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Bioremediation of heavy metals by melanised and non-melanised feathers and heavy metal resistant feather-degrading bacteria

Biorremediación de metales pesados por plumas melanizadas y no melanizadas y bacterias degradantes de plumas, resistentes a metales pesados

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Abstract

Heavy metals are toxic and detrimental water pollutants. The continuous discharge of cadmium (Cd) and lead (Pb) containing effluents from industries within and around residential areas in Nigeria is worrisome since they do not only affect human beings, but also beneficial microbes, animals and vegetation due to their mobility in aqueous ecosystem, toxicity and non-biodegradability. The aim of this study is to compare the ability of black (melanised) and white (non melanised) feathers to adsorb Cd and Pb in two aqueous media and to isolate Cd and Pb tolerant feather-degrading bacteria that will degrade the Cd and Pb polluted feather generated. Black and white chicken feathers were used as bio-adsorbent material in two Cd and Pb containing aqueous solutions (distilled water (DW) and feather meal broth (FMB). The sorption capacity of the feathers was tested by atomic absorption spectrometry and gravitational methods. Cd and Pb tolerant bacterium identified as *Bacillus* sp. was isolated from manure of local chicken waste and was used to degrade Cd and Pb polluted feathers. Result shows that black feathers possessed higher uptake capacity of Cd and Pb in FMB than white feathers, as about 15 and 30 ppm of Pb and Cd were adsorbed in FMB in 1hr from initial concentrations of 25 and 50 ppm respectively, slightly higher than 13 and 23 ppm adsorbed by white feathers. A Cd and Pb resistant feather-degrading bacteria isolated from chicken manure were able to degrade about 40% of Cd polluted feathers and 30% of Pb polluted white feathers in 7 days. Even though the bacterium grew faster in FMB containing Cd polluted feathers, the degradation of Cd polluted white feathers was faster than black. Hydrolysates produced after complete degradation of polluted feathers contained low concentration of heavy metals. Bioremediation of heavy metals and recalcitrant chicken feather wastes can be achieved by melanised and non melanised feathers and metal resistant feather-degrading bacteria (FDB).

Keywords: Feathers, melanised, cadmium, lead, heavy metals, bioremediation, biodegradation.

Resumen

Los metales pesados son contaminantes tóxicos y perjudiciales para el agua. La continua descarga de efluentes que contienen cadmio (Cd) y plomo (Pb) por parte de las industrias dentro y alrededor de las áreas residenciales en Nigeria es preocupante, ya que no solo afectan a los seres humanos, sino que también a los microbios, animales y la vegetación debido a su toxicidad, no biodegradabilidad y movilidad en ecosistemas acuosos. El objetivo de este estudio es comparar la capacidad de las plumas negras (melanizadas) y blancas (no melanizadas) para adsorber Cd y Pb en dos medios acuosos, y aislar bacterias tolerantes a Cd y Pb que degradarán las plumas contaminadas con estos metales. Se usaron plumas negras y blancas de gallina como material bioadsorbente en dos soluciones acuosas que contenían Cd y Pb (agua destilada (DW) y caldo de harina de plumas (FMB)). La capacidad de sorción de las plumas fue testeada por espectrometría de absorción atómica y métodos gravitacionales. La bacteria tolerante a Cd y Pb identificada como *Bacillus* sp. se aisló de desecho de estiércol de gallina local y fue usada para degradar las plumas contaminadas con Cd y Pb. Los resultados muestran que las plumas negras poseen mayor capacidad de adsorción de Cd y Pb en FMB que las plumas blancas; en 1 h desde una concentración inicial de 25 y 50 ppm de Pb y Cd, se adsorbieron 15 y 30 ppm respectivamente, siendo ligeramente superior a los 13 y 23 ppm adsorbidos por las plumas blancas. La bacteria aislada de estiércol de gallina resistente a Cd y Pb degradadora de plumas pudo degradar aproximadamente el 40% de plumas contaminadas con Cd y el 30% de plumas blancas contaminadas con Pb en 7 días. Pese a que la bacteria creció más rápido en FMB que contenía plumas contaminadas con Cd, la degradación de las plumas blancas contaminadas con Cd fue más rápida que las negras. Los hidrolizados producidos después de la completa degradación de las plumas contaminadas contenían baja concentración de metales pesados. La biorremediación de metales pesados y desechos de plumas recalcitrantes puede lograrse mediante plumas melanizadas y no melanizadas y bacterias resistentes a metales degradantes de plumas (FDB).

Palabras clave: Plumas, melanizado, cadmio, plomo, metales pesados, biorremediación, biodegradación.

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1 Introduction

In recent years, pollution has become one of the most serious environmental problems with heavy metals as among the major inorganic pollutants that contaminate various ecosystems including water and soil (Buendía-González *et al*., 2019; Karamba *et al*., 2020; Zakaria *et al*., 2020; Ibrahim *et al*., 2020). Heavy metals that have been reported to be toxic metals are disseminated into the environments such as surface water bodies (e.g. lakes, ponds, and reservoirs), ground water, soil and plants through natural and artificial activities (Cabrera *et al*., 2006; Muñoz *et al*., 2012; Padrilah *et al*., 2017; Cruz *et al*., 2018).

The problem associated with the existence of heavy metal pollutant in our environment is their recalcitrant nature, which makes them hard to be degraded by either microbes or their enzyme that accumulates in living tissues, leading to various disease and disorders (Guo *et al*., 2010; Ahmad *et al*., 2016; Sabullah *et al*., 2017; Alcázar-Medina *et al*., 2019; Tengku-Mazuki *et al*., 2020; Zahri *et al*., 2020). In Nigeria, the location of industries within the vicinity of residential areas and un-control discharge of untreated and semi treated effluents into sand and plastic blocked community sewers has made it possible for the presence of different heavy metals in the environment. Some of the commonest heavy metal pollutants reported in different environmental and agricultural samples include copper (Cu), arsenic (As), chromium (Cr), lead (Pb) nickel (Ni), mercury (Hg) and zinc (Zn) (Monachese *et al*., 2012; Chaudhari *et al*., 2013; Yusuf *et al*., 2015; Fadzil *et al*., 2019; Hernandez-Botello *et al*., 2020). As explosion in population continue to force people to reside in locations where plastic, paint and tannery industries are generating a lot of Cd and Pb as effluents, possibility of Cd and Pb toxicity is imminent.

The conventional means of removal of toxic heavy metals from different sources are available but are too expensive in terms of set up and maintenance especially in developing countries (Malik, 2004; Pires *et al*., 2011). Thus, alternative, less expensive and environmental friendly method becomes necessary.

Similarly, poultry processing industries in Nigeria also have enormous environmental, economic and health implications. The waste generated from poultry production processes are mainly feathers which in addition to causing environmental pollution, also serve as reservoirs for many bacteria. Effective disposal of such feather wastes is a major environmental problem in Nigeria, since significant amount of them finally settled in ditches, culverts and major water channels. Adsorption of heavy metals by different biological wastes including chicken feather has been reported (Al-Asheh *et al*., 2003; Pan dey *et al*., 2007; Wang *et al*., 2013; Yusuf *et al*., 2015; Ahmad *et al*., 2018). The content of feather which consists of about 91% keratin, 1.3% fat and 7.9% water, has make them potential bio-absorbents for the removal of heavy metals (Al-Asheh *et al*., 2003; Yusuf *et al*., 2015; Bhange *et al*., 2016; Bhari *et al*., 2018). However, comparison in terms of adsorption capacity of black and white chicken feathers due to differences in their composition has not been well documented.

This study therefore aims at studying the difference in the ability of black and white feathers to adsorb cadmium (Cd) and lead (Pb) and to isolate Cd and Pb tolerant feather degrading bacteria that have the potential to degrade the Cd and Pb polluted feather generated.

2 Materials and methods

2.1 *Chemicals*

Heavy metal stock solutions and other chemicals used in this study were of analytical grade and purchased from Sigma-Aldrich Corporation, (St Louis, USA) except where contrarily indicated.

2.2 *Samples collection and processing*

Black and white chicken feathers were collected from slaughterhouses located in three major markets in Kano, North west Nigeria. The collected feather samples were washed several times with detergent and distilled water followed by soaking them in a solution of distilled water, ethanol, methanol and chloroform for 12 h to remove the remnant of organic matters present in the feathers. Finally, feathers were washed again with distilled water, dried in an oven at 50°C and then cut into small sizes with scissors (Yusuf *et al*., 2015).

Samples of soil, sewage and chicken manure were also collected from different feather dump and chicken rearing sites in Kano, Nigeria. They were transported to the Microbiology laboratory, Bayero University Kano for isolation of feather-degrading bacteria (FDB).

2.3 Isolation of feather degrading bacteria (FDM)

Isolation of FDB was carried out on feather meal agar which contained (g/L): 1.0 feather, 0.5 NaCl, 0.7 K_2HPO_4,1.4 KH_2PO_4, 0.001 $MgSO_4 \cdot 6H_2O$ and 1.5 agar as previously described (Yusuf *et al*., 2016, 2019). Feather degradation studies using both black and white feathers were carried out in 100 mL feather meal broth (FMB) incubated at 30°C on rotatory shaker for 10 days. FMB contains 10.0 g/L of feather, 0.5 g/L of NaCl, 0.7 g/L of K_2HPO_4, 1.4 g/L of KH_2PO_4, and 0.001 g/L of $MgSO_4 \cdot 6H_2O$ pH 8.0 as described by Tork *et al*. (2010). The residual feather at the end of the incubation period were filtered by pre-weight filter paper, rinsed with distilled water, dried in an oven at 60°C to obtain a constant weight. Percentage of feather degradation was calculated as described by Yusuf *et al*. (2016) using the formula;

$$\text{Percentage feather degradation} = \frac{(X + Zx)}{(Y + Zy)} \times 100 \tag{1}$$

where X=final weight of feather, Zx= final weight of filter paper, Y=initial weight of feather and Zy=final weight of feather.

2.4 Screening of FDB for Cd and Pb tolerance

Stock solutions (1000 ppm) of Cd and Pb were prepared and appropriate aliquot was pippeted and placed into 100ml of FMB to give a final concentration of 1 and 5 ppm. Five percent of standardized inocula of the isolated FDB were then inoculated into 100 ml of FMB containing Cd and Pb polluted feathers. Other sets of FMB without heavy metals in triplicates were used as control. FDB that grew and degraded feather faster in the presence of Cd and Pb at 1 ppm or above was selected and inoculated onto fresh FMA (Yusuf *et al*., 2016). The identity of the selected bacterium was detected by studying its morphological and biochemical characteristics as described elsewhere (Yusuf *et al*., 2019).

2.5 Generation of Cd and Pb polluted feathers

A 25 and 50 ppm concentration of Cd and Pb solutions was prepared from their respective 1000 ppm stock solutions by diluting an appropriate volume of the stock in 100 ml of FMB and DW followed by measurement of their respective pH (Yusuf *et al*., 2019). Five grams of black and white feathers were placed in appropriately labelled 250 ml capacity flasks containing 100 ml of FMB or DW and placed on orbital shaker at 150 rpm for 1 h. The feathers were then harvested from the respective flasks, dried in an oven at 50°C for 30 min, and then stored in a refrigerator until further use. The analysis of residual Cd and Pb left in FMB and DW flasks were carried out using atomic absorption spectrometer (model AA-6880, Shimadzu Japan) equipped with a dual atomizer system, a metal furnace, a high-performance boosted hollow cathode lamp, and a data processing module following nitric acid digestion. The operating parameters for each metal was set as recommended by the manufacturer [wavelength (nm) =228.8 for Cd and 217 for Pb; slit width (nm) =1.2 for both Cd and Pb; lamp current (mA) =2 for Cd and 3 for Pb].

Further, using gravimetric method, the final weight of the feather was determined on weight balance to quantify the amount of heavy metals adsorbed using the formula; $Xf - Xi$. Where Xf is the final dry feather weight obtained after adsorption and Xi is the initial weight of the dry feather before adsorption (Yusuf *et al*., 2019). A setup of FMB and DW without the heavy metals which were also placed on orbital shaker at 150 rpm for 1 h was used as controls.

2.6 Biodegradation of Cd and Pb polluted feathers by heavy metal tolerant feather-degrading bacterium

Ten grams of black and white feathers polluted with Cd and Pb were used as substrate in 100 ml FMB and were inoculated with 5% of standardized inoculate of the FDB. The flasks were then incubated at 30°C on a rotatory shaker at 150rpm for 10 d. Percentage of feather degradation was calculated at the end of incubation period as previously described. Flasks which contain Cd and Pb polluted feathers but without FDB served as control. Residual Cd and Pb in the hydrolysates produced after incubation was determined by atomic absorption spectrometry. The bacterial growth in each flask was estimated by colony forming units techniques in duplicates as previously described (Yusuf *et al*., 2019).

2.7 Data analysis

Data analysis was performed using Minitab (version 16) software. All experiments were run in triplicates except where contrarily indicated. The means and standard deviations (SD) were then determined from the triplicate (or duplicates where applicable) samples at each time to check for errors and variation among the samples, and the values are represented as the means ± SD. Differences in adsorption and degradation rate between black and white feathers were analyzed by Student's t test at 95% confidence level.

3 Results and discussion

3.1 *Adsorption of Cd and Pb by melanised and non-melanised feathers*

In this study, adsorption capacity of black and white feathers was compared in two aqueous solutions. Black feathers adsorb Cd and Pb better than white feathers in DW and FMB at 25 ppm concentration. About 50% mean reduction in the initial amount of Cd present in FMB and DW was observed when both feather types were used for the adsorption (Table 1). However, black feathers resulted in 41.6% and 39.2% reduction in the concentration of Pb and Cd respectively when initial concentration was 50ppm against 51% and 56% when white feathers were used. Even though the reason for the disparity is not yet understood, it may be due to the differences in presence of melanin and amount of amino acids present in melanised and non-melanised chicken feathers (Yusuf *et al*., 2019). It is worthy of note that the final initial pH of the FMB and DW were about 8.5±3.6 and 7.8±6.3 respectively before adsorption experiment. This could be the reason why adsorption of the heavy metals (Cd and Pb) by both black and white feathers took place slightly higher in FMB than in DW. This is in line with previous studies, where adsorption of different heavy metals was reported to be pH dependent (Yusuf *et al*., 2019) and was higher when pH shifted toward slight alkaline region. Similar trend was observed when the net weight of the feathers used to adsorb Cd and Pb was determined after 1h of shaking (Table 2). The net gain in feather weight could have resulted from heavy metal adsorption by the feathers. The possibility of adsorbing other metals present in FMB by the feathers was rule-out when the net weight of Pb and Cd polluted feathers were slight but significantly higher in distilled water than in FMB.

3.2 *Isolation and characterization of Cd and Pb resistant feather degrading bacteria*

A total of 7 FDB were isolated from soil and chicken manure samples collected from five different locations (L1, L2, L4, L5 and L6). Extent of degradation varies from one bacterium to another. Among the seven bacteria isolated, 3 degraded only white feathers effectively, while 2 (from L2 and L5) were able to degrade white and black feather at almost an equal rate. The highest degradation of black feathers was observed with bacteria isolated from L2 and L6. Until now, only few bacteria have been reported to degrade melanised feathers faster than non-melanised (Kumar *et al*., 2011; Okoroma *et al*., 2012). The presence of eumelanin in black feather has been indicated by scientist to be the reason why melanised feather are more recalcitrant to microbial degradation (Gunderson *et al*., 2008; Zduniak *et al*., 2014). Upon screening for heavy metal tolerance, one of the FDB (L6) isolated from chicken manure of local free range chickens was found to tolerate Cd and Pb, while one of the FDB isolated from soil collected from a dumpsite near a busy market was able to tolerate only Cd (Table 3). Metal tolerance and resistance have been reported in many bacteria systems and the rate of resistance is often high in bacteria isolated from heavily polluted environments than in clean environments (Lima de Silva *et al*., 2012). Bacteria isolated from manure of chicken that feed freely in the environment possess metal resistance system, particularly to Cd and Pb. Isolation of Cd and Pb tolerant FMB from L6 and not others could indicate that the tolerant bacteria are present in fecal samples of the chicken possibly from the long exposure to different contaminants due to the ability of the chicken to feed on contaminated environment. Previous studies have reported the presence of multi-drug resistant bacteria in the poultry droppings reared in Nigerian farms due to the excessive use of antibiotics in poultry production. The co-presence of antibiotic and heavy metal resistance genes on the same plasmid could be another reason why heavy metal resistant bacteria were recovered from the chicken (Hassen *et al*., 1998). The feather degrading Cd and Pb tolerant bacterium isolated in pure culture was identified based on cultural, morphological and biochemical characteristics as *Bacillus* sp., which is a Gram positive rod shape bacterium, is capable of completely degrading heavy metals free and polluted feathers completely in 7 and 10 days, respectively (Fig. 1).

Table 1. Residual concentration of Cd and Pb in FMB and DW after feather biosorption.

Heavy metals	**Initial conc. in FMB and DW (ppm)**	**Residual conc. of heavy metal after bioabsorption with feather (ppm)**			
		Black feather (10 g/L)		White feather (10 g/L)	
		FMB	DW	FMB	DW
Pb	26.2±0.4	11.3±0.6	16.6±3.1	13.3±3.2	14.2±1.4
Pb	50.9±0.8	21.1±1.8	31.2±2.6	26.2±0.6	28.4±3.2
Cd	25.4±0.2	9.6±2.6	11.4±1.0	11.6±1.6	14.1±1.2
Cd	50.2±0.1	19.7±0.6	24.7±0.7	28.2±1.0	26.5±1.5

Table 2. Residual weight of black and white feathers after use as Cd and Pb adsorbent in FMB and DW

Heavy metals	**Initial conc. in FMB and DW (ppm)**	**Residual weight of feather after bioabsorption (g)**			
		Black feather (10 g/L)		White feather (10 g/L)	
		FMB	DW	FMB	DW
Pb	24.2±0.4	13.6±0.6	12.4±5.1	12.1±3.0	10.8±2.6
Pb	52.1±0.8	16.2±1.8	15.7±1.2	14.3±6.1	13.8±1.6
Cd	26.7±0.2	17.3±4.4	17.6±2.6	15.5±2.4	14.3±3.2
Cd	49.4±0.1	16.5±2.8	14.4±1.4	15.8±0.6	16.2±4.6

Initial weight of feathers is 10g/L.

Table 3. Isolation of feather degrading bacteria and screening of the isolates for Cd and Pb tolerance.

Sample code/nature of sample	No of FDB isolated	No of FDB tolerant to Cd	No of FDB tolerant to Pb	No of FDB tolerant to Cd and Pb
L1 (soil)	2	1	0	0
L2 (soil)	1	0	0	0
L3 (sewage)	0	0	0	0
L4 (soil)	1	0	0	0
L5 (manure)	1	0	1	0
L6 (manure)	2	1	1	1

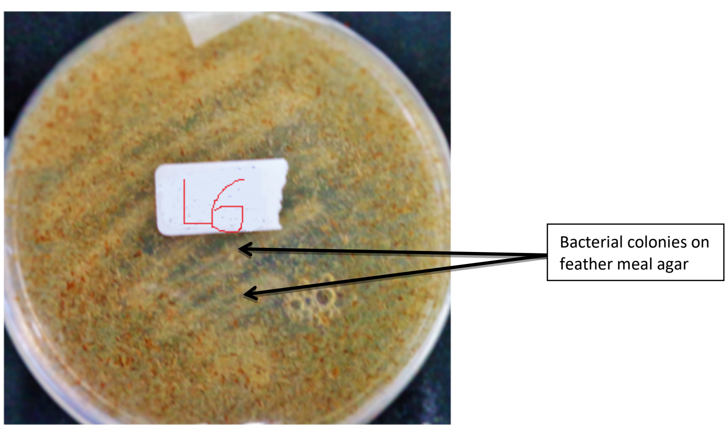

Fig. 1. Pure colonies of the selected feather-degrading bacterium on feather meal agar indicating its ability to utilise feather as a sole source of carbon and nitrogen.

Table 4. Mean growth (CFU/ml) of FDB in FMB.

Day	Mean growth (CFU/ml) of FDB in FMB with and without Pb and Cd							
	Black feather				White feather			
	Pb		Cd		Pb		Cd	
	Polluted	Non polluted	Polluted	Non polluted	Polluted	Non polluted	Polluted	Non polluted
1	$3.1\times10^6\pm2.1$	$3.9\times10^6\pm4.0$	$3.6\times10^6\pm0.4$	$2.9\times10^6\pm1.2$	$3.4\times10^6\pm2.8$	$3.2\times10^6\pm0.2$	$2.8\times10^6\pm2.1$	$4.1\times10^6\pm1.0$
2	$3.6\times10^6\pm0.5$	$3.7\times10^6\pm2.6$	$4.1\times10^8\pm0.8$	$3.3\times10^6\pm3.4$	$3.8\times10^6\pm0.6$	$3.4\times10^6\pm0.8$	$4.5\times10^7\pm0.6$	$2.9\times10^6\pm0.6$
3	$1.3\times10^7\pm2.0$	$2.1\times10^7\pm1.8$	$3.3\times10^8\pm1.6$	$4.0\times10^6\pm2.1$	$3.1\times10^7\pm1.8$	$3.7\times10^6\pm2.6$	$3.7\times10^8\pm2.0$	$3.2\times10^6\pm2.8$
4	$2.6\times10^8\pm1.1$	$4.3\times10^7\pm0.4$	$1.7\times10^8\pm4.4$	$2.8\times10^7\pm3.1$	$4.3\times10^7\pm2.0$	$4.4\times10^6\pm4.0$	$3.9\times10^8\pm1.0$	$2.3\times10^7\pm1.0$
5	$2.9\times10^8\pm0.6$	$2.9\times10^8\pm1.0$	$4.1\times10^7\pm2.0$	$3.6\times10^8\pm1.0$	$3.3\times10^8\pm3.2$	$2.8\times10^7\pm3.8$	$1.2\times10^8\pm5.6$	$3.6\times10^7\pm4.3$
6	$3.9\times10^7\pm0.8$	$2.8\times10^8\pm5.2$	$4.5\times10^6\pm1.4$	$5.1\times10^8\pm0.6$	$4.0\times10^8\pm1.2$	$4.6\times10^7\pm1.8$	$4.1\times10^7\pm3.6$	$4.3\times10^7\pm3.6$
7	$4.4\times10^7\pm4.1$	$3.3\times10^8\pm3.2$	$3.2\times10^6\pm7.2$	$2.1\times10^8\pm1.2$	$2.1\times10^8\pm3.8$	$3.2\times10^8\pm4.4$	$3.7\times10^7\pm4.4$	$2.2\times10^8\pm6.2$
8	$3.1\times10^7\pm2.7$	$2.9\times10^7\pm3.0$	$2.5\times10^6\pm5.6$	$3.5\times10^7\pm6.4$	$4.2\times10^7\pm1.0$	$2.5\times10^8\pm6.3$	$2.9\times10^6\pm2.0$	$3.1\times10^8\pm0.2$
9	$5.6\times10^6\pm2.2$	$3.8\times10^6\pm1.2$	3.5 x105±2.6	$2.9\times10^7\pm4.4$	$2.5\times10^7\pm0.5$	$3.7\times10^7\pm0.2$	$3.1\times10^6\pm0.8$	$1.9\times10^7\pm2.4$
10	$2.3\times10^6\pm1.8$	$4.1\times10^6\pm2.6$	2.2 x105±1.4	$3.9\times10^6\pm2.8$	$2.1\times10^7\pm0.2$	$2.3\times10^7\pm1.8$	$2.6\times10^6\pm1.2$	$2.1\times10^7\pm2.0$

Results are expressed as mean ± SD

Transmitting the hard to treat bacteria to human through direct contact with chicken or their products is possible. Previous study by Oves *et al*. (2013) has identified a *Bacillus* sp. isolated from contaminated soil in India as a good sorbent of Pb, Cd, Cr, Cu and Ni (Oves *et al*., 2013).

3.3 Biodegradation of Cd and Pb polluted feathers

Un-polluted white feathers suffered from higher degradation by the FDB than the un-polluted black feathers (Fig. 1). This is in agreement with previous studies which indicated white feathers as more easily degraded feather than melanised black feathers by FDB (Jaouadi *et al*., 2013; Bhange *et al*., 2016). Similarly, Cd polluted white feathers were significantly ($p<0.05$) degraded than Cd polluted black feathers under similar conditions. This could be as a result of higher sorption capacity of the melanised feathers over the non melanised feathers which exposes the FDB to a much higher concentration of Cd and Pb.

An unexpected phenomenon was observed in FMB containing Cd polluted feathers. The bacteria number significantly increased from 3.6×10^6 cfu/mL to 4.1×10^8 cfu/mL in 2 days, while it took the bacteria 4 and 5 days to reach the same range in FMB containing un-polluted and Pb polluted feathers respectively (Table 4). This relative fast growth in FMB containing polluted feathers may indicate growth efficiency of the bacteria in the presence of metals. However, the earlier growth of the FDB in FMB containing Cd polluted feathers does not translate into faster and efficient feather degradation as observed in FMB containing unpolluted feathers (Fig. 2).

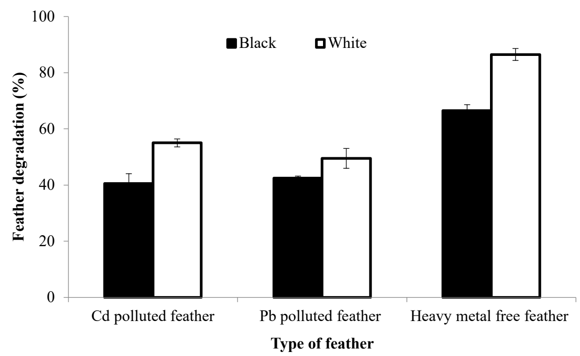

Fig. 2: Biodegradation of heavy metal polluted and unpolluted black and white feathers by *Bacillus* sp.

The resultant feather protein hydrolysate often produced after degradation of feathers by FDB have a lot of applications in making biofertilizers, animal feeds, antioxidants and exopolysacchrides due to high amounts of free amino acids it contains (Tork *et al*., 2010; Okoroma *et al*., 2012; Bhange *et al*., 2016). However, presence of heavy metals in such hydrolysates in high concentration may affect its application in for instance formulation of animal feeds due to toxicity of the metals. Interestingly, the amount of Cd and Pb recovered in the hydrolysates produced after complete degradation of polluted feathers in 10 days in this study were significantly lower than the amount present in the FMB in day 1 (Table 5). This suggests that the bacteria, in addition to possessing feather degrading property, also possess Cd and Pb accumulation property. In addition, the level of Pb in the hydrolysates in FMB containing Pb polluted white feathers was slightly higher than in FMB containing Pb polluted black feathers. This may suggest that the bacteria only tolerate Pb toxicity to certain limit and accumulation of Pb is not pronounced.

Table 5. Residual concentration of Cd and Pb in hydrolysates after complete degradation of 10 g/L of polluted feathers.

Type of feather in FMB	Residual concentration of heavy metal (ppm) after incubation	
	Day 1	Day 10
Cd polluted white feather	13.3±2.4	1.9±1.0
Cd polluted black feather	16.1±1.0	2.1±0.4
Pb polluted white Feather	11.7±0.5	4.6±1.1
Pb polluted black feather	13.2±0.9	3.3±3.2
Control (non polluted feather)	0.1 ±0.2	0.0±0.0

Conclusions

In conclusion, black chicken feather wastes possess higher capacity to adsorbed Cd and Pb in feather meal broth than white feathers which adsorb better in distilled water. The Cd and Pb polluted feathers were effectively degraded by a newly isolated heavy metal FDB belonging to genus *Bacillus*. The bacterium in addition to degradation of heavy metal polluted feathers also accumulate significant amount of the heavy metals (Cd and Pb) in the FMB, thereby producing feather hydrolysates potentially safe for further applications. Black feathers in addition to white feathers can be potentially used in bioremediation of Cd and Pb in aqueous solution. Biodegradation of polluted feathers to yield metal free hydrolysates by heavy metal resistant feather degrading bacteria will do well in managing environments polluted with heavy metals and recalcitrant chicken feathers.

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