °C and pH 7.0 for 30 days to study the growth

and production of enzyme by *A. calcoaceticus*. The experiment set was replicated thrice. PBM was prepared afresh for the characterization studies of the enzymes.

2.3 Manganese peroxidase assay

Manganese peroxidase (MnP) activity was investigated by determining the rate of oxidation of 1.8 mM 2,2'-azino-di-[3-ethyl benzothiazoline-6-sulphonic acid] (ABTS) buffered with 0.1M Tris-HCl buffer (pH 7) in the presence of 2.5 mM H₂O₂ (Ogunjemite *et al.*, 2023). The reaction was monitored at 414 nm for 5 minutes in a visible spectrophotometer. The reaction mixture (6 mL) contained 2 mL ABTS, 2 mL of culture filtrate, and 2 mL H₂O₂. One unit (U) of manganese peroxidase activity was defined as the amount of enzyme oxidizing 1 μmol ABTS per minute at pH 7.0 and 30 °C with a molar extinction coefficient for the ABTS radical cation (the reaction product) of ε_{414 nm} = 31100 M⁻¹ cm⁻¹.

2.4 Lignin peroxidase assay

Lignin peroxidase activity was determined by measuring the rate of oxidation of pyrogallol to purpurogallin in the presence 0.15 M H₂O₂ (Evans *et al.*, 2015). The reaction mixture (6 mL) contained 2 mL of pyrogallol (1 mM) in 5 mM Sodium acetate buffer at pH 5.0, 2 mL of culture filtrate and 2 mL H₂O₂. The reaction mixture was monitored at 450 nm for 10 minutes. One unit (U) of lignin peroxidase was defined as the amount of enzyme required to oxidizing 1 μmol of pyrogallol at pH 5.0 and 30 °C with a molar extinction coefficient for pyrogallol at ε_{450 nm} = 2470 M⁻¹cm⁻¹.

2.5 Esterase assay

Esterase activity was detected by measuring the release of p-NP butyrate (Ramnatha *et al.*, 2017). The substrate mixture contained 0.5 mM p-NP butyrate (C4) in methanol, 50 mM tris-HCl buffer (pH 8) and 0.1% Triton X-100. The reaction mixture contained 200 μl of substrate mixture and 20 μl of the crude supernatants incubated at 37 °C for 1 h. Enzyme activity was determined by measuring the release of p-NP at an absorbance of 405 nm. One unit (U) of enzyme activity was defined as the amount of enzyme required to release 1 nM of p-NP per min under the assay conditions 405 nm (ε_{405 nm} ¼ 18,000 nm⁻¹M⁻¹).

2.6 Determination of protein content

Protein concentration was determined using bovine serum albumin (BSA) (Olajuyigbe *et al.*, 2016). In the assay, 200 fold diluted dye reagent was pipetted into 10 uL of sample solution. The reaction mixture was then incubated at room temperature for 15 min for proper color development. The absorbance was measured at 595 nm against blank. The specific activity of manganese peroxidase was expressed as U/mg protein.

2.7 Characterization of MnP, LiP and esterase produced from *A. calcoaceticus*

2.7.1 Effect of pH on activity and stability of MnP, LiP and esterase

The effect of pH on MnP, LiP and esterase was assessed over a range of pH 3.0 – 11.0 (glycine-HCl (pH 3.0), sodium acetate (pH 5.0), Tris-HCl (pH 7.0) and glycine-NaOH (pH 9.0 and 11.0). The activities of MnP, LiP and esterase were investigated using the standard assay method earlier described. The stability of the pH was ascertained by incubating each enzyme at a different pH for 180 minutes at room temperature. The residual activities of enzymes were assessed using the described standard enzyme activity assay.

2.7.2 Effect of temperature on activity and stability of MnP, LiP and esterase

The activities of MnP, LiP, and esterase were evaluated by incubating the assay mixture at 30 °C to 80 °C for 35 minutes after which enzyme activities were recorded. Enzyme stability at the selected temperature was assessed at 30-minute intervals for 3 hours of incubation and residual activity was determined using the described standard enzyme activity assay.

2.7.3 Effect of metal ions and EDTA on activity of MnP, LiP and esterase

Metallic chlorides of selected concentrations (1, 5 and 10 mM) were used to investigate the effects of some cations (Ca^{2+} , Cu^{2+} , Mg^{2+} , Mn^{2+} , Hg^{2+}) on MnP, LiP and esterase activities. Ethylene diamine tetraacetic acid (EDTA) was also prepared in selected concentration (5 and 10 mM) to determine the effect of EDTA on the crude enzyme. Each metallic chloride was added to the reaction mixture and incubated for 30 minutes at the obtained optimum temperature and pH of the enzyme, thereafter enzyme activity was determined.

2.8 Biodegradation efficiency of *A. calcoaceticus* on polyethylene sheet

The biodegradation potentials of *A. calcoaceticus* on polyethylene was evaluated using 0.15 g PE sheet of 3 cm X 2 cm. The PE sheets were sterilized in a solution of 90% (v/v) ethanol and sterile water. The sterile 0.15 g PE sheets were then aseptically added into a conical flask with freshly prepared 500mL *A. calcoaceticus* culture. Conical flask containing only PE sheets in 250mL sterile nutrient medium was used as the control. The conical flasks were tightly covered with aseptic cotton wool with aluminum foil and incubated at pH 7.0 and 50 °C, and 180 rpm for 30 days. The experiment was in triplicate. At the end of the experiment, PE sheets were retrieved from the in vitro biodegradation setup and washed with several milliliters of aseptic distilled water. The PE sheets were air-dried overnight until a constant weight was obtained and they were subsequently analyzed. Fourier transform infrared (FTIR) spectrophotometer (Shimadzu FTIR spectrophotometer) was used to investigate variations in bonds and structure of the bio-degraded PE sheet compared to control PE sheet (Peng *et al.*, 2020).

2.9 Statistical analysis

Results are recorded as mean \pm standard deviation. The data were analyzed using the one-way analysis of variance. The mean values were correlated with the Duncan test and statistical package for social sciences (SPSS) by IBM version 16 was used.

3. Results and Discussion

3.1 Production of MnP, LiP and esterase

The production of MnP, LiP and esterase by *A. calcoaceticus* in PBM are shown in Figure 1. The ability of *A. calcoaceticus* to produce these extracellular enzymes shows its PE biodegradative potentials because these enzymes have been revealed to play pivotal roles during bio-transformation of PE (Zuriash *et al.*, 2023). In Figure 1, MnP activity was optimum on the tenth day (7.92 U/mg) in PBM. Manganese peroxidase production enhanced the ability of *A. calcoaceticus* to bio-fragment polyethylene polymer into smaller subunits.

The monomers are then ingested into the microbial cells, and utilized as carbon source for the production of energy, water, and carbon dioxide (Sowmya *et al.*, 2020). Similarly, lignin peroxidase (LiP) activity was optimum on the tenth day (8.26 U/mg). *A. calcoaceticus* also produced LiP activity because of its unique ability to work in the presence of various mediators enhancing its contribution to the degradation process of complex polymers like polyethylene (Evans *et al.*, 2015). In addition, esterase activity was produced as well by *A. calcoaceticus* on the tenth day and 5.70 U/mg specific

activity was observed. Esterase was produced by the bacterium to hydrolyze ester bonds present in PE, breaking down polyethylene into smaller monomers and thereby facilitating the overall biodegradation process.

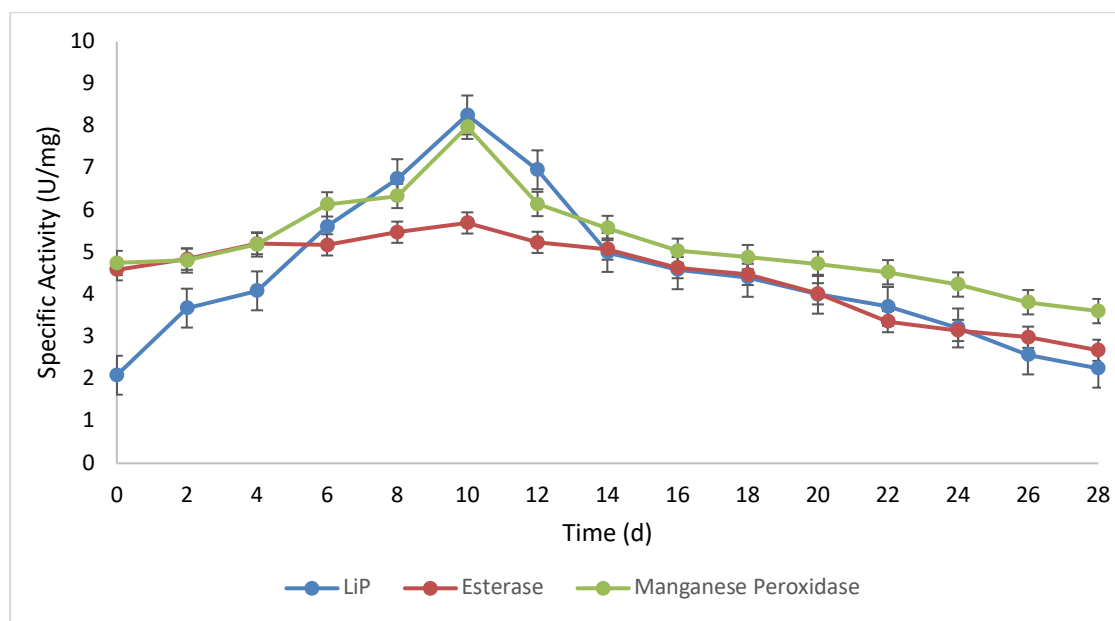


Figure 1. (a) Production of manganese peroxidase, lignin peroxidase and esterase from *A. calcoaceticus* (error bars represent mean \pm standard deviation)

3.2 Effect of pH on activity and stability of MnP, LiP and esterase

The pH profile of MnP, LiP and esterase produced from *A. calcoaceticus* exhibiting wide range of pH activities (between pH 3 to pH 11) is presented in Figures 2a-f. MnP showed 62 % relative activity at pH 3 which increased to 83 % at pH 5 (Figure 2a). Optimum MnP activity was recorded at pH 7 and decline in activity was seen afterwards as 40 % relative activity was finally obtained at pH 11. MnP activity was optimum at neutral pH because the amino acid residues at the active site were functional and stable at neutral pH which invariably enhanced effective PE binding and catalysis (Verónica *et al.*, 2021). Additionally, MnP was optimally stable at pH 7 and 81% residual activity was recorded after incubation for 3 h (Figure 2d). Optimum stability was also seen at pH 7.0 because neutral pH produces extra hydrogen bonds and salt bridges which helps to stabilize the heme pocket of MnP (Verónica *et al.*, 2021). Manganese peroxidase produced by *Penicillium italicum* with optimum activity and stability at pH 7.0 was also reported as an effective bio-degrader of polyethylene (Ogunjemite *et al.*, 2023).

In Figure 2b, lignin peroxidase (LiP) activity was optimum at pH 5 after which there was decrease in LiP relative activity from 77.8 % to 38.9 % in pH 7.0 and pH 11.0. LiP activity was optimum at pH 5 because acidic pH facilitates the protonation of PE, making it more reactive for LiP catalysis (Pham *et al.*, 2021). Similarly, optimum LiP stability was displayed at pH 5.0 and 80 % residual activity was obtained (Figure 2e). Seventy percent (70 %) and 49 % residual activities were observed at pH 7.0, and pH 11.0 respectively. LiP was optimally stable at pH 5.0 because the acidic pH helps to retain LiP structural integrity and maintains its catalytic activity (Fernández-Fueyo *et al.*, 2014).

Esterase was depicted with an increasing relative activity from pH 3.0 until (100 %) optimum activity was attained at pH 9.0 in Figure 2c. Esterase activity was optimum at pH 9.0 because alkaline conditions improves substrates ionization, reaction and hydrolysis of ester bonds present in the substrate (Rodríguez-Mejía *et al.*, 2024). Esterase retained 81 % activity at pH 7.0 and exhibited

optimum stability at pH 9.0 with 83.5 % residual activity and 40 % residual activity was recorded at pH 11.0 (Figure 2f). Esterase was optimally stable at pH 9.0 as well because this alkaline pH helps to maintain the enzyme's activity over a longer period, making it more efficient at pH 9.0 (Yi *et al.*, 2021).

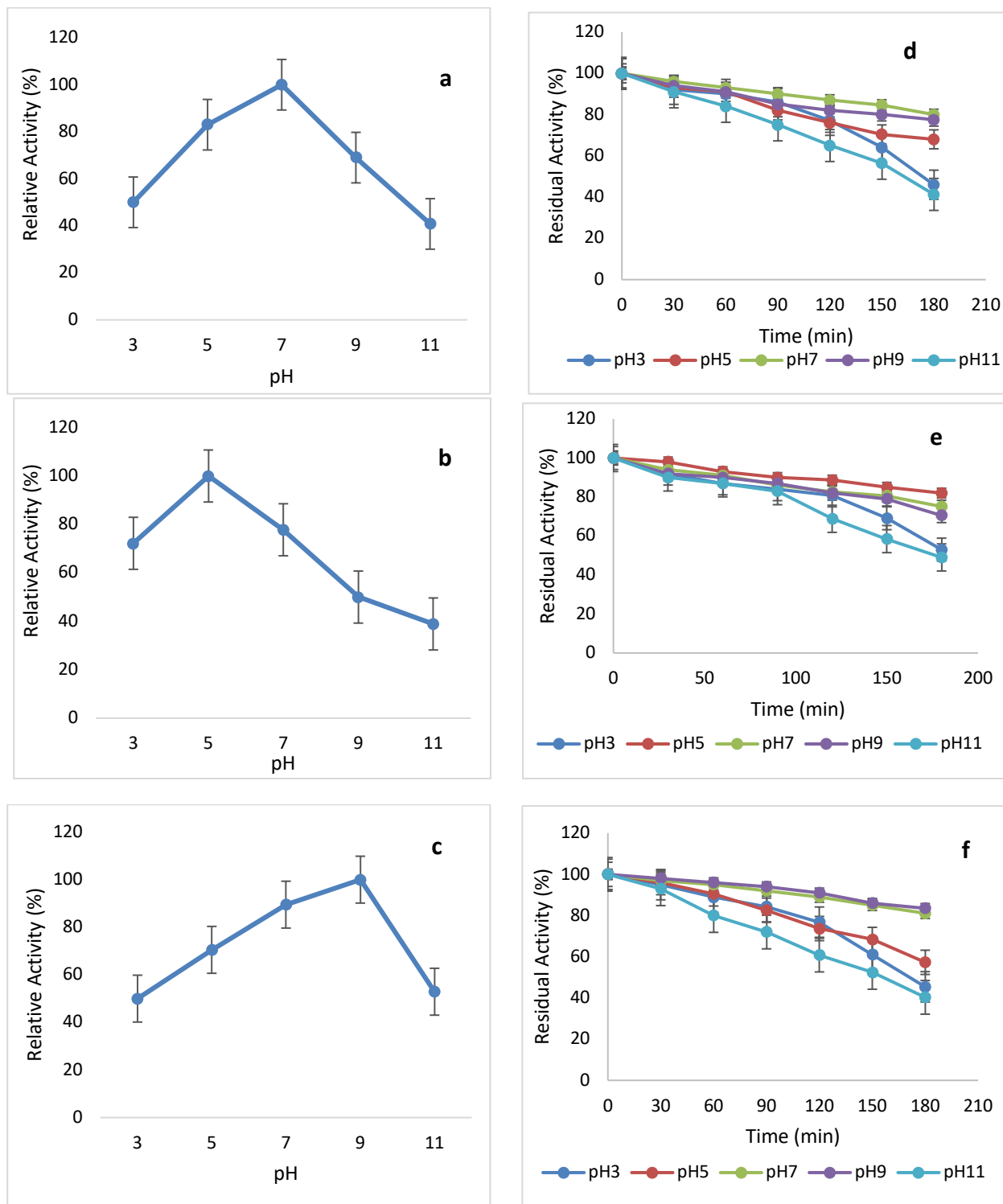


Figure 2. Effect of pH on (a) MnP activity, (b) LiP activity, (c) esterase activity (d) MnP stability, (e) LiP stability and (f) esterase stability from *Acinetobacter calcoaceticus* (Error bars represent Mean \pm standard deviation)

3.3 Effect of temperature on activity and stability of MnP, LiP and esterase

The optimum temperature for the activities and stabilities of MnP, LiP and esterase produced from *A. calcoaceticus* is illustrated in Figures 3a-f. MnP activity was peak at 60 °C and 44 % relative activity was recorded at 80 °C (Figure 3a). MnP activity was optimum at 60 °C because high temperatures increase the kinetic energy of molecules, leading to more frequent and effective collisions between MnP and the substrate (Plácido and Capareda, (2020). The activity of manganese peroxidase obtained from *Trametes spp* was also optimum at 60 °C (Huy *et al.*, 2017). Likewise, after incubation of the enzyme for 3 hours at different temperatures, MnP activity was optimally stable at 60 °C retaining approximately 70 % of its activity while it showed 42 % residual activity at 80 °C (Figure 3d). The enzyme was stable at 60°C due to the rigid protein structures which helps to maintain the functional shape and activity at elevated temperatures (Plácido and Capareda, (2020).

In Figure 3b optimum LiP activity was also obtained at 60 °C. The relative activity of LiP reduced from 78 % to about 55% at 70 °C and 80 °C. Similarly, in Figure 3e optimum LiP stability was achieved at 60 °C and 50 % residual activity was seen at 80 °C. LiP was stable at 60 °C because it has ability to structurally adapted and withstand high temperatures without denaturing. Furthermore, Figure 3 (c) reveals esterase was optimally active at 50 °C. Eighty-seven percent (87 %), 73 % and 50 % of relative activities were recorded at 60 °C, 70 °C and 80 °C. Esterase was also optimally stable at 50 °C and 50 % residual activity was displayed at 80 °C (Figure 3f). Esterase was active and stable at 50 °C because the temperature facilitated the solubility of the substrate, making it more accessible to the enzyme and enhanced the enzymatic reaction (Barman and Dkhar, 2022).

3.4 Effect of metal ions and EDTA on activity of MnP, LiP and esterase

The reactions of MnP, LiP, and esterase to different metallic chlorides (Cu^{2+} , Ca^{2+} , Mg^{2+} , Mn^{2+} , Ba^{2+} , and Hg^{2+}) are illustrated in Figures 4a-c. Manganese peroxidase (MnP) activity was only increased in the presence of 5 mM and 10 mM Mn^{2+} (123 % and 150 % respectively) (Figure 4a). The significant increase in MnP activity when manganese ion was added shows Mn^{2+} as an activator. MnP specifically requires manganese (Mn) ions to function effectively because the active site is designed to bind Mn ions, facilitating the oxidation of Mn (II) to Mn (III) (Zhang *et al.*, 2022). Previous report revealed Mn^{2+} as an activator of MnP (Yan *et al.*, 2022). Reduction in MnP activity was observed in the presence of Cu^{2+} , Ca^{2+} , Mg^{2+} , Ba^{2+} , and Hg^{2+} as 29 %, 63 %, 43 %, 55 %, and 51 % relative activities were recorded respectively. Reduction in MnP activity was recorded because they do not fit into the active site of MnP and metal ions, like Hg and Cu, can inhibit MnP activity (Saroj *et al.*, 2013). EDTA inhibited the activity of MnP. EDTA inhibition is caused by the metal chelating characteristic of EDTA which makes metal ions that are co-factors for the activity of manganese peroxidase unavailable to the enzymes.

Inhibition of MnP, by EDTA has been reported in previous studies (Olajuyigbe *et al.*, 2016, Zhang *et al.*, 2022). Figure 4b shows LiP activity was enhanced by 10 mM Ca^{2+} (113 %), and Mg^{2+} (122 %). Calcium ion and Mg^{2+} activated LiP because they are essential for the stability LiP, however, it may not be involved in the catalytic process of the enzyme significant increase in LiP activity in the presence of Cu^{2+} (43 %), Mn^{2+} (57 %), Ba^{2+} (72 %) and Hg^{2+} (69 %) because LiP typically requires hydrogen peroxide (H_2O_2) as a co-substrate for its catalytic activity and do not rely on metallic chlorides for activation (Pham *et al.*, 2021). Ditto, Hg and Cu, can inhibit LiP activity by binding to the enzyme and disrupting its structure or interfering with its active site. EDTA also inhibited the activity of LiP as 23 % relative activity was seen. Furthermore, Figure 4 (c) reveals esterase activity was elevated by 10 mM Ca^{2+} (125 %), Mg^{2+} (118 %) and Ba^{2+} (110 %). These metal ions enhanced esterase activity

because they help to stabilize the three-dimensional structure of esterase, ensuring that the enzyme maintains its active conformation which is crucial for catalytic activity (Rodríguez-Mejía, *et al.*, 2024).

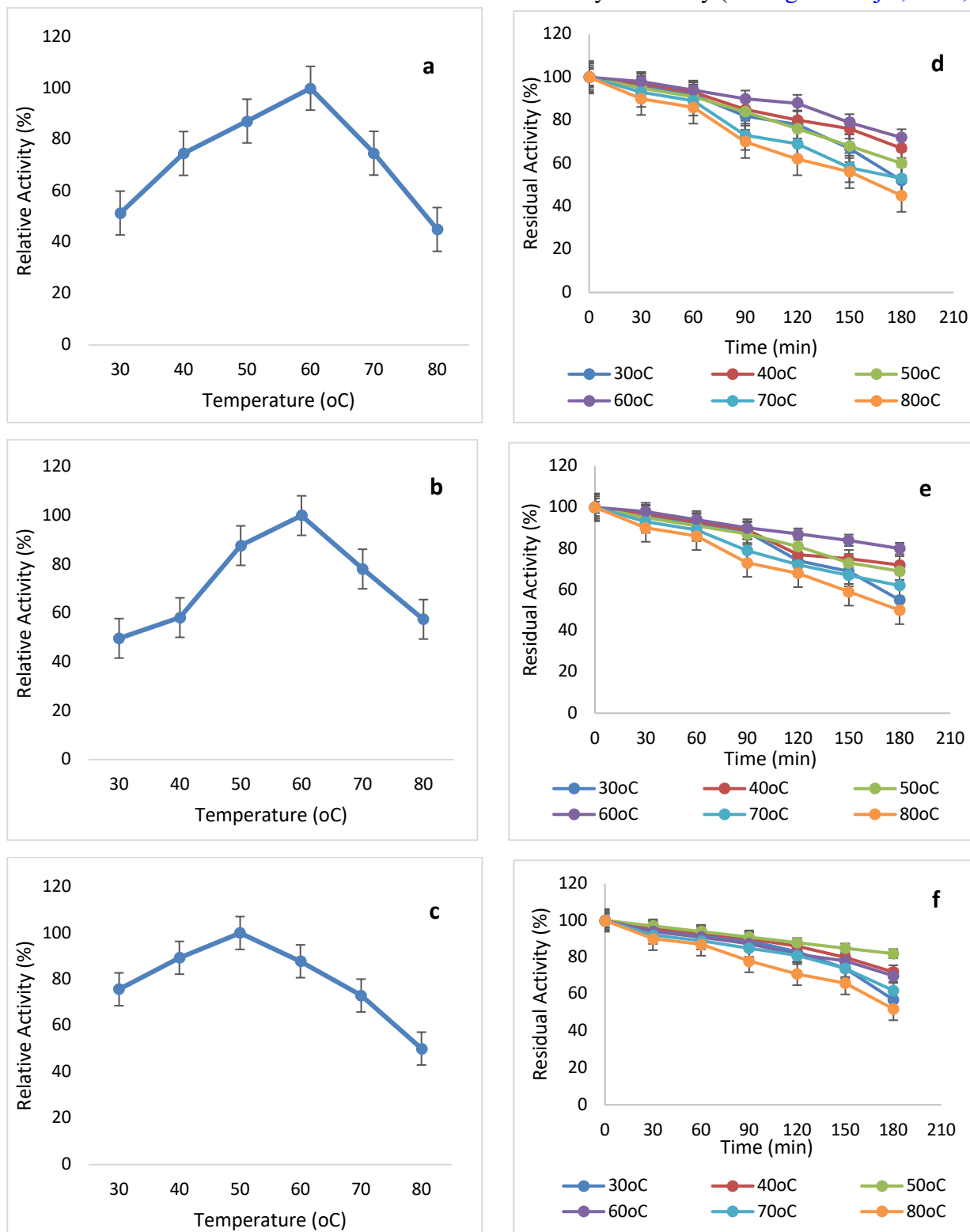


Figure 3. Effect of temperature on (a) MnP activity, (b) LiP activity, (c) esterase activity (d) MnP stability, (e) LiP stability and (f) esterase stability from *Acinetobacter calcoaceticus* (Error bars represent Mean \pm standard deviation)

There was no significant increase in esterase activity in the presence of Cu^{2+} (50 %), Mn^{2+} (68 %), and Hg^{2+} (33 %) because they bind to the active site of the enzyme, preventing the substrate from

binding and thus inhibiting the enzyme's activity (Rodríguez-Mejía *et al.*, 2024). EDTA also chelated the activity of esterase and 28 % relative activity was recorded.

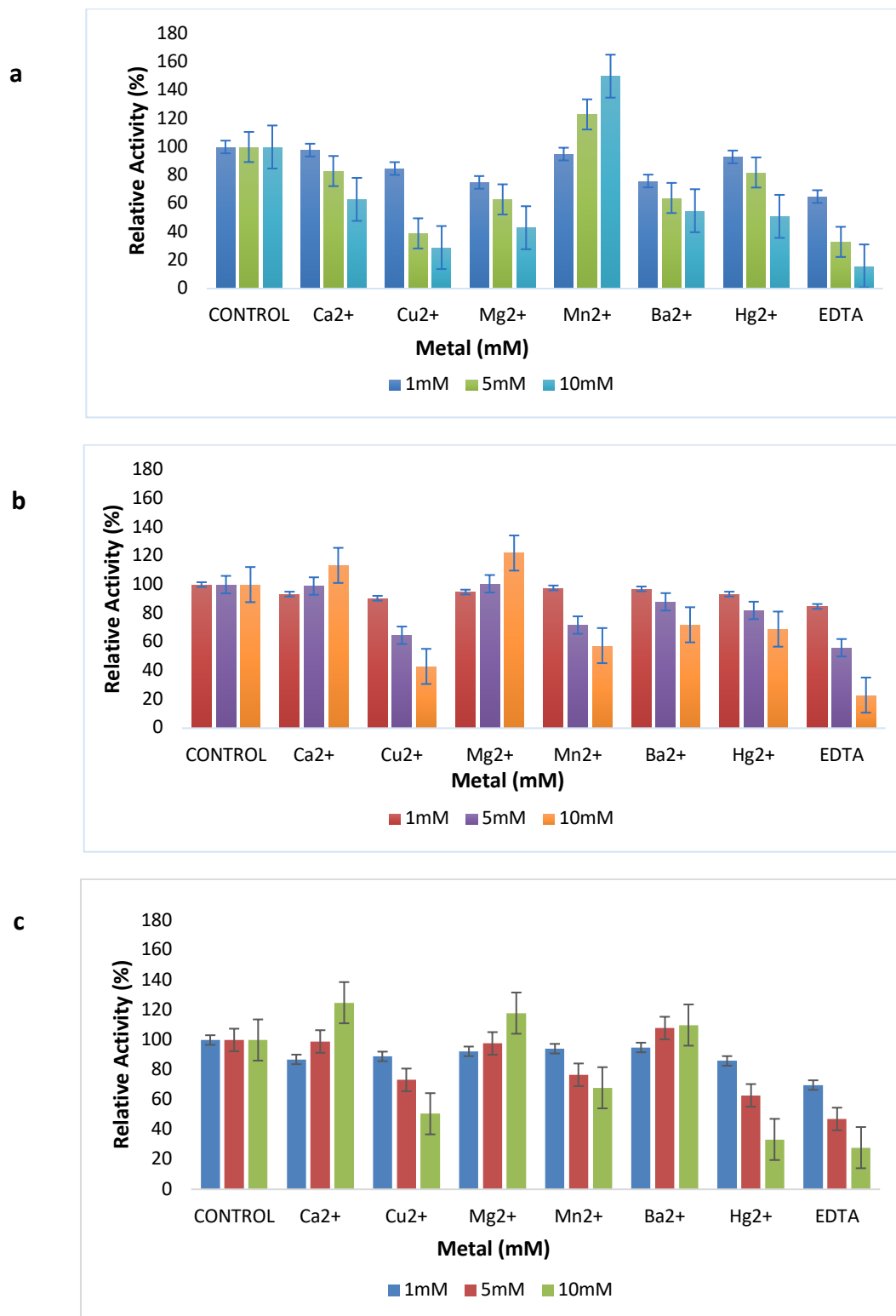


Figure 4. Effect of metal ions on (a) MnP activity, (b) LiP activity, and (c) esterase activity from *Acinetobacter calcoaceticus* (Error bars represent Mean \pm standard deviation)

3.5 Assessment of PE biodegradation potential of *A. calcoaceticus*

The fourier transform infrared spectrophotometer spectra of the PE sheet treated with *Acinetobacter calcoaceticus* is presented in Figure 5. The FTIR spectra of the degraded PE sheet showed shift to the left in existing peaks at wavelength 2017 to 2019 nm which corresponds to C=O esters, and at 2850 nm which signals the presence of C-H alkanes when compared with the un-degraded PE. Similarly, there were shiftings to the right in the biodegraded PE peaks at 873 nm to 773 nm corresponding to C-C vibrations, 1062 nm to 966 nm, signaling alky substituted ether group. At 1313 nm there was a shift to 1303 nm showing the presence of O-H plane bend. In addition, there was a shift from 1541 nm to 1471 nm which depicts to secondary amide (NH) bend, and 1377 nm shifted to 1367 nm showing an symmetric methyl (-CH₃) bend compared to the un-degraded PE. Furthermore, new peaks were observed at 2918 nm showing the C-C aromatic group, and the presence of hydrogen-bonded O-H carboxylic acid was displayed at 3431 nm as seen in Figure 5b.

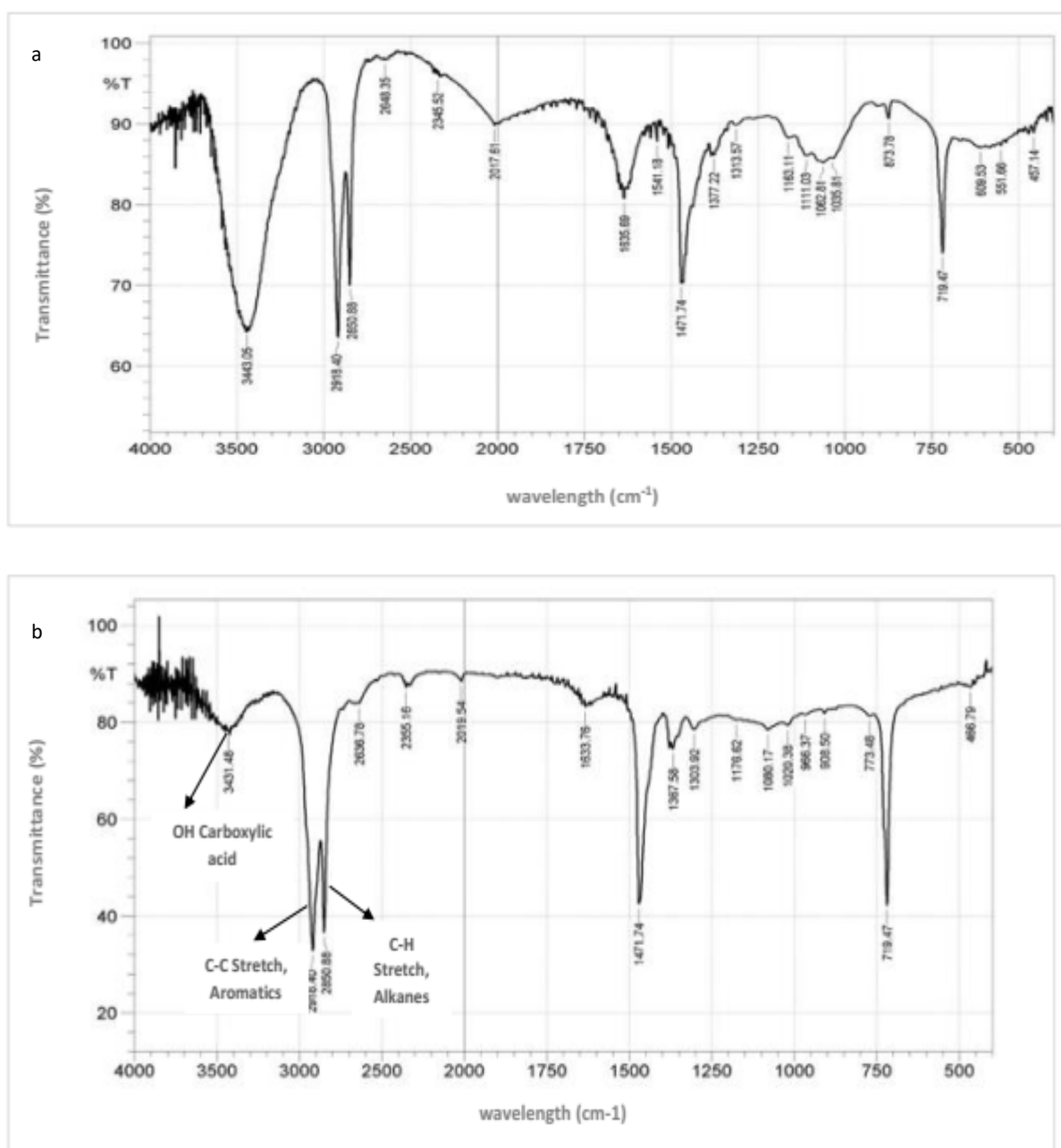


Figure 5. FTIR spectra of (a) control PE sheet and (b) PE sheet treated with *A. calcoaceticus*

The FTIR spectra results revealed that *Acinetobacter calcoaceticus* degraded PE as shown by formation of new functional groups in PE such as aromatic OH group and carboxylic group (-OH, -COOH) which are products of the hydrolysis and oxidation of PE (Peng *et al.*, 2020), (Li *et al.*, 2020). A study by also revealed that the shifts in C-H stretch during degradation indicate the loss of di-acid and di-alcohol groups from the polymer, suggesting the hydrolysis of PE (Spinal *et al.*, 2021). However, this result is in contrast to the report that concluded that microorganisms only degrade chemically or physically pre-treated polyethylene (Vimala and Mathew, (2016).

Conclusion

In this study, *Acinetobacter calcoaceticus* produced thermostable manganese peroxidase (MnP), lignin peroxidase (LiP), and esterase activities. MnP activity was optimum and stable at 60 °C, also at 60 °C LiP activity was optimally stable, while esterase activity was optimally stable at 50 °C. The enzymes showed activities over a broad range of pH and at pH 7.0, pH 5.0, and pH 9.0 MnP, LiP and esterase were optimally active and stable. The activities of LiP and esterase were inhibited by Cu²⁺, and Hg²⁺ and enhanced by Mg²⁺ and Ca²⁺ while MnP activity was only elevated by Mn²⁺. However, EDTA chelated the activities of all the enzymes. The FTIR spectra of the degraded PE sheet showed shiftings and formation of new peaks at different wavelengths most especially the formation of aromatic C-C functional group, and the OH carboxylic acid at 2918 nm, and 3431 nm when compared with control PE sheet. The results reveal the ability of *Acinetobacter calcoaceticus* to produce thermostable manganese peroxidase (MnP), lignin peroxidase (LiP), and esterase with activities over a broad pH range which are essential for effective biodegradation of PE. In addition, the formation of aromatic C-C functional group, and the OH carboxylic acid displayed in the FTIR spectra of the degraded PE sheet confirms the biodegradation activity and potential of *Acinetobacter calcoaceticus* on PE. Hence, we recommend industrial and domestic exploration of *Acinetobacter calcoaceticus* for effective biodegradation of different environmental pollutants most especially polyethylene.

Disclosure statement: *Conflict of Interest:* The authors declare that there are no conflicts of interest.

Compliance with Ethical Standards: This article does not contain any studies involving human or animal subjects.

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