Biosorption Using Tilapia (*Oreochromis* niloticus *Linn*) Scales in Chromium – Induced Wastewater

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Abstract: Biosorption is a process of using waste biomass to clean contaminated environment. This study was conducted to determine the capability of O. niloticus Linn fish scale as biosorbent for chromium contaminated aqueous solutions. Scales were washed and air dried before exposure to induced-wastewater that simulates high contamination in aqueous solutions. Initial concentration of synthetic wastewater was controlled and final concentration was sent to CRL Laboratory for measurement of total Chromium. Variations on contact time and weigh of dried scales were used to measure the biosorption capabilities of the fish scales where length of exposure used were 24, 48, and 96 hours and weight of dried scales were 1, 3 and 5 grams. Analysis of variance showed significant differences on both length of exposure and weight of biosorbent. Furthermore, the study showed a negative correlation on the chromium-induced wastewater respectively. Moreover, exposure of biomass to highly contaminated aqueous solution showed a drastic decline on chromium concentration with increasing weight of fishscales as well as longer contact times. Future study is highly recommended on the use of O. niloticus Linn and other fish scales on natural heavy metal contaminated freshwater ecosystem.

Keywords: Biosorption, fish scales, experimental, Zambales, Philippines.

1. INTRODUCTION

Rivers are one of the most vital sources of water an agriculture country may have. The continuous flow of water supply plants and carries the nutrients it need in order to grow and reproduce in a normal state. However, contamination of the river system may occur and alter the normal production of plants that will in due time affect higher level of organisms.

Water pollution is one of the prevalent issues affecting the provinces in our country that is relatively affected by irresponsible agricultural, domestic and mining activities. By definition, water pollution is any chemical, physical or biological change in the quality of water that has a harmful effect on living organisms that thrive and/or utilize such resource (Lenntech, 2015). As of 2013, 50 of the 421 rivers in the country are already considered "biologically dead" which means that the nutrients and oxygen levels are already depleted, thus, cannot support any organisms thriving on such ecosystem. One source of water contamination comes from heavy metals which includes arsenic, cadmium, chromium, copper, nickel, lead, and mercury (Aguilar, 2013).

Developed by the modern necessity and demand, heavy metals are on the top of the mining list and simply defined as a metal that is heavy in its atomic weight. Heavy metal is a member of loosely-defined subjects of elements that exhibit metallic properties which mainly includes the transition metals, some metalloids, lanthanides, and actinides. Many definitions have been proposed – some based on density, some on atomic number or atomic weight, and some on chemical properties or toxicity.

Chromium, for the past decades has been a primary source for manufacturing steel and other alloys and is a naturally occurring element in rocks, animals, plants, soil and volcanic dust and gasses. In addition, it is present predominantly in one of two valence states: Trivalent chromium (III) and Hexavalent chromium (VI), which, along with the less common metallic chromium (0), is most commonly produced by industrial processes. Chromium compounds, either Chromium

(III) or Chromium (VI) form are used for chrome plating, manufacturing of dyes and pigments, leather and wood preservation, and treatment of cooling water towers. Smaller amounts in addition, are used in drilling muds, textiles, and toner for copying machines.

Numerous ways have been proven to remediate or at least alleviate the issue of heavy metal contamination on the river systems in our country. Adsorptive removal of heavy metals from aqueous solutions has received much attention in recent years. Biosorption is a convenient and efficient way of removing heavy metal contaminants especially on liquid mediums and ecosystems. Furthermore, bioremediation consist of applications which involve detoxification of hazardous solutions by transferring the heavy metal ions to another medium by means of microbes or plants known as bioremediation and phytoremediation respectively. In addition, this also eliminates the need to transfer contaminated samples as this may cause cross-contamination (Gavrilescu, 2004). Plants and trees are often used for biosorption, however, organic materials such as fish scales are recently being used because of its similar ability to accumulate heavy metals through the process of adsorption high enough to conduct further study.

Biosorbents are prepared from naturally – abundant and/or waste biomass which are cost – effective but with high uptake capacity (Volesky and Holan, 1995). Moreover, biosorption is a physico-chemical adsorption process whereby metal ions become attached to the biomass surface. The ability to adsorb metals has been investigated for many materials, including wool, rice, straw, coconut husks, peat moss, fungi, algae and yeast (Prabu, 2014). However, little information has been develop regarding the ability of skin appendages of animals such as chicken plumage and fish scales.

Zambales province is a good source of aquatic freshwater fishes such as *Oreochromis niloticus* locally known as the tilapia fish. In addition, locals normally discard the scales of *O. niloticus* and other freshwater fish before consumption which makes and regards such scales of no use and waste to people but could be a good product for biosorption. Consequently, Zambales is one of the provinces with active mining economy to which several heavy metals are being excavated causing its release and probable source of unintentional contamination of the river system.

Therefore, the researcher aims to use such considered waste biomass as probable material for biosorption of heavy metals to remediate the freshwater ecosystem in our province. Thus, promoting increase in economy but safeguarding the environmental equilibrium. In connection with this, the researchers would like to assess the *O. niloticus* scales biosorption capabilities on induced wastewater to contribute in finding an eco – friendly remediating procedures in our province. Results of the study can then be utilized by the government to enhance other methods of remediating contaminated waters in our province.

A. Biosorption: Fish scales cleaning heavy metals:

Biosorption is defined as the ability of biological materials to accumulate heavy metals from wastewater through metabolically mediated or physico-chemical pathways of uptake (Ahalya et al., 2004). The biological materials used in the process are usually inexpensive dead biomass that are naturally abundant or waste biomass of algae, moss, fungi or bacteria (Kratochvil and Volesky, 1998). Advantages of biosorption are the significant amount of energy savings from a more efficient wastewater treatment system operating for fewer hours and it is economically attractive because waste biomass is inexpensive and widely available (Mustafiz et al., 2004). Biosorption also offers low operating cost, minimization of chemical and biological sludge, and no additional nutrient requirements (Kratochvil and Volesky 1998).

The removal of heavy metals from our environment especially wastewater is now shifting from the use of conventional adsorbents to the use of biosorbents. The presence of heavy metals in the environment is of major concern because of their toxicity, bioaccumulating tendency, and threat to human life and environment. In recent years, many low cost sorbents such as algae, fungi, bacteria and agricultural by-products have been investigated for their biosorption capacity towards toxicology of heavy metals and the biosorption capacity of biosorbents compared to conventional adsorbents (Igwe and Abia 2006).

In the study of Prabhu et al. (2012) entitled "*Biosorption studies of heavy metals from synthetic effluents*", the researchers used the scales of *Catlacatla* for the sorption of chromium (VI). The scales of South Asian freshwater fish of the carp family was used due to its abundance in their country. The removal of Cr (VI) from aqueous solutions of synthetic chromium, Nickel and Copper were investigated as a function of biosorbent concentration.

In addition to studies of biosorption of heavy metals using fish scales, Huang 2007, also observed biosorption capabilities using fish scales of tilapia under different biosorbent to copper mass ratios and contact times. With the use of a biosorbent

to copper mass ratio of 13.5, 33.988% of the copper was removed after two hours. After eight days, 97.2% of the heavy metal was removed with the application of biosorbent to copper ratio of 10.0. Larger amounts of biosorbent added and longer contact times yielded higher sorption percentage (Huang, 2007).

Aside from heavy metals, dyes and pigments represent one of the water pollution being emitted from various industrial branches, mainly from dye manufacturing and textile finishing and also from food colouring, cosmetics, papers and carpet industries. In a study of Huriye et al., adsorption of acid red 337 which is a dye on fish scales was investigated in a batch system. The adsorption is highly dependent on initial pH, temperature, initial dye concentration and adsorbent concentration (Huriye et al., 2013).

Usage of other biomaterials for biosorption was also conducted by Petersen et al. entitled *"Removal of Heavy metals from water by use of Biomaterials"*, induced process was used. Prepared aqueous solutions containing different heavy metals such as zinc, nickel, lead and copper was introduced to various biomaterials which consists of tobacco dust, saw dust, peat moss, sea weeds and fish scales.

Fish scales was also been utilized by Rodriguez, (no date), in the study entitled "Fish scales of Chanos chanos and Tilapia nilotica as natural sorption material in the recovery of Astaxanthin". In this study, the researcher constructed a Fish Scale Adsorption Apparatus (FSAA) containing unshredded and shredded scales where synthetic astaxanthin dissolved in water was made to flow through. Results showed that the compressed shredded Tilapia nilotica scales were most effective in adsorbing astaxanthin. The comparison of fish scales configurations considered the discoloration of filtrate, the total volume of filtrate collected, the time it took the first flow of filtrate to be released from the FSAA and the length of time the flow lasted. The occurrence of slits on the sclerits of the unshredded tilapia scales and the presence of debris like collagen materials on unshredded tilapia scales as revealed by the scanning electron micrographs were observed to retain astaxanthin better than milkfish scales by producing greater amount of clear, colorless filtrate and greater retention time before filtrate is released (Rodriguez, 2013).

There are two principal modes of adsorption of molecules on surfaces, namely physical adsorption (physisorption) and chemical adsorption (chemisorptions). The basis of distinction is the nature of the bonding between the molecule and the surface. Physical Adsorption, the only bonding is by weak Van der Waals – type forces. There is no significant redistribution of electron density either in the molecule or at the substrate surface. Chemical adsorption on the other hand is a chemical bond, involving substantial rearrangement of electron density, is formed between the adsorbate and substrate. The nature of this bond may lie anywhere between the extremes of virtually complete ionic or complete covalent character. There is a possibility that the molecules can decay (Aldrich et al., 2005).

B. Laws and Mandates Concerning Environmental Protection:

In lieu with the increasing problem regarding the environmental degradation in relation with the stationary agricultural productivity and performances of the different agricultural sectors, the government of the Philippines enacted several laws and decrees to address the problem in the Philippine environment and agricultural productivity.

The government of the Philippine enacted RA 7942 or known as the Philippine Mining Act of 1995 which allows and governs mining companies and local individuals in excavating raw minerals in our country. Moreover, RA 7942 Section 39 entitled Environmental Protection which emphasizes on environmental and enhancement program through rehabilitation, regeneration and reforestation of the area as well as the stabilization of the mined-out and tailings covered areas.

Republic Act 6969 know as Toxic Substances and hazardous and Nuclear Wastes Control Act of 1990 is a policy of the State to regulate, restrict or prohibit the use and disposal of chemical substances and mixtures that may pose threat and risk to health as well as to the environment (Robles, 2014).

Executive Order No. 79 otherwise known as Institutionalizing and Implementing Reforms in the Philippine Mining Sector Providing Policies and Guidelines to Ensure Environmental Protection and Responsible Mining in the Utilization of Mineral Resources Section 2 promulgates the full enforcement of environmental standards in mining reinforcing that mining companies and small scale miners shall include in their program plans and ways on rehabilitating the mining site to safeguard the health of the environment and its people.

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Republic Act 9275 known as *Philippine Clean Water Act* enacts the annual and semi-annual assessment, monitoring and preservation of the quality of Philippine waters, inclusive of the organisms within it, as well as save it from pollution through a sustainable plan.

Republic Act No. 7160 known as *Local Government Code of the Philippines of 1991* provides that LGUs have duties and authorities to protect and co-manage the environment and enhance the right of the people to balanced ecology (Robles, 2014).

With these laws, it can be established that it is the liability of the miners, the locals and the government to maintain or at least ensure that the environment will not be destroyed. Rehabilitating mining sites through research should also be now considered an essential contingency plan.

Based on the aforementioned republic acts and presidential decree, it is therefore the responsibility and obligation not only the contractors, miners, and government but also the community to secure and maintain a healthy environment to preserve the Philippine agricultural sector consequently the food safety and security of the country.

Furthermore, Zambales is also a leading province in producing raw materials from mining. Chromite mining has boosted the economy of several towns of Zambales. Harboring some of the heavy metal mining companies in Zambales, it also poses a threat to another livelihood which is fishing. Intentional or unintentional contamination may occur thru its river systems, thus, a need for immediate action should be conducted.

2. RESEARCH OBJECTIVES

The main objective of the study is to assess the biosorption capability of fish scales with a heavy metal pollution in water in terms of amount of scales used and exposure.

Specifically, the study aims the following to:

1. Prepare and simulate the chromium contamination in aqueous solution in laboratory.

2. Assess the biosorption capability of *Oreochromis niloticus Linn* scales on induced Chromium (VI) aqueous solution in terms of length of exposure and amount of scales.

3. RESEARCH METHODOLOGY

3.1 Preparation of Fish Scales:

O. niloticus L. scales were collected from the local fish market of Iba, Zambales. Fish scales were washed and soaked with distilled water to remove dust and other impurities that adhered for 24 hours and were rinsed with distilled water for three times. The biosorbent was then transferred on a laboratory tray and air dried for 24 hours. Dried *O. niloticus L.* biosorbents were then weighed and transferred to 500 mL Erlenmeyer flask containers and were covered.



Figure 1: Researcher weighing the dried fish scales in various weights

3.2 Preparation of Chromium Solution:

The researchers had prepared solutions of chromium to simulate chromium wastewater in the concentration amount of 100 μ g/mL. 1414 mg of Potassium Dichromate (K₂Cr₂O₇) was dissolved to a 5 liter of distilled water and stirred until fully dissolved to obtain the 100 μ g/mL.

3.3 Chromium Level in Induced Water Samples:

Synthetic chromium solutions were submitted to CRL Laboratory Testing Center in Clark Pampanga for pre and post measurement analysis of total chromium concentration of the prepared chromium wastewater.

3.4 Biosorbent Exposure to Synthetic Solution:

Fish scales were preserved and introduced to the prepared chromium solution in two separate set –ups: 1) Length of Time Exposure and 2) Amount of *O. niloticus L.* biosorbent (in grams) used. Length of exposure varies from 24, 48 and 96 hours (See Figure 2) while amount of *O. niloticus L.* biosorbent varies from 1, 3 and 5 grams (See Figure 3).



Figure 2: Exposure of biosorbent to synthetic wastewater with varying contact time



Figure 3: Samples with varying contact time and amount of biosorbent in triplicates

3.5 Calculation on Specific Uptake:

After the exposure of fish scales, the biosorbent was filtered out using filter paper. Although no biosorbent was added to the untreated sample, it was filtered before the determination of the final chromium to control any chromium sorption caused by the filtering process. The filtrate was then collected and the final chromium concentrations were determined in CRL laboratory in Clark Pampanga. The calculated initial and final (Cr) were used to determine the specific uptake (Q) in units of gram of Cr removed per gram of biosorbent used, and the sorption percentage (%S). \mathbf{Q} and %S were calculated using the equations,

Q = <u>(initial [Cr] – final [Cr]) X volume of sample</u> Mass of bio sorbent

> %**S**= <u>(initial [Cr] – final [Cr])</u> X100% Initial [Cr]

3.6 Varying Contact Time:

Each sample was treated under constant amount of biosorbent (5 grams) while at different contact times (24 hours, 48 hours and 96 hours). The biosorbent was then filtered out and the final [Cr] of the filtrate was determined. The **%S** and the removal rate were calculated. ./The removal rate (R_r), in units of grams of Cr removed per minute, was calculated using the equation,

Rr= <u>(initial [Cr] – final [Cr])</u> Contact time

3.7 Statistical Analysis:

To present the result effectively, the researcher applied the following statistical analysis.

3.7.1 Analysis of Variance:

This was used to measure the difference between a specific weight to other various weights of fish scales. The researcher made used of the SPSS, a statistical software used in processing data.

3.7.2 Pearson r Coefficient:

This was used to identify the significant difference of the weight of fish scales versus the concentration of Cr (VI).

3.7.3 Regression:

This was used to further show the decline of concentration of contaminants in wastewater.

4. RESULTS AND DISCUSSION

The study assessed the biosorption capabilities of *Oreochromis niloticus L*. (Tilapia) scales when exposed to chromium contaminated wastewater in an induced method. The synthetic wastewater total chromium was analyzed after exposure of the chosen biomass to observe the decrease in the heavy metal ions using Flame Atomic Absorption Spectrophotometry. Furthermore, effect of the mass of fish scales and length of exposure was measured using statistical analysis.

4.1 Characterization of Oreochromis niluticus:

Oreochromis niloticus L. locally known as Tilapia fish is a widely distributed organism throughout the world which dates back during the ancient Egyptian era. *O. niloticus L.* species are grayish to black in color with pectoral, dorsal and caudal fins becoming reddish in coloration which exhibits no sexual dimorphism and are sexually matured at five to six months of age. Moreover, such species are omnivorous type that feeds on a wide range of nutrition. *O. niloticus* are eurythermal organisms with optimum temperature range of 31to 36° C causing it to be proliferative specie in the tropical regions including Philippines and are locally culture in ponds and tanks. Furthermore, such species contain cycloid types of scales (FAO, 2015).

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Scales are overlapping hard plates that protect fishes against infection, abrasions and predation. Moreover, scales are classified based on their shape and appearance. The cycloid type fish scale of *O. niloticus L.* and is also the common type of scales found on freshwater fishes. Cycloid scales are circular shaped with smooth posterior margins and lacks ctenii or spiny extentions. Moreover, cycloid scales exhibits spacing known as circuli that are large during conducive environment and close during unfavorable ones (Creaser, 2000). Adsorptive capabilities of the cycloid scales of *O. niloticus L.* will be discussed in the succeeding section.

4.2 Length of Exposure of O. niloticus Scales in Relation to Induced Chromium:

A 5 gram dried *O. niloticus* fish scales were used and exposed to 100 μ g/mL induced chromium wastewater in different time frame such as 24 hours, 48 hours and 96 hours. The induced chromium wastewater were assessed before and after the exposure of scales to measure the sorption capabilities of the selected scales.

	Pre exposure to biosorbent (Cr Range in µg/mL)	Post exposure to biosorbent (Cr Range in µg/mL)	Sorption percentage (%S)
24 hours	100	82.03	17.97
24 hours	100	82.07	17.93
24 hours	100	82.02	17.98
48 hours	100	67.13	32.87
48 hours	100	67.03	32.97
48 hours	100	67.14	32.86
96 hours	100	48.70	51.03
96 hours	100	48.77	51.23
96 hours	100	48.93	51.07
Standard Limit	$0.0005 - 0.002 \ \mu g/mL^a$		

Table 1: Pre and post exposure of scales to wastewater at three various time frames

^a based on World Health Organization/ Chromium in surface water of 2004

Table 1 showed that levels of chromium ions decrease drastically as hours of exposure increases as observed with 24 hour exposure with about 18 μ g/mL decrease or 18% sorption percentage in chromium concentration to about 51 μ g/mL decrease or 51% sorption percentage of chromium that is found on the fish scales.

Furthermore, there is a high significant correlation between the increase in exposure time to the decrease in chromium concentration with a value of -0.991. Moreover, this signifies that there is an almost negative linear relationship which strengthens the result of decreasing chromium concentration with increasing contact time (See Appendix B). Additionally, when run with regression analysis, the negative linear correlation showed that there is a decrease of chromium concentration per hour of exposure to about 0.0047 μ g/mL (See Appendix B).

High concentration of chromium ions was used for pre exposure to simulate a high contamination in a river. In addition, based on WHO, the standard limit of chromium concentration of bodies of water should be 0.5-2 μ g/ mL. Based on the results, fish scales could lower the high chromium concentration on the water if exposed on long duration until the scales are not deteriorated and decayed.

4.3 Variation in weight of O. niloticus Scales in Relation to Induced Chromium:

A varying weight of dried *O. niloticus* fish scales in 1 gram, 3 grams and 5 grams measurements were used and exposed to 100 μ g/mL induced chromium wastewater for 4 days. The induced chromium wastewaters were assessed before and after the exposure of scales to measure the sorption capabilities of the selected varying scales weight.

	Pre exposure to biosorbent (µg/mL)	Post exposure to biosorbent µg/mL	Sorption percentage (%S)
1 gram	100	92.78	7.22
1 gram	100	92.76	7.24
1 gram	100	92.71	7.29
3 grams	100	77.09	22.91
3 grams	100	77.03	22.97
3 grams	100	77.02	22.98
5 grams	100	48.69	51.31
5 grams	100	48.72	51.28
5 grams	100	48.70	51.26

Table 2: Measurement of Pre and Post exposure of varying scales weight and Sorption percentage

Table 2 showed that levels of chromium ions decrease drastically with increasing amount of fish scales or biosorbent, that is, a 5 gram of dried scale showed about 51% sorption percentage or in other words had removed 51 μ g/mL of chromium on the induced wastewater which is different with the 7 % or 7 μ g/mL chromium concentration decrease.

Table 2 shows that with increase in weight of biomass results to decrease in concentration of chromium ions. Based on one – way analysis of variance, there is a significant difference between the chromium concentration and weight of the biosorbent at alpha level of 0.05. Moreover, there is a high significant correlation between the increasing amount of biosorbent and the amount of chromium concentration on the induced wastewater with an r – value of – 0.987. That is, there is a 98.7% noticeable decrease on the amount of chromium as indicated by the negative sign with increasing amount or weight of the scales. Having this said, it is also calculated based on the regression analysis that there is a negative linear relationship, thereby, strengthening the result of the Pearson - r Correlation. Furthermore, based on the regression analysis, there is about 11 µg of chromium being adsorb per gram of fish scale. (See Appendix C and Appendix D). In addition, it is very evident that sorption percentage is above 50% which makes it a very effective way of reducing chromium ions in wastewater.

Discussion:

Biosorption is affected by several factors both on the adsorbent scales as well as on the adsorbate or the induced wastewater. In relation to this, the characteristics of the adsorbent such as the amount, diameter, surface area, pore size, chemical composition of the scales all contribute to its capacity to adsorb heavy metals. Consequently, the characteristics of the induced chromium wastewater should also be observed such as its contact time (DESOTEC, 2013).

One of the possible factors contributing to the lowering of chromium concentration is the presence of high number of circuli on the *O. niloticus* cycloid scales. Circuli or the space in between the ridges on the individual scales serves as reservoir where chromium elements could settle and bond with protein substances that constitute the fish scales further tapping the said elements within the scales and not suspended on the liquid medium. Furthermore, such high number of spaces increases the surface area where high amount chromium elements could be trapped and bonded on the scales.

In accordance with this, *O. niloticus* scales contain collagen which is an organic protein . Collagen contains different functional groups that could bind to the metal Cr^+ such as the phosphate, carboxyl, amine and amides which are negatively charged, thereby, attracting and binding more Cr elements to the surface of the scale (Prabu, et. al., 2012).

Therefore, a long exposure of the biosorbent on the wastewater paves way into more chromium elements bonding with the chemical constituents of the scales. Furthermore, the longer the time of exposure means that there are more chromium elements that settle on the large surface area as contributed by the high number of circuli.

Moreover, with a constant time frame, a large amount of fish scales causes more areas for bonding sites and larger surface areas given by the high amount of circuli that each scales contribute.

Moreover, there are other possible contributing factors to Cr removal such as the presence of microbes onn the fish scales. The study done by Mustafiz et al. (2003) suggested that microbes were responsible in heavy metal removal with the application of fish scales as a biosorbent. Furthermore, longer contact time allowed the microbes to be released into the Cr solution, thereby, either remediating or absorbing Cr with their cellular body. (Mustafiz, et al, 2003).

Consequently, based on the study, biosorption using fish scale as way of rehabilitating the area with high concentrations of Chromium could do several methodologies to achieve greater results. One of which is by using large amounts of scales and exposing it in a longer duration of time as permitted by the lifespan of the scale. Other methodology is by subjecting the wastewater to small amount of fish scale but with a longer exposure time and another is high amount of fish scales in short duration of time (Huang, 2007). Either ways, the main objective is to lower the heavy metals that contaminate the environment in an effective and eco – friendly way.

5. CONCLUSION

The initial research locale is situated in a river next to a closed chromite mining site within Cabangan, Zambales. However, tests showed that the total chromium is within the normal range for surface water. Thus, the researchers opted to conduct the experiment in an induced method. Additionally, tilapia fish scales were collected from a fish vendor in Iba, Zambales wet market and was washed and rinsed repeatedly and air dried to remove adhering dust and mucus. Total chromium was measured using Flame Atomic Absorption Spectrophotometry after exposure under different length of contact and different weight of biomass.

Induced method was used to simulate a severe contamination of chromium in an aqueous solution. Standard limit of total chromium in surface water published by the World Health Organization was used for reference.

One – way analysis of variance at alpha 0.05 evidently showed that there is a significant difference in weight and length of exposure. Furthermore, statistics show that correlation of chromium to mass of fish scales is measured at -.987 stating that as weight of scales increases, final concentration of wastewater decreases. In addition, correlation is measured at - 0.991 in terms of length of exposure to heavy metal concentration stating that as hours of exposure increases, final concentration of heavy metal decreases.

Chromium are naturally present in food and water but in minimal or in within normal range. Excessive amount of chromium pose a threat to human health which causes dermatitis and causes gastrointestinal effects in humans and animals, including abdominal pain, vomiting, and hemorrhage if ingested.

6. RECOMMENDATIONS

Based from the findings and conclusions, the researchers hereby formulated the following recommendations:

- 1. Locate an exisiting chromium contaminated river within the province to showcase effect of fish scales.
- 2. Different heavy metal can also be used to see if the same interaction will be observed.
- 3. Different fish scales can also be used for comparison to seek the best biomass in terms of specie.

4. The researchers highly recommend to seek funding for Electron Microscopy for graphic representation of the adsorption process

REFERENCES

- Aguilar, A. (2013). Greenpeace to DENR: Implement pollution disclosure to save our rivers. Retrieved from http://www.greenpeace.org/seasia/ph/press/releases/Greenpeace-to-DENR-Implement-pollution-disclosure-to-saveour-rivers/ (BLOG/WEBSITE)
- [2] Ahalya, N. et al. (2003). Biosorption of heavy metals. Res. J. Chem. Environ 7:71-78. Retrieved last February 2016 from www.ces.iisc.ernet.in/energy/water/paper/biosorption/biosorption.html

- [3] Begum, H. et al. (2013). Removal of Brilliant Red from Aqueous Solutions by Adsorption of Fish scales. Dhaka Univ. J. Sci. 61(1): 7-12 Retrieved from http://www.banglajol.info/index.php/DUJS/article/viewFile/ 15089/10714
- [4] Creaser, C. (2000). The Structure and Growth of the Scales of Fishes in Relation to the Interpretation of their Life-History, with special Reference to the Sunfish *Eupomotis gibbosus*. University of Michigan. Pp.12-31 http:// deepblue.lib.umich.edu/bitstream/handle/2027.42/56262/MP017.pdf?sequence=1
- [5] DESOTEC Laboratories, 2014. Adsorption. Retrieved from http://www.desotec.com/activated-carbon/what-is-adsorption/ (WEBPAGE)
- [6] Environmental Protection Agency (EPA). (2013). National Rivers and Streams Assessment 2008 2009. Washington: Office of the Research and Development. Retrieved from http://water.epa.gov/type/rsl/monitoring/ riverssurvey/
- [7] Food and Agriculture Organization (FAO). (2015) *Oreochromis niloticus*. Retrieved last February 2016 from http://www.fao.org/fishery/culturedspecies/Oreochromis_niloticus/en#tcNA0078
- [8] Gavrilescu, M. (2004). Removal of Heavy Metals from the Environment by Bio sorption Engineering in Life Sciences 4(3):219 – 232 Retrieved from http://onlinelibrary.wiley.com/doi/10.1002/elsc.200420026/abstract
- [9] Igwe, J. C. and Abia, A. A. (2006). A bioseparation process for removing heavy metals from waste water using biosorbents. African Journal of Biotechnology Vol. 5 (12), pp. 1167-1179, Retrieved from http://www.uiennieu ws.nl/kennis/docs/A%20bioseperation%20process%20for%20removing%20heavy%20metals%20 from%20waste% 20water%20using%20biosorbents.pdf
- [10] Huang, E. (2007). Use of Fish Scales as Biosorbent for the Removal of Copper in Water. Retrieved from http://nature.berkeley.edu/classes/es196/projects/2007final/Huang.pdf
- [11] Huriye et al. (2013). Adsorption of Telon Red FRL From Aqueous Solutions On Fish Scale. J. Selcuk Univ. Nat. Appl. Sci. (2013) 795–804.Retrieved from http://www.josunas.org/login/index.php/josunas/article/download /269/243
- [12] Kratochivil, D. and B. Volesky (1998). Advances in the biosorption. Volume 16, Issue 7, p291–300 Retrieve last February 2016 from doi:http://dx.doi.org/10.1016.S0167-7799(98)01218-9
- [13] Lenntech, B. (2015). Water Treatment Solutions. Retrieved from http://www.lenntech.com/water-pollution-faq.htm (WEBPAGE)
- [14] Mustafiz, S. (2003) The Application of Fish Scales in Removing Heavy Metals from Energy Produced Waste Streams: The Role of Microbes. *Energ. Source*, 25:905–916. Retrieved last February 2016 from doi:10.1080/ 00908310390221255
- [15] Prabu, K. et. al..(n.d.) A Biosorption of heavy metal ions from aqueous solutions using fish scales (*CatlaCatla*).
 World Journal of Fish and Marine Sciences 4 (1): 73-77 Retrieved from www.idosi.org/wjfms/wjfms4% 281%2912/13.pdf
- Petersen, F. et al. (n.d.). Biosorption of Heavy Metals from Aqueous Solutions. WRC Report, vol. 1, iss. 100, 2005 Retrieved from http://www.wrc.org.za/Knowledge%2520Hub%2520Documents/Research%2520Reports/1259-1-05.pdf
- [17] Rodriguez, I. (2013).Fish scales of *Chanos chanos* and *Tilapia nilotica* as natural sorption material in the recovery of Astaxanthin. Retrieved last February 2016 from www. studymode.com/essays/Fish-Scales-Wastes-For-The-Recovery-13416111.html (WEBPAGE)
- [18] Volesky, B. and Holan, Z. R.: 1995, 'Review: biosorption of heavy metals', Biotechnol. Progr.11, 235-250.
- [19] Zambrano and Gruba (2013). Philippine mining law update: features of the new executive order 79. ZGLAW.com/wp/Philippines-mining-law-update-features-of-the-new-eo-79/ (WEBPAGE)

APPENDIX- A

Table 3: Pearson - r correlation of length of exposure Chromium concentration

Correlations					
		Biosorption Percentage	Length of Exposure (hours)		
	Pearson Correlation	1	991**		
Biosorption Percentage	Sig. (2-tailed)		.000		
	Ν	9	9		
	Pearson Correlation	991**	1		
Length of Exposure (hours)	Sig. (2-tailed)	.000			
	N	9	9		
**. Correlation is significant at the	he 0.01 level (2-tailed).				



Figure 4: Graph showing decline in the concentration as length of exposure increases

Table 4: Pearson - r correlation on mass of biosorbent to concentration

Correlations						
		Weight of fish scales	Concentration of Chromium			
Weight of fish scales	Pearson Correlation	1	987**			
	Sig. (2-tailed)		.000			
	Ν	9	9			
	Pearson Correlation	987**	1			
Concentration of Chromium	Sig. (2-tailed)	.000				
	Ν	9	9			
**. Correlation is significant at the 0.01 level (2-tailed).						



Figure 4: Graph showing decline on final concentration as weight of biomass increases