Korespondensi Artikel dengan Judul

The Metals Oxide of Abandoned Tin Mining Pit Waters as An Indicator for Bacterial Diversity and Aquaculture

adec kurnia <andri_pangkal@yahoo.co.id>
Kepada:zoobiomag2004@yahoo.com
Min, 21 Jun 2020 jam 19.44

Article title:

THE METALS OXIDE OF ABANDONED TIN MINING PIT WATERS AS AN INDICATOR FOR BACTERIAL DIVERSITY AND AQUACULTURE

Name of the author**: Andri Kurniawan**

Hereby I would like to submit the manuscript entitled "**The Metals Oxide of Abandoned Tin Mining Pit Waters as An Indicator for Bacterial Diversity and Aquaculture**" to Aquaculture, Aquarium, Conservation & Legislation - International Journal of the Bioflux Society.

This manuscript was not submitted or published to any other journal.

The author declare that the manuscript is an original paper and contain no plagiarised text. All authors declare that they are not currently affiliated or sponsored by any organization with a direct economic interest in subject of the article. My co-authors have all contributed to this manuscript and approve of this submission.

Corresponding author

Andri Kurniawan

Tudor Papuc <ptudor2008@yahoo.com>
Kepada:andri_pangkal@yahoo.co.id
Jum, 26 Jun 2020 jam 02.03
Dear Dr. Andri Kurniawan,

This is a message of confirmation regarding the reception of your manuscript **The Metals Oxide of Abandoned Tin Mining Pit Waters as An Indicator for Bacterial Diversity and Aquaculture**, submitted to our journal AACL Bioflux. I will soon contact you with further information regarding your manuscript.

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For any questions, please contact me, on this email address.

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Thank you for considering our journal.

Best regards,

Tudor Papuc

Editor, Bioflux SRL

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I send my revised my manuscript with entitled

The metals oxide of abandoned tin mining pit waters as an indicator for bacterial diversity and aquaculture

I hope my manuscript can be published on your journal.
Thank you for attentions
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Kepada:adec kurnia
Sel, 30 Jun 2020 jam 22.09
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Tudor Păpuc
Editor, Bioflux

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andri_pangkal <andri_pangkal@yahoo.co.id>

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Sab, 18 Jul 2020 jam 02.03
Dear Tudor Papuc

Let me to know my manuscript progress on your journal and how could i get the fast track way for my manuscript publishing?

thank you for your information

regards,

andri

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Sen, 20 Jul 2020 jam 14.57
Yes, you can choose fast track.
In that case, I will give you an answer (almost certainly preliminary accepted) and payment details in about 1 week.

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Sen, 20 Jul 2020 jam 20.05

Thank you for the information. I hope I can choose fast track for my article publishing.

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Rab, 5 Agu 2020 jam 02.50

Hello,

Your paper is preliminary accepted for publication. Please find attached the preliminary acceptance letter. The final acceptance or rejection will be confirmed after receiving the double-blind peer reviews. If you can, please recommend 1-2 reviewers for your paper, because it would go much faster that way.

After this acceptance you should pay 300 USD for publication (open access - priority processing).

As the payment and bank transfer may be time consuming you may start the process of payment right now; we will send you the invoice immediately after the payment.

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Let me know if you need more details.

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Hello
Dear Tudor Papuc

I would like to say thank you for this information.

I recommend 2 reviewer based on their capability on bacterial and aquaculture science.
1. Dewi Embong Bulan, Ph.D Institution: University of Mulawarman
email: dewi.embong@fpik.unmul.ac.id
Scopus ID: 57195241377

2. Dr. Agus Setiawan
Institution: University of Lampung email: agusu.san@gmail.com agus.setyawan@fp.unila.ac
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For the publication payment, I want to apologize because my funder wants my manuscript to be declared accepted first, then after that the publication funds will be given.
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thank you.

Regards

Tudor Papuc <ptudor2008@yahoo.com>
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Sen, 10 Agu 2020 jam 14.01
Yes, this is ok.
I have contacted the reviewers and expect replies.
After they give me the positive evaluations, I will give you a final acceptance letter, and you can make the payment afterwards.

Thank you,

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Tudor Păpuc
Editor, Bioflux

Tudor Papuc <ptudor2008@yahoo.com>
Kepada:andri_pangkal

Sab, 12 Sep 2020 jam 02.57
We have the 2 positive evaluations.
I am attaching the final acceptance letter. I can say that the paper will be published, but at this point, I can't know exactly if in issue 5 or issue 6.
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Thank you,

Best Regards,
Tudor Păpuc
Editor, Bioflux

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adec kurnia <andri_pangkal@yahoo.co.id>
Kepada: Tudor Papuc
Sab, 12 Sep 2020 jam 05.41
Dear Tudor Păpuc
Editor, Bioflux

I would like to say thank you for your information and the final acceptance decision.
I will make the publication payment soon, I hope I can pay and tell you about the payment on Wednesday (16/9/2020). I will send the proof of payment after I pay it.

for this moment, I want to ask a question...could I add "an acknowledgment" on my paper for my funder?
I would like to say thank you for Bangka Belitung University for the funding of this publication.
I hope you can add it on my paper.

Thank you for your help.

Best Regards,
Andri Kurniawan

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adec kurnia <andri_pangkal@yahoo.co.id>

Kepada: Tudor Papuc
Jum, 18 Sep 2020 jam 10.12

Dear Tudor Papuc
I want to tell you that i have paid my publication fee amount 300 USD and all the bank transfer costs are in my concern and not to the publisher. I attach the transfer form from the bank.
Thank you for your attentions.

Best Regards
Andri Kurniawan

Tudor Papuc <ptudor2008@yahoo.com>
Kepada:adec kurnia
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Thank you, we are verifying the payment, but everything should be ok.
I am sending you soon the paper with comments.

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Tudor Păpuc
Editor, Bioflux

---

andri_pangkal <andri_pangkal@yahoo.co.id>
Kepada:ptudor2008@yahoo.com
Sab, 10 Okt 2020 jam 06.05
thank you for the invoice dan information.
I am waiting for the paper.

best regards,

Tudor Papuc <ptudor2008@yahoo.com>
Kepada:andri_pangkal
Jum, 23 Okt 2020 jam 02.24
Ok, I am sending you the paper with comments, English corrected and formatted, and references checked.
There is one big problem, but we both can find a solution, and eventually publish the paper. You mention very few things in Material and Method, and this can simply be corrected (you will see the comments). But in Results and Discussion, you seem to present the results from other studies of yours, because you have citations for every results.

If you have your own results, please make that clear, and present them (this would be the ideal solution). If not, please try to restructure the paper as a short review, because you have many references, and it would work as a short review. You would have to change some phrases in Material and Method mostly, and in some other places, but it could be done. You could even remove the Material and Method, because reviews do not need to respect exactly the same structure as original research articles.

Ok, so please choose 1 of the 2 scenarios, and try to repair the paper. If you have other suggestions as to how to proceed, I am open to them. Also, please mark all changes with yellow highlight or track changes, so I can check them, and work on this version of the manuscript. Thank you,

Best Regards,
Tudor Păpuc
Editor, Bioflux

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adec kurnia <andri_pangkal@yahoo.co.id>

Kepada: Tudor Papuc
Sen, 26 Okt 2020 jam 04.16
Dear Tudor Papuc

I am sending my manuscript revision.
Thank you for your attentions.

Regards,
andri

Tudor Papuc <ptudor2008@yahoo.com>
Kepada:adec kurnia
Kam, 29 Okt 2020 jam 14.17
I received the document and will soon send you the final version.
Thank you,

Best Regards,
Tudor Păpuc
Editor, Bioflux

Tudor Papuc <ptudor2008@yahoo.com>
Kepada:adec kurnia
Kam, 19 Nov 2020 jam 14.38
Hello, please find attached the final form of the manuscript. The revisions you made were all accepted, with the following exceptions:

1. In the title the aquaculture part was removed, because there was too little text about aquaculture in mining pit waters, and there was no experiment about this.

2. From the text, the parts were removed because they mentioned the possibility of aquaculture without considering all aspects, like the higher cost of aquaculture because of improper water parameters, or heavy metal contamination in the flesh of fish destined for human consumption.

3. The references were corrected (again). Please do no change them. For example, the reference Fernandes et al 2018 was changed to Cesario Fernandes et al 2018, because the family name is Cesario Fernandes, not just Fernandes (Spanish people do this often, and in this case, Cecilia is the first name - woman name - and Cesario Fernandez the family name; please leave it like this even though in some other places is cited Fernandes C. C. - which is incorrect).

4. Also, the English was corrected again, but there were only minor changes.

So please read carefully the manuscript and let me know if you agree with the publication. The publication date might be 30 October, because that issue is still open, and there are a few more places for papers there. If the issue closes, the paper will be published with the first dates of November as the publishing date (1-3 Nov).

Thank you,

Best Regards,

Tudor Păpuc
Editor, Bioflux

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adec kurnia <andri_pangkal@yahoo.co.id>
Kepada: Tudor Papuc
Jum, 20 Nov 2020 jam 02.42
Dear Tudor Păpuc

I agree with the final revision.
Thank you.

Best Regards,
Andri

Tudor Papuc <ptudor2008@yahoo.com>
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Congratulations! The manuscript has been published!
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Sen, 23 Nov 2020 jam 08.44
Dear Tudor Păpuc
I would like to thank for your attentions in my publication.

Best Regards
Andri

THE METALS OXIDE OF ABANDONED TIN MINING PIT WATERS AS AN INDICATOR FOR BACTERIAL DIVERSITY AND AQUACULTURE

Andri Kurniawan[1]*

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ABSTRACT

This study aims to provide information about the form of metal oxides identified in post-mining underwater waters with different ages and explain their relationship to the diversity of acidophilic bacteria in acid mine waters. Analysis of metal oxides was carried out using *X-Ray Fluorescence* (XRF). The sixteen oxide forms of heavy metals identified show the highest concentrations of Iron (III) Oxide or Fe_2O_3, Tantalum (V) Oxide or Ta_2O_5, Tin (IV) Oxide or SnO_2, Manganese (II) Oxide or MnO is also found in high concentrations in all spheres of different ages. The form of metal oxides can represent the metal content contained in these waters. The metal oxide also explains that the pattern of metal oxide concentration decreases with the duration of the underwater chronoseconds such as As_2O_3, Cr_2O_3, CuO, NiO, PbO, and ZnO, although not all metals experience a pattern of decreasing concentration over the duration of chronosequences in these waters like Ga_2O_3, HfO_2, MnO, TeO_2, and ThO_2. The implications of the presence of metals and their oxides can affect the acidity (pH) and indirectly have an impact on the diversity of microbes, especially bacteria that affect the environment and aquaculture commodities.

Keywords: Element Oxides, Pits, Tim Mining, X-Ray Fluorescence, Bacterial, Aquaculture.

INTRODUCTION

A number of metals have been identified in aquatic ecosystems, especially in artificial lakes (pit) after tin mining activities. These metals become components of ecosystem pollutants (Dinis & Fiuza 2011; Guan et al 2014; Kurniawan 2016) including those classified as heavy metals such as Pb, Zn, Mn, Fe, Cr, Cu, Ni, Cd, Sn, and As (Ashraf et al 2011a; Henny 2011; Ashraf et al 2012a; Rosidah & Henny 2012; Daniel et al 2014).

The use of the general term heavy metal refers to a group of metals and semi-metals (metalloids) associated with contamination, having a density > 5 $g.cm^{-3}$ or in other studies explained above 3.5-7.0 $g.cm^{-3}$, atomic weights between 63.546 (≈ 63.6) - 200.590 (≈ 200.6), and specific gravity > 4.0 or 5.0 (Duffus 2002; Srivastava & Majumder 2008; Aslam et al 2011).

Heavy metals are not always described as dangerous metals (toxic metals), but their chemical structure determines the biological properties and toxicity of these elements (Templeton 2015). Some metals in certain structures are needed by the body as essential micromineral (trace element), but in other structures it is dangerous (Kurniawan & Mustikasari 2019). Elements such as chromium in the form of Cr(III) are important trace elements, but Cr(VI) can cause cancer (Govind & Madhuri 2014), Hg(II) is more toxic than Hg(0) (Azimi & Moghaddam 2013); anorganic Mn(III) is more toxic than oxidation forms such as $Mn(II)Cl_2$ and $Mn(IV)O_2$; As(III) is more toxic than As(V); element V(V) is more toxic than V(IV) (Templeton 2015), and Fe(II) is more and significantly absorbed by cells than Fe(III) (He et al 2008). The chemical structure is confirmed in the ionization that occurs and gives an idea of the toxicity of an element (element) is. Ionization of heavy metals can potentially be disruptive and dangerous to health and can even damage the vitality of the body' systems (Abdi & Kazemi 2015).

Microecosystem changes can be indicated by the diversity of microorganisms in an ecosystem because it can be related to variations in water characteristics (Ashraf et al 2011b) which are determined through weather, geomorphology, and geochemical conditions (Ashraf et al 2012b). Studies on the relationship between bacterial diversity and its role in the biogeochemical cycle and the interaction of microorganisms with their environment have been widely conveyed (Bhowal & Chakraborty 2015; Fashola et al 2015) including Methane-Oxidizing Bacteria (MOB), Ammonia-Oxidizing Bacteria (AOB) (Sow et al 2014a; Sow et al 2014b), and Arsenic-Resistant Bacteria (ARB) (Jareonmit et al 2010; Valverde et al 2011) found in post-mining ecosystems. The study of the diversity of microorganisms and their activities as ecological bioindicatory agents is an important step to predict ecosystem conditions and environmental changes (Niemi & McDonald 2004; Moscatelli et al 2005), including tin mining. This is because microorganisms have the capacity to respond to changes that occur in the environment quickly, including in soil and aquatic ecosystems (Paerl et al 2003; Lau & Lennon 2012).

This short communication aimed to provide information related to the form of metal oxides, especially the heavy metals identified in post pit mining waters with different ages (chronosequences) and to explain their relationship to the diversity of acidophilic bacteria in acid mine waters and the potential of the waters for aquaculture.

EXPERIMENTAL SECTION

The study was conducted by testing the metal content in water taken from under the tin mining post with different ages, namely < 1 year, 5-10 years, and > 15 years. Water sampling was carried out in the pit located in Bangka Regency, Bangka Belitung Islands Province. The research stations were coded Station A for under the age of < 1 year, Station B for under the age of 5-10 years, and Station C for under the age of > 15 years. The coordinates of the research station were

Station A at 01°59' S and 106°06' E; Station B at 01°59' S and 106°06' E; and Station C at 01° 55' S and 106°06' E (**Figure 1**) (Kurniawan 2019; Kurniawan et al 2019).

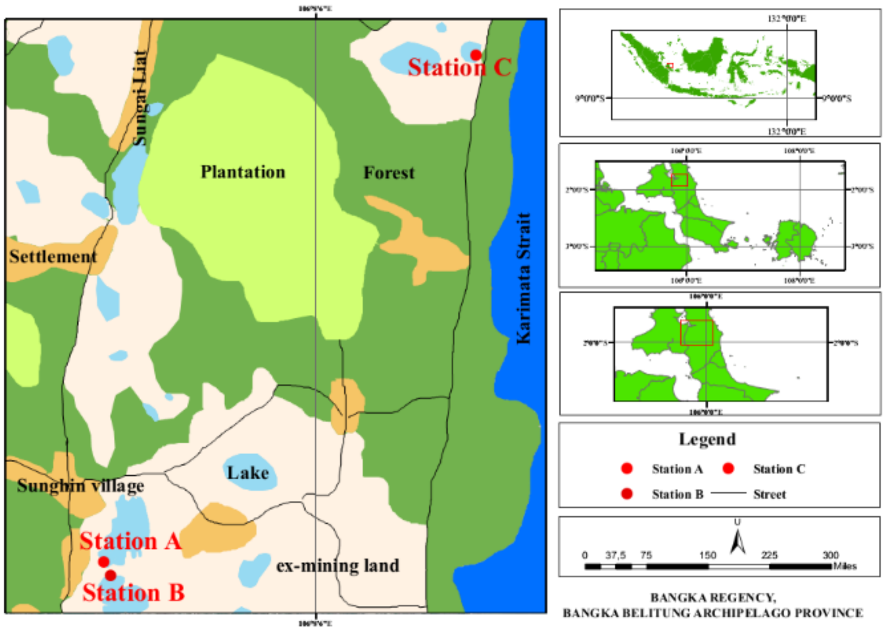

Fig. 1. Map of the research station, which is the ex- tin mining pit in Bangka Regency, Bangka Belitung Islands Province.

Analysis of the metal content or elements was carried out using X-Ray Fluorescence (XRF) instruments (Kodom et al 2012) Rigaku NEX CG with cross-section specifications of three refracting metals, namely copper (Cu), molybdenum (Mo), and aluminum (Al). Data analysis was performed descriptively to explain the concentration of metal oxides identified in the sample.

RESULTS AND DISCUSSION

The results of the heavy metals analysis identified in previous studies, namely As, Co, Cr, Cu, Fe, Ga, Hf, Mn, Ni, Pb, Sn, Ta, Te, Th, V, and Zn (Ashraf et al 2011b; Ashraf et al 2012b). The sixteenth metal shows oxidation as shown in **Table 1**.

Table 1. Concentrations of metal oxides in post pit mining waters

Metals Form	Metals Oxide Form	Name of Oxide Form	Average of Metals Oxide Concentration at the Research Station (ppm)		
			A	B	C
As	As_2O_3		5.70	8.49	3.04
Co	Co_2O_3		14.20	ND	9.34
Cr	Cr_2O_3		13.80	14.50	1.96
Cu	CuO		7.03	7.83	6.91
Fe	Fe_2O_3		1424.3	2307.67	1571.93
Ga	Ga_2O_3		11.50	12,00	12.65
Hf	HfO_2		7.74	8.76	11.07
Mn	MnO		33.70	35.10	39.05
Ni	NiO		10.40	7.32	4.64
Pb	PbO		14.50	13,00	9.40

Sn	SnO_2	89.73	64.77	74.53
Ta	Ta_2O_5	987.33	1373.33	888.73
Te	TeO_2	13,00	9.48	14.50
Th	ThO_2	10.50	9.97	15.75
V	V_2O_5	3,00	ND	2.38
Zn	ZnO	3.00	ND	ND

Noted: ND (not detected).

The highest concentration was Fe_2O_3 or Iron(III) Oxide. Other formations were Ta_2O_5 or Tantalum(V) Oxide, SnO_2 or Tin(IV) Oxide, and MnO or Manganese(II) Oxides were also found to be highly concentrated in all pits of different ages. The form of metal oxides can indicated or represented the metal content contained in these waters. The content of Fe_2O_3 has the highest value under the age of 5-10 years compared to under the age of < 1 year and > 15 years which indicated that the highest Fe metal was also found under the age of 5-10 years. Fe metals were also highest in all voids compared to other metals.

The highest MnO found in under the age of > 15 years indicated that the potential for heavy metal contamination was still very large under the old age. This indicator explained that not all metals experience a pattern of decreasing concentrations as the length of chronosequences in these waters. This condition were also seen in metals or oxides from Gallium (Ga) as Ga_2O_3, Hafnium (Hf) as HfO_2, Manganese (Mn) as MnO, Tellurium (Te) as TeO_2, and Thorium (Th) as ThO_2. The decreasing pattern of metal or its oxide concentration with the duration of chronosequence were shown in Arsenic (As) as As_2O_3, Chrome (Cr) as Cr_2O_3, Copper (Cu) as CuO, Nickel (Ni) as NiO, Lead (Pb) as PbO, and Zinc (Zn) as ZnO.

DISCUSSION

A number of metals and their oxides contributed to water quality parameters such as pH. It was seen that under the age of <15 years showed a low pH value or acidic (pH = 3), while the under the age of> 15 years had a neutral pH value (pH = 7) (Kurniawan 2019; Kurniawan et al 2019). A number of metals indicated by the formation of the oxide, namely Fe_2O_3, As_2O_3, Cr_2O_3, CuO, NiO, PbO, and ZnO were found high under <15 years old which has an acidic pH or under a 15 year old which has a neutral pH.

A number of minerals undergo chemical reactions that form acidic pH are categorized as Potentially Acid Forming (PAF) such as Cu, Fe, Pb, Zn (Celebi & Oncel 2016), Al, As, Cd, Co, Cr, and Mn which are formed from leaching of mined minerals (Campaner et al 2014). Oxidation and hydrolysis reactions in elements such as S, Fe, Cu, Zn, Ni, Cr, and Pb cause it to change to form cations of Cu^{2+}, Zn^{2+}, Ni^{2+}, Cr^{3+}, and Pb^{2+}. The increasing in proton H^+ contributes to the increase in acidity. An increasing number of H^+ ions can cause pH conditions in these environments to become more acidic (Gaikwad & Gupta 2008; Hatar et al 2013). The existence of these metals can directly or indirectly affect the pH value (De Saedeleer et al 2010; Zhao et al 2010; Fernandes et al 2011; Strom et al 2011; Huang et al 2012; Sadeghi et al 2012; Zhang et al 2014) and form acidity in mining waters known as Acid Mine Drainage (AMD) or Air Asam Tambang (Ashraf et al 2011b; Kurniawan 2019; Kolmert & Johnson 2001; Bigham & Nordstrom 2000; Tan et al 2007). The consequences of the acidity of waters can cause not all organisms to be able to live optimally at the pH conditions. Organisms at the microscopic level such as bacteria and archaea that are acidophilic (acidophile) have the ability to survive and live optimally in extreme acidic conditions as in acid mine waters (Navarro et al 2013). Acidophilic groups have the capacity to modify the physical and chemical conditions of waters by detoxifying or exploiting their metabolism [48] and play an important role in the biogeochemical cycles of iron and sulfur (Fashola et al 2015).

Acidophilic is grouped into two, namely true achidophile (extreme acidophiles) can live at pH 2.7 and even some can live at pH < 1.0 with optimum growth pH < 3.0 and moderate acidophile that can live in the pH range 3.0 -7.2 with optimum growth pH of 3.0-5.0 (Johnson & Hallberg 2008; Mendez et al 2008; Oren 2010). Various studies explain that a number of groups of bacteria that are found in mining areas are derived from Phyla Proteobacteria, Acidobacteria, Chloroflexi, Cyanobacteria, Actinobacteria, Nitrospirae, Firmicutes, Planctomycetes, Bacteroidetes, and Chlorobi (Gupta 2000; Lefebvre et al 2010; Hua et al 2015; Mesa et al 2017;Teng et al 2017; Fernandes et al 2018).

The optimum growth of acidophilic bacteria takes place between pH 1.0 and pH 5.0. This distinguishes it from neutrophilic that lives tolerant in the pH range of 5.0-9.0 with the optimum pH of 7.0 and alkaliphilic which is tolerant of life at pH 6.0-12.0 with an optimum pH above 9.0 or often between pH 10.0 and pH 12.0 (Gupta et al 2014; Horikoshi 2016). Acidophilic bacterial groups have the ability to reduce and oxidize metals such as iron, sulfur and other minerals (Islam et al 2004; Hallberg 2010; Harahuc et al 2000; Yli-Hemminki et al 2014), help the carbon cycle flow (Wegner & Liesack 2017; Hausmann et al 2018; Sun et al 2018), nitrogen retarders (Gargaud et al 2011; Sun et al 2015), and play a role decomposing organic matter (Khare & Arora 2015). This capability is generally used in the process of detoxification of contaminated waters (Davis-Belmar & Norris 2009; Johnson et al 2009; Murali et al 2014; Shivlata & Satyanarayana 2015; Hu et al 2018) both inorganic and organic.

The presence and biological activity of microbial groups in environments containing sulfide minerals can accelerate the formation of acidic conditions in the environment (Rawlings 2005). Acidic pH conditions involve oxidation processes and complex chemical reactions to produce H^+ ions, sulfates (SO_4^{2-}), Mn^{3+}, and other ions. More and more of these ions that form in an environment causes acidity to increase and pH to be low (Gaikwad & Gupta 2008; Hatar et al 2013; Nurofiq et al 2016). The acceleration of the formation of acidic conditions can involve biological interactions such as microbial metabolic activity (Violante et al 2010).

The oxidation process of sulfide elements such as iron and sulfur in acidic waters produces an energy reaction which is used by acidophilic microbes for growth and metabolic functions. Sulfur oxidizing microbes utilize the iron cycle under acidic pH conditions to use ferrous iron ions (Fe^{2+}) as electron donors and ferric iron (Fe^{3+}) as electron acceptors (Lei et al 2016). The oxidation process involving acidophilic bacteria is due to the ability of metabolism such as enzymes. Sulfur metabolic enzymes work to oxidize the element sulfur with sulfur dioxygenase, sulfur oxygenase reductase, and Hdr-like complex; thiosulfate oxidizing enzymes such as sulfuroxidizing enzyme and thiosulfate dehydrogenase; sulfide oxidizing enzymes such as sulfide:quinone oxidoreductase (Wang et al 2019) and iron oxidizing enzymes (Li et al 2017).

The presence and use of acidophilic bacteria in acid mine waters can be optimized by studying their biochemical characteristics. Individuals who have the potential to be bioremediators can be explored to tackle environmental pollution in acidic waters. This contributes as an effort to improve the condition of the aquatic environment so that it can be utilized by humans and comfortable habitats for aquatic organisms in aquaculture.

CONCLUSION

The presence of a number of metals and their oxides in ex-tin mining or abandoned pits waters has shown an association with the formation of pH conditions in the environment. The implication of the change in pH, especially on acidic pH causes a number of acidophilic groups to appear. Acidophilic groups have the ability to live in these extreme environments and then move biogeochemically. The potential ability of these acidophilic bacteria can be utilized to carry out a detoxification process for inorganic and organic contamination found in underwater waters. Optimizing the role of acidophilic bacteria is expected to be beneficial for environmental improvement so that it becomes better and beneficial to humans and is suitable for the life of aquatic organisms.

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Ashraf, M.A., Maah, M.J., and Yusoff, I., 2011b, Analysis of physio-chemical parameters and distribution of heavy metals in soil and water of ex-mining area of Bestari Jaya, Peninsular Malaysia, *Asian Journal of Chemistry*, 238, 3493-3499.

Ashraf, M.A., Maah, M.J., and Yusoff, I., 2012b, Morphology, geology and water quality assessment of former tin mining catchment, *The Scientific World Journal*, 2012, 1-15.

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The metal oxides of abandoned tin mining pit waters as an indicator for bacterial diversity and aquaculture

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Abstract. This study aimed to provide information about the form of metal oxides identified in abandoned tin mining pit waters with different ages and explain their relationship to the diversity of acidophilic bacteria in acid mine waters. The analysis of metal oxides was carried out using X-Ray Fluorescence (XRF). The 16 oxide forms of heavy metals identified showed that iron (III) oxide (Fe_2O_3), tantalum (V) oxide (Ta_2O_5), tin (IV) oxide (SnO_2), manganese (II) oxide (MnO) were found in high concentrations in all mine waters of different ages. The form of metal oxides can represent the metal content contained in these waters. The pattern of metal oxide concentration decreases with the duration of the pit waters chronosequences, such as As_2O_3, Cr_2O_3, CuO, NiO, PbO, and ZnO, although not all metals experience a pattern of decreasing concentration over the duration of chronosequences in these waters, exceptions being Ga_2O_3, HfO_2, MnO, TeO_2, and ThO_2. The implications of the presence of metals and their oxides can affect the acidity (pH) and indirectly impact the diversity of microbes, especially bacteria that affect the environment and aquaculture commodities.

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Key Words:

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Introduction. A number of metals have been identified in aquatic ecosystems, especially in artificial lakes (pits) after tin mining activities. These metals become components of ecosystem pollutants (Dinis & Fiuza 2011; Guan et al 2014; Kurniawan 2016), including as heavy metals such as Pb, Zn, Mn, Fe, Cr, Cu, Ni, Cd, Sn, and As (Ashraf et al 2011a; Henny 2011; Ashraf et al 2012a; Rosidah & Henny 2012; Daniel et al 2014). The use of the general term "heavy metal" refers to a group of metals and semi-metals (metalloids) associated with contamination, with a density higher than 3.5-5 g cm^{-3}, atomic weights between 63.546 (≈63.6) and 200.59 (≈200.6), and specific gravity higher than 4 (Duffus 2002; Srivastava & Majumder 2008; Aslam et al 2011).

Heavy metals are not always described as dangerous metals (toxic metals), but their chemical structure determines the biological properties and toxicity of these elements (Templeton 2015). Some metals in certain structures are needed by the body as essential microminerals (trace elements), but in other structures are dangerous (Kurniawan & Mustikasari 2019). Elements such as chromium in the form of Cr (III) are important trace elements, but Cr (VI) can cause cancer (Govind & Madhuri 2014), Hg (II) is more toxic than Hg (0) (Azimi & Moghaddam 2013), anorganic Mn (III) is more toxic than oxidation forms such as Mn (II) Cl_2 and Mn (IV) O_2; As (III) is more toxic than As (V); element V (V) is more toxic than V (IV) (Templeton 2015), and Fe (II) is more and significantly absorbed by cells than Fe (III) (He et al 2008). The chemical structure is confirmed by ionization. Ionization of heavy metals can potentially be disruptive and dangerous to health and can even damage the vitality of the systems in a body (Abdi & Kazemi 2015).

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Microecosystem changes can be indicated by the diversity of microorganisms system because it can be related to variations in water characteristics (Ashraf et al 2011b), which can be determined through weather, geomorphologic, and geochemical conditions (Ashraf et al 2012b). Studies on the relationship between bacterial diversity and its role in the biogeochemical cycle and the interaction of microorganisms with their environment have been widely conveyed (Bhowal & Chakraborty 2015; Fashola et al 2015), including methane-oxidizing bacteria, ammonia-oxidizing bacteria (Sow et al 2014a; Sow et al 2014b), and arsenic-resistant bacteria (Jareonmit et al 2010; Valverde et al 2011). The study of the diversity of microorganisms and their activities as ecological bioindicatory agents is an important step to predict ecosystem conditions and environmental changes (Niemi & McDonald 2004; Moscatelli et al 2005). This is because microorganisms have the capacity to respond to changes that occur in the environment quickly, including in soil and aquatic ecosystems (Paerl et al 2003; Lau & Lennon 2012).

This short communication aimed to provide information related to the form of metal oxides, especially the heavy metals identified in post pit mining waters with different ages (chronosequences). Furthermore, it shortly reviews some relationships between metals and the diversity of acidophilic bacteria in acid mine waters and the potential of the waters for aquaculture.

Material and Method. The study was conducted by testing the metal content in water collected from under the tin mining post with different ages, namely less than 1 year (Station A), 5-10 years (Station B), and more than 15 years (Station C). Water sampling was carried out in pits located in Bangka Regency, Bangka Belitung Islands Province, Indonesia. The coordinates of the research station were: Station A - 01°59'S, 106°06' E; Station B - 01°59'S, 106°06'E; Station C - 01°55'S, 106°06'E (Figure 1) (Kurniawan 2019; Kurniawan et al 2019).

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how many repetitions were there?

how were the samples tested? with what method and with what tools/instruments?

was the water tested on site, or was it transported somewhere for analysis?

what containers were used for sampling?

what was the sample size?

from what depth was the water collected? near the shore/middle of the lakes?

you need to present some environmental conditions, because, as you said in the introduction, weather/geomorphological conditions affect bacteria distribution; so please present some methods and tools used for at least the following parameters: water depth, temperature, salinity, pH, dissolved oxygen (because it is important for aquaculture and bacterial life); and in Results and Discussion, please present the results of these measurements

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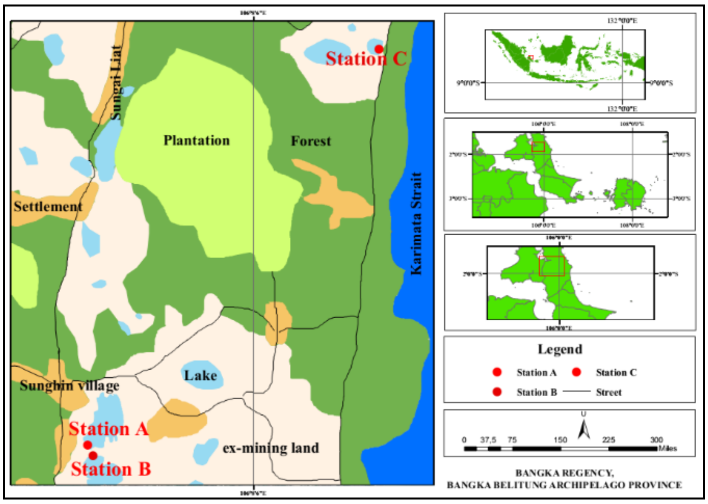

Figure 1. Research stations in ex-tin mining pits in Bangka Regency, Bangka Belitung Archipelago Province, Indonesia.

Analysis of the metal content was carried out using X-Ray Fluorescence (XRF) instruments Rigaku NEX CG (Kodom et al 2012) with cross-section specifications of 3 refracting metals, namely copper (Cu), molybdenum (Mo), and aluminum (Al). Data analysis was performed descriptively to explain the concentration of metal oxides identified in the samples.

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Results and Discussion. The results of the heavy metal analysis identified in previous studies the next metals: As, Co, Cr, Cu, Fe, Ga, Hf, Mn, Ni, Pb, Sn, Ta, Te, Th, V, and Zn (Kurniawan 2019; Kurniawan et al 2019) are presented in Table 1.

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Table 1

Concentrations of metal oxides in post pit mining waters

Metal form*	Metal oxide form	Name of oxide form	Average metal oxide concentration at the each station (ppm)		
			A	*B*	*C*
As	As_2O_3		5.70	8.49	3.04
Co	Co_2O_3		14.20	ND	9.34
Cr	Cr_2O_3		13.80	14.50	1.96
Cu	CuO		7.03	7.83	6.91
Fe	Fe_2O_3		1424.3	2307.67	1571.93
Ga	Ga_2O_3		11.50	12,00	12.65
Hf	HfO_2		7.74	8.76	11.07
Mn	MnO		33.70	35.10	39.05
Ni	NiO		10.40	7.32	4.64
Pb	PbO		14.50	13,00	9.40
Sn	SnO_2		89.73	64.77	74.53
Ta	Ta_2O_5		987.33	1373.33	888.73
Te	TeO_2		13,00	9.48	14.50
Th	ThO_2		10.50	9.97	15.75
V	V_2O_5		3,00	ND	2.38
Zn	ZnO		3.00	ND	ND

Note: * - sources: Kurniawan (2019); Kurniawan et al (2019); ND - not detected.

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The highest concentration was in the case of Fe_2O_3. Ta_2O_5, SnO_2, MnO were also concentrated in stations. The form of metal oxides can indicate or represent the metal content in these waters. The content of Fe_2O_3 has the highest value in waters under the age of 5-10 years. Fe metals were also highest in all voids compared to other metals.

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The highest MnO found in waters under the age of 15 years indicated that the potential for heavy metal contamination was still high. According to the values, not all metals experience a pattern of decreasing concentrations as the length of chronosequences in these waters. This condition was also observed in metals or oxides of gallium (Ga) as Ga_2O_3, hafnium (Hf) as HfO_2, manganese (Mn) as MnO, tellurium (Te) as TeO_2, and thorium (Th) as ThO_2. The decreasing pattern of metals or their oxide concentrations with the duration of chronosequence were observable for arsenic (As) as As_2O_3, chromium (Cr) as Cr_2O_3, copper (Cu) as CuO, nickel (Ni) as NiO, lead (Pb) as PbO, and zinc (Zn) as ZnO.

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A number of metals and their oxides contribute to water quality parameters, such as pH. In water under the age of 15 years, a low pH value (pH=3) occured, while water with age higher than 15 years had a neutral pH value (pH=7) (Kurniawan 2019; Kurniawan et al 2019). A number of metals indicated by the formation of the oxides, namely Fe_2O_3, As_2O_3, Cr_2O_3, CuO, NiO, PbO, and ZnO were found in high concentrations in waters less than 15 years old with acidic pH.

A number of minerals undergo chemical reactions that form acidic pH are categorized as potentially acid forming: Cu, Fe, Pb, Zn (Celebi & Oncel 2016). Al, As, Cd, Co, Cr, and Mn are formed from leaching of mined minerals (Campaner et al 2014). Oxidation and hydrolysis reactions of elements such as S, Fe, Cu, Zn, Ni, Cr, and Pb cause cation formations of Cu^{2+}, Zn^{2+}, Ni^{2+}, Cr^{3+}, and Pb^{2+}. The increase in proton H^+ contributes to the increase in acidity. An increasing number of H^+ ions can cause more acidic pH conditions in these environments (Gaikwad & Gupta 2008; Hatar et al 2013). The existence of these metals can directly or indirectly affect the pH value (De Saedeleer et al 2010; Zhao et al 2010; Fernandes et al 2011; Strom et al 2011; Huang et al 2012; Sadeghi et al 2012; Zhang et al 2014) and form acidity in mining waters known as acid mine drainage (Ashraf et al 2011b; Kurniawan 2019; Kolmert & Johnson 2001; Bigham & Nordstrom 2000; Tan et al 2007). The consequences of the acidity of waters include the disruption of the life of some organisms. Organisms at a microscopic level such as bacteria and archaea that are acidophilic have the ability to survive and live optimally in

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extreme acidic conditions as in acid mine waters (Navarro et al 2013). Acidophilic groups have the capacity to modify the physical and chemical conditions of waters by detoxifying or exploiting their metabolism [48] and play an important role in the biogeochemical cycles of iron and sulfur (Fashola et al 2015).

Acidophilic bacteria can be true acidophil (extreme acidophiles), which live at pH lower than 2.7, even lower than 1, with optimum growth under a pH of 3, and moderate acidophil that live in the pH range of 3-7.2, with optimum growth in pH of 3-5 (Johnson & Hallberg 2008; Mendez et al 2008; Oren 2010). Various studies explain that some bacteria found in mining areas are derived from the following phyla: Proteobacteria, Acidobacteria, Chloroflexi, Cyanobacteria, Actinobacteria, Nitrospirae, Firmicutes, Planctomycetes, Bacteroidetes, and Chlorobi (Gupta 2000; Lefebvre et al 2010; Hua et al 2015; Mesa et al 2017; Teng et al 2017; Cesario Fernandes et al 2018).

The optimum growth of acidophilic bacteria takes place between pH 1.0 and pH 5.0. This distinguishes it from neutrophilic bacteria that tolerates a pH range of 5-9, with the optimum pH of 7, and alkaliphilic bacteria, tolerant of 6-12 pH, with an optimum above 9, or often between 10 and pH 12 (Gupta et al 2014; Horikoshi 2016). Acidophilic bacterial groups have the ability to reduce and oxidize metals, sulfur and other minerals (Islam et al 2004; Hallberg 2010; Harahuc et al 2000; Yli-Hemminki et al 2014). They also help the carbon cycle flow (Wegner & Liesack 2017; Hausmann et al 2018; Sun et al 2018), are nitrogen retarders (Gargaud et al 2011; Sun et al 2015), and play a role in decomposing organic matter (Khare & Arora 2015). This capability is generally used in the process of detoxification of contaminated waters (Davis-Belmar & Norris 2009; Johnson et al 2009; Murali et al 2014; Shivlata & Satyanarayana 2015; Hu et al 2018).

The presence and biological activity of microbial groups in environments containing sulfide minerals can accelerate the formation of acidic conditions in the environment (Rawlings 2005). Acidic pH conditions involve oxidation processes and complex chemical reactions to produce H^+ ions, sulfates (SO_4^{2-}), Mn^{3+}, and other ions. More and more of these ions that form in an environment cause an increase in acidity (Gaikwad & Gupta 2008; Hatar et al 2013; Nurofiq et al 2016). The acceleration of the formation of acidic conditions can involve biological interactions such as microbial metabolic activity (Violante et al 2010).

The oxidation process of iron and sulfur in acidic waters produces an energy reaction which is used by acidophilic microbes for growth and metabolic functions. Sulfur oxidizing microbes utilize the iron cycle under acidic pH conditions for ferrous iron ions (Fe^{2+}) as electron donors and ferric iron ions (Fe^{3+}) as electron acceptors (Lei et al 2016). The oxidation process involving acidophilic bacteria occurs due to the ability of metabolism such as enzymes. Sulfur metabolic enzymes work to oxidize the element sulfur with sulfur dioxygenase, sulfur oxygenase reductase, and Hdr-like complex; thiosulfate oxidizing enzymes such as sulfuroxidizing enzyme and thiosulfate dehydrogenase; sulfide oxidizing enzymes such as sulfide:quinone oxidoreductase (Wang et al 2019) and iron oxidizing enzymes (Li et al 2017).

The presence and use of acidophilic bacteria in acid mine waters can be optimized by studying their biochemical characteristics. Individuals who have the potential to be bioremediators can be explored to tackle environmental pollution in acidic waters. This contributes as an effort to improve the condition of the aquatic environment so that it can be utilized for aquatic organisms in aquaculture.

Conclusions. The presence of a number of metals and their oxides in ex-tin mining or abandoned pit waters showed an association with the formation of pH conditions in the environment. The change in pH, especially acidic pH, causes a number of acidophilic groups to appear. Acidophilic groups have the ability to live in these extreme environments and then move biogeochemically. The potential ability of these acidophilic bacteria can be utilized to carry out detoxification processes for inorganic and organic contamination found in some waters. Optimizing the role of acidophilic bacteria is expected to be beneficial for environmental improvement, and even suitable for aquaculture.

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The metal oxides of abandoned tin mining pit waters as an indicator for bacterial diversity and aquaculture

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Abstract. This study aimed to provide information about the form of metal oxides of heavy metals that identified in abandoned tin mining pit waters with different ages and explain their relationship to the diversity of acidophilic bacteria and the prensence of fish in acid mine waters in Bangka Regency, Indonesia. The analysis of metal oxides was measured by X-Ray Fluorescence (XRF) and the bacterial was identified by Next Generation Sequencing (NGS). The sixteen oxide forms of heavy metals identified showed that Iron(III) Oxide (Fe_2O_3), Tantalum(V) Oxide (Ta_2O_5), Tin(IV) Oxide (SnO_2), and Manganese(II) Oxide (MnO) were found in high concentrations in all mine waters of different ages. The presence of heavy metals and their oxide affected the water quality, especially pH value and they made the acidic condition by oxidation process. This conditions contributed to the presence of acidophilic bacterial group such as Phylum Proteobacteria, Bacteriodetes, Planctomycetes, Actinobacteria, Chloroflexi, Firmicutes, Chlorobi, Acidobacteria, Cyanobacteria, etc. They play the important role in biogeochemical process and made a changes in this environment. The consequence of changes among chronosequence in this ecosystem can supported the life of organisms such as fish of *Aplocheilus* sp., *Rasbora* sp., *Betta* sp., *Puntius* sp., *Channa* sp., *Oreochromis* sp., *Belontia* sp., *Anabas* sp., and *Trichopodus* sp. Further, the presence of fish can produced organic material as product of their metabolism. The organic material was decomposed by bacterial to be anion and functional group which can reacted to proton and then caused neutralization of pH conditions.
Key Words: acidophilic bacterial, chronosequence, fish, heavy metals, interaction

Introduction. A number of metals have been identified in aquatic ecosystems, especially in artificial lakes (pits) after tin mining activities. These metals become components of ecosystem pollutants (Dinis & Fiuza 2011; Guan et al 2014; Kurniawan 2016), including as heavy metals such as Pb, Zn, Mn, Fe, Cr, Cu, Ni, Cd, Sn, and As (Ashraf et al 2011a; Henny 2011; Ashraf et al 2012a; Rosidah & Henny 2012; Daniel et al 2014). The use of the general term "heavy metal" refers to a group of metals and semi-metals (metalloids) associated with contamination, with a density higher than 3.5-5 g cm^{-3}, atomic weights between 63.546 (≈63.6) and 200.59 (≈200.6), and specific gravity higher than 4 (Duffus 2002; Srivastava & Majumder 2008; Aslam et al 2011).

Heavy metals are not always described as dangerous metals (toxic metals), but their chemical structure determines the biological properties and toxicity of these elements (Templeton 2015). Some metals in certain structures are needed by the organisms's body as essential microminerals (trace elements), but in other structures are dangerous (Kurniawan & Mustikasari 2019). Elements such as chromium in the form of Cr (III) are important trace elements, but Cr (VI) can cause cancer (Govind & Madhuri 2014), Hg (II) is more toxic than Hg (0) (Azimi & Moghaddam 2013), anorganic Mn (III) is more toxic than oxidation forms such as Mn (II) Cl_2 and Mn (IV) O_2; As (III) is more toxic than As (V); element V (V) is more toxic than V (IV) (Templeton 2015), and Fe (II) is more and significantly absorbed by cells than Fe (III) (He et al 2008). The chemical structure is confirmed by ionization. Ionization of heavy metals can potentially be disruptive and dangerous to health and can even damage the vitality of the systems in a body (Abdi & Kazemi 2015).

Microecosystem changes can be indicated by the diversity of microorganisms system because it can be related to variations in water characteristics (Ashraf et al 2011b), which can be determined through weather, geomorphologic, and geochemical conditions (Ashraf et al 2012b). Studies on the relationship between bacterial diversity and its role in the biogeochemical cycle and the interaction of microorganisms with their environment have been widely conveyed (Bhowal & Chakraborty 2015; Fashola et al 2015), including methane-oxidizing bacteria, ammonia-oxidizing bacteria (Sow et al 2014a; Sow et al 2014b), and arsenic-resistant bacteria (Jareonmit et al 2010; Valverde et al 2011). The study of the diversity of microorganisms and their activities as ecological bioindicatory agents is an important step to predict ecosystem conditions and environmental changes (Niemi & McDonald 2004; Moscatelli et al 2005). This is because microorganisms have the capacity to respond to changes that occur in the environment quickly, including in soil and aquatic ecosystems (Paerl et al 2003; Lau & Lennon 2012).

This short communication aimed to provide information related to the form of metal oxides, especially the heavy metals identified in post pit mining waters with different ages (chronosequences). Furthermore, it shortly reviews some relationships between metals and the diversity of acidophilic bacteria in acid mine waters and the potential of the waters for aquaculture.

Material and Method. The study was conducted by testing the metal content in water collected from under the tin mining post with different ages, namely Station A (age of pit < 1 year), Station B (age of pit between 5-10 years), and Station C (age of pit >15 years). The coordinates of the research station were Station A - 01°59'S, 106°06' E; Station B - 01°59'S, 106°06'E; Station C - 01°55'S, 106°06'E (Figure 1). Water sampling was carried out in these pits which located in Bangka Regency, Bangka Belitung Islands Province, Indonesia in 2017-2018.

There were four water samples as much as 1.5 L were collected from < 4 m and > 4 m in depth with five sampling points for repetitions from each stations. The collected water samples were put into sample bottle, transported by cold box container, and then analyzed ex situ in laboratory. Analysis of the metal content was carried out using X-Ray Fluorescence (XRF) instruments Rigaku NEX CG (Kodom et al 2012) with cross-section specifications of 3 refracting metals, namely copper (Cu), molybdenum (Mo), and aluminum (Al). While, the presence of bacterials was identified with Next Generation Sequencing (NGS). Then, data analysis was performed descriptively by Excel 2010 Program and Origin 8 Program to explain the concentration of metal oxides identified in the samples and their pattern in the research location.

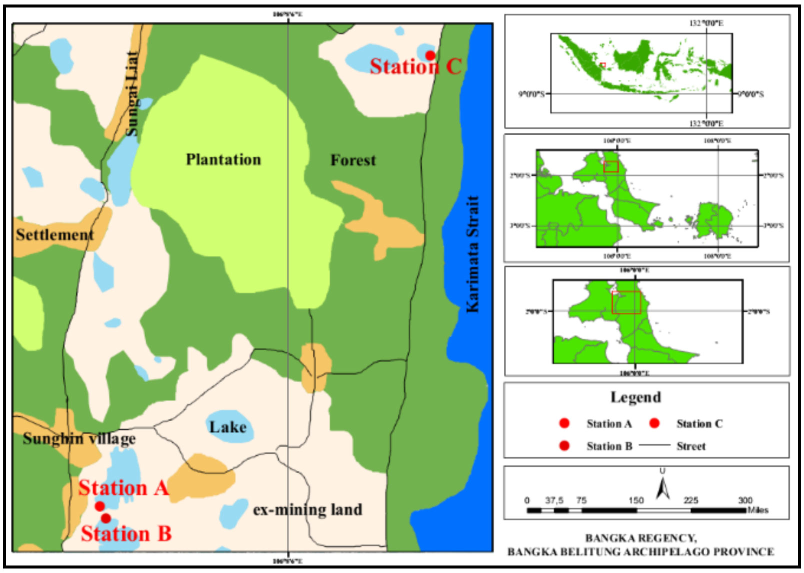

Figure 1. Research stations in ex-tin mining pits in Bangka Regency, Bangka Belitung Archipelago Province, Indonesia.

Results and Discussion. This reseach have identified some heavy metals from abandoned tin mining pits. There were sixteen heavy metals were identified, namely As, Co, Cr, Cu, Fe, Ga, Hf, Mn, Ni, Pb, Sn, Ta, Te, Th, V, and Zn. These heavy metals showed oxide form by XRF analysis as presented in Table 1. Metal oxide's functional properties are strongly dependent on oxide's crystal structure, composition, native defects, doping, etc., which govern their optical, electrical, chemical and mechanical characteristics (Grilli 2020).

Table 1

Concentrations of metal oxides in abandoned tin mining pits waters

Metal form	*Metal oxide form*	*Name of oxide form*	*Average metal oxide concentration in the each station (ppm)*		
			A	*B*	*C*
As	As_2O_3	Arsenic(III) Oxide	5.70	8.49	3.04
Co	Co_2O_3	Cobalt(III)) Oxide	14.20	ND	9.34
Cr	Cr_2O_3	Chromium(III) Oxide	13.80	14.50	1.96
Cu	CuO	Cupric(II) Oxide	7.03	7.83	6.91
Fe	Fe_2O_3	Iron(III) Oxide	1424.3	2307.67	1571.93
Ga	Ga_2O_3	Gallium(III) Oxide	11.50	12,00	12.65
Hf	HfO_2	Hafnium(IV) Oxide	7.74	8.76	11.07
Mn	MnO	Manganese(II) Oxide	33.70	35.10	39.05
Ni	NiO	Nickel(II) Oxide	10.40	7.32	4.64
Pb	PbO	Lead(II) Oxide	14.50	13,00	9.40
Sn	SnO_2	Tin(IV) Oxide	89.73	64.77	74.53
Ta	Ta_2O_5	Tantalum(V) Oxide	987.33	1373.33	888.73
Te	TeO_2	Tellurium(II) Oxide	13,00	9.48	14.50
Th	ThO_2	Thorium(II) Oxide	10.50	9.97	15.75
V	V_2O_5	Vanadium(V) Oxide	3,00	ND	2.38
Zn	ZnO	Zinc Oxide	3.00	ND	ND

Note: ND - not detected; Station A (age of pit < 1 year), Station B (age of pit between 5-10 years), and Station C (age of pit >15 years).

The oxide form and their concentrations (Tabel 1) indicated that there were potential of contamination of heavy metals in abandoned tin mining pit waters. The highest concentration found was Fe_2O_3 which identified at Station B with age of pit between 5-10 years and this oxide form was also found as the highest concentrations in all abandoned tin mining pit. The oxide form such as Ta_2O_5, SnO_2, MnO were also concentrated at all stations with high value, while the others showed low value.

The form of Fe_2O_3, Ta_2O_5, SnO_2, MnO also showed their distribution patterns in the waters. The Fe_2O_3 and Ta_2O_5 had increasing pattern of concentration in abandoned tin mining pit with age between 5-10 years compared age < 1 year and then the value were decrease in pit with age > 15 years. The SnO_2 had decreasing pattern of concentration in abandoned tin mining pit with age between 5-10 years compared age < 1 year and then the value were increase in pit with age > 15 years. While, the MnO showed the increasing pattern of concentration among the chronosequence of abandoned tin mining pit waters. The presence of these oxide indicated the potential for heavy metal contamination were still high, although the chronosequence of abandoned tin mining pits were more than 15 years. In additions of MnO, the other heavy metals also had high value in pit with age > 15 years, namely Ga_2O_3, HfO_2, TeO_2, and ThO_2. While, the other pattern of the oxide form was decreasing pattern that was shown by As_2O_3, Cr_2O_3, CuO, NiO, PbO, and ZnO. The value indicted concentration decreasing during chronosequence for more than 15 years.

The presence of heavy metals correlated with pH conditions. According to Kurniawan et al (2019) in the previous study in this reseach location, a number of metals and their oxides contribute to water quality parameters such as pH. In water under the age of 15 years, a low pH value (pH=3) occured, while water with age higher than 15 years had a neutral pH value (pH=7). A number of minerals undergo chemical reactions that form acidic pH are categorized as potentially acid forming: Cu, Fe, Pb, and Zn (Celebi & Oncel 2016). The acidic condition also can be fomed by association of Al, As, Cd, Co, Cr, and Mn with environment material (Campaner et al 2014). Oxidation and hydrolysis reactions of elements such as S, Fe, Cu, Zn, Ni, Cr, and Pb cause cation formations of Cu^{2+}, Zn^{2+}, Ni^{2+}, Cr^{3+}, and Pb^{2+}. The increase in proton H^+ contributes to the increase in acidity. An increasing number of H^+ ions can cause more acidic pH conditions in these environments (Gaikwad & Gupta 2008; Hatar et al 2013). The existence of these metals can directly or indirectly affect the pH value (De Saedeleer et al 2010; Zhao et al 2010; Fernandes et al 2011; Strom et al 2011; Huang et al 2012; Sadeghi et al 2012; Zhang et al 2014) and form acidity in mining waters known as acid mine drainage (Ashraf et al 2011b; Kurniawan 2019; Kolmert & Johnson 2001; Bigham & Nordstrom 2000; Tan et al 2007).

The consequences of the acidity of waters include the disruption of the life of some organisms. Organisms at a microscopic level such as bacteria and archaea that are acidophilic have the ability to survive and live optimally in extreme acidic conditions as in acid mine waters (Navarro et al 2013). Acidophilic groups have the capacity to modify the physical and chemical conditions of waters by detoxifying or exploiting their metabolism and play an important role in the biogeochemical cycles of iron and sulfur (Fashola et al 2015). Acidophilic bacteria can be the true acidophil (extreme acidophiles), which live at pH lower than 2.7, even lower than 1, with optimum growth under a pH of 3, and the moderate acidophil that live in the pH range of 3-7.2, with optimum growth in pH of 3-5 (Johnson & Hallberg 2008; Mendez et al 2008; Oren 2010). Various studies explain that some bacteria found in mining areas are derived from the following phyla: Proteobacteria, Acidobacteria, Chloroflexi, Cyanobacteria, Actinobacteria, Nitrospirae, Firmicutes, Planctomycetes, Bacteroidetes, and Chlorobi (Gupta 2000; Lefebvre et al 2010; Hua et al 2015; Mesa et al 2017; Teng et al 2017; Fernandes et al 2018). In these research locations, the presence of bacterial was identified and was shown in Table 2.

Some species of these phylum can be grouped as acidophilic bacterial and they have the ability to reduce and oxidize metals, sulfur and other minerals (Islam et al 2004; Hallberg 2010; Harahuc et al 2000; Yli-Hemminki et al 2014). They also help the carbon cycle flow (Wegner & Liesack 2017; Hausmann et al 2018; Sun et al 2018), are nitrogen retarders (Gargaud et al 2011; Sun et al 2015), and play a role in decomposing organic matter (Khare & Arora 2015). This capability is generally used in the process of detoxification of contaminated waters (Davis-Belmar & Norris 2009; Johnson et al 2009; Murali et al 2014; Shivlata & Satyanarayana 2015; Hu et al 2018).

The presence and biological activity of microbial groups in environments containing sulfide minerals can accelerate the formation of acidic conditions in the environment (Rawlings 2005). Acidic pH conditions involve oxidation processes and complex chemical reactions to produce H^+ ions, sulfates (SO_4^{2-}), Mn^{3+}, and other ions. More and more of these ions that form in an environment cause an increase in acidity (Gaikwad & Gupta 2008; Hatar et al 2013; Nurofiq et al 2016). The acceleration of the formation of acidic conditions can involve biological interactions such as microbial metabolic activity (Violante et al 2010).

The oxidation process of iron and sulfur in acidic waters produces an energy reaction which is used by acidophilic microbes for growth and metabolic functions. The acidophilic bacterial use sulfur metabolic enzymes to oxidize the element sulfur with sulfur dioxygenase, sulfur oxygenase reductase, and Hdr-like complex; thiosulfate oxidizing enzymes such as sulfuroxidizing enzyme and thiosulfate dehydrogenase; sulfide oxidizing enzymes such as sulfide:quinone oxidoreductase (Wang et al 2019) and iron oxidizing enzymes (Li et al 2017) to utilize the iron cycle under acidic pH conditions for ferrous iron ions (Fe^{2+}) as electron donors and ferric iron ions (Fe^{3+}) as electron acceptors (Lei et al 2016).

The presence of acidophilic bacterial in low pH value due to its capability to survive and use sulfide minerals such as sulfur and iron as their energy source for growth by oxidation process (Korehi et al 2013) such as $S^0 + O_2 + H_2O \rightarrow SO_4^{2-} + 2H^+$ or Fe^{2+} +

O_2 + $2H^+$ → Fe^{3+} + H_2O (Rawlings 2005). The relative abundance of them in long chronosequence can also contribute to decompose organic material such as water plants, metabolism products of water organisms, and the dead organisms. The decomposting process can affect to CO_2 increasing and then interact to H_2O to form carbonic acid (H_2CO_3) by reaction $CO_2 + H_2O \rightarrow H_2CO_3 \rightarrow H^+ + HCO_3^-$ (Loerting & Bernard 2010; Ghoshal & Hazra 2015). Dissociation of H_2CO_3 to ion carbonate (HCO_3^-) can neutralize ion hydrogen (H^+) so it can increase the pH value to neutral condition (Andersen 2010). The decomposition product of organic acid from organic materials have functional group R-COOH as dissociated organic anion and it can use cation H^+ so it can make the concentration of ion H^+ decrease in environment and pH reach neutral condition (Rukshana et al 2010).

Table 2

The presence of bacterial in abandoned tin mining pits

No	*Phylum*	*The presence of phylum*		
		Station A	*Station B*	*Station C*
1	Proteobacteria	+	+	+
2	Actinobacteria	+	+	+
3	Chloroflexi	+	+	+
4	Firmicutes	+	+	+
5	Acidobacteria	+	+	+
6	Planctomycetes	+	+	+
7	Bacteroidetes	+	+	+
8	Chlorobi	+	+	+
9	Cyanobacteria	+	+	+
10	Gemmatimonadetes	+	+	+
11	OD1 (candidate of Phylum Parcubacteria)	+	+	+
12	Spirochaetes	+	+	+
13	Thermi	-	+	+
14	Nitrospirae	+	+	+
15	Verrucomicrobia	+	+	+
16	Armatimonadetes	+	-	+
17	Chlamydiae	+	+	+
18	Elusimicrobia	+	-	+
19	Caldiserica	-	-	+
20	Chaldithrix	-	-	+
21	Lentisphaerae	-	-	+
22	Fibrobacteres	-	-	+

The presence of heavy metals and their oxide and also the pH change in environment can impact to life of macroorganism such as fish. There were some fishes found in the abandoned tin mining pit (Table 3). The presence of fish in abandoned tin mining pit indicated some species of fish can survive in the environment. It also can explained that the species can be introduced as aquaculture organisms in an extreme habitat such as abandoned mining waters. The capability of the species can be used to help the succession in the habitat by producing organic substance from their metabolism. Their metabolism product were used by decompose bacterial such as Phylum Bacteriodetes to produce CO_2 and carboxylate functional group ($COOH^-$) for recovery pH value to neutral condition (Andersen 2010; Loerting & Bernard 2010; Rukshana et al 2010; Ghoshal & Hazra 2015).

In addition, the presence of CO_2 in water can be changed into complex organic molecule and oxygen (O_2) in aerobic photosynthesis by reaction $H_2O + CO_2 \rightarrow CH_2O + O_2$ (Johnson, 2016). The presence of organic molecule can be used as nutrition for organism's life and the optimum O_2 also can supported their life. The CO_2 also can be used as carbon source for some bacterials for anaerobic photosynthesis such as Green

Sulfur Bacteria (GSB) like Chlorobium, Green Non-Sulfur Bacteria (GNSB) like Chloroflexus, Purple Sulfur Bacteria (PSB) like Thiospirillum, and Purple Non-Sulfur Bacteria (PNSB) like Rhodobacter. The anorganic photosynthesis use H_2S, H_2, and S as electron donor by reaction $6CO_2 + 12H_2S \rightarrow C_6H_{12}O_6 + 6H_2O + 12S^0$ (Nisbet et al 2003). The organic materials as product of this reaction was used as nutrition in an environment and can recovered the water quality.

Table 3

The presence of fish in abandoned tin mining pits

No	*Genus*	*The presence of fish*		
		Station A	*Station B*	*Station C*
1	*Aplocheilus* sp.	-	-	+
2	*Rasbora* sp.	+	+	+
3	*Betta* sp.	+	+	+
4	*Puntius* sp.	+	+	-
5	*Channa* sp.	+	+	+
6	*Oreochromis* sp.	-	+	-
7	*Belontia* sp.	+	+	+
8	*Anabas* sp.	+	+	-
9	*Trichopodus* sp.	+	+	+

The information of environment condition form abandoned tin mining pits showed there was an interaction between the presence heavy metals and their oxide, pH value, acidophilic bacterial and fish's life. The capability of acidophilic bacterial and some fish in acid mine waters can be optimized by studying of their biochemical characteristics as an effort to improve the condition of the aquatic environment.

Conclusions. The presence of a number of metals and their oxides in ex-tin mining or abandoned tin mining pit waters showed an association with the formation of pH conditions in the environment. The change in pH, especially acidic pH causes the presence a number of acidophilic groups. Acidophilic groups have the ability to live in this extreme environment and then move biogeochemically. The potential ability of these acidophilic bacterial can be utilized to carry out detoxification processes for inorganic and organic contamination found in some waters. The changes of environment quality can impacted to the other organism's life such as fish. Further, the presence fish can produced organic materials as product of their metabolism and it can supported biogeochemical process for chronosequence in this environment.

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The metal oxides of abandoned tin mining pit waters as an indicator for bacterial diversity

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Abstract. This study aimed to provide information about the form of metal oxides of heavy metals identified in abandoned tin mining pit waters with different ages and explain their relationship to the diversity of acidophilic bacteria and the presence of fish in acid mine waters in Bangka Regency, Indonesia. The analysis of metal oxides was carried out using X-Ray Fluorescence (XRF). The 16 oxide forms of heavy metals identified showed that iron (III) oxide (Fe_2O_3), tantalum (V) oxide (Ta_2O_5), tin (IV) oxide (SnO_2), manganese (II) oxide (MnO) were found in high concentrations in all mine waters of different ages. The presence of heavy metals and their oxides affected the water quality, especially the pH value, decreasing it by oxidation processes. This condition contributed to the presence of acidophilic bacterial groups such as phyla Proteobacteria, Bacteriodetes, Planctomycetes, Actinobacteria, Chloroflexi, Firmicutes, Chlorobi, Acidobacteria, Cyanobacteria, etc. They play the important role in biogeochemical processes, changing the environment. The changes of chronosequences in the ecosystem can support the life of organisms such as fish (*Aplocheilus* sp., *Rasbora* sp., *Betta* sp., *Puntius* sp., *Channa* sp., *Oreochromis* sp., *Belontia* sp., *Anabas* sp., and *Trichopodus* sp.). Furthermore, the fish produce organic material. The organic material was decomposed by bacteria in anions and functional groups, which react to protons and cause the neutralization pH.

Key Words: acidophilic bacteria, chronosequence, fish, heavy metals, interaction.

Introduction. A number of metals have been identified in aquatic ecosystems, especially in artificial lakes (pits) after tin mining activities. These metals become components of ecosystem pollutants (Dinis & Fiuza 2011; Guan et al 2014; Kurniawan 2016), including heavy metals such as Pb, Zn, Mn, Fe, Cr, Cu, Ni, Cd, Sn, and As (Ashraf et al 2011a; Henny 2011; Ashraf et al 2012a; Rosidah & Henny 2012; Daniel et al 2014). The general term "heavy metal" refers to a group of metals and semi-metals (metalloids) associated with contamination, with a density higher than 3.5-5 g cm^{-3}, with atomic weights between 63.546 (≈63.6) and 200.59 (≈200.6), and specific gravity higher than 4 (Duffus 2002; Srivastava & Majumder 2008; Aslam et al 2011).

Heavy metals are not always described as dangerous metals (toxic metals), but their chemical structure determines the biological properties and toxicity of these elements (Templeton 2015). Some metals in certain structures are needed by the body of an organism as essential microminerals (trace elements), but can be dangerous in other structures (Kurniawan & Mustikasari 2019). Elements such as chromium in the form of Cr (III) are important trace elements, but Cr (VI) can cause cancer (Govind & Madhuri 2014). Hg (II) is more toxic than Hg (0) (Azimi & Moghaddam 2013), inorganic Mn (III) is more toxic than oxidation forms such as Mn (II) Cl_2 and Mn (IV) O_2; As (III) is more toxic than As (V); element V (V) is more toxic than V (IV) (Templeton 2015), and Fe (II) is more significantly absorbed by cells than Fe (III) (He et al 2008). The chemical structure is confirmed by ionization. Ionization of heavy metals can potentially be disruptive and dangerous to health and can even damage the vitality of the systems in a body (Abdi & Kazemi 2015).

Microecosystem changes can be indicated by the diversity of microorganisms system because it can be related to variations in water characteristics (Ashraf et al 2011b), which can be determined through weather, geomorphologic, and geochemical conditions (Ashraf et al 2012b). Studies on the relationship between bacterial diversity and its role in the biogeochemical cycle and the interaction of microorganisms with their environment have been widely conveyed (Bhowal & Chakraborty 2015; Fashola et al 2015), including methane-oxidizing bacteria, ammonia-oxidizing bacteria (Sow et al 2014a; Sow et al 2014b), and arsenic-resistant bacteria (Jareonmit et al 2010; Valverde et al 2011). The study of the diversity of microorganisms and their activities as ecological bioindicators is an important step in predicting ecosystem conditions and environmental changes (Niemi & McDonald 2004; Moscatelli et al 2005). This is because microorganisms have the capacity to respond to changes that occur in the environment quickly, including in soil and aquatic ecosystems (Paerl et al 2003; Lau & Lennon 2012).

This short communication aimed to provide information related to the form of metal oxides, especially the heavy metals identified in post mining pit waters with different ages (chronosequences). Furthermore, it shortly reviews some relationships between metals and the diversity of acidophilic bacteria in acid mine waters and the potential of the waters for aquaculture.

Material and Method. The study was conducted by testing the metal content in water collected from under the tin mining post with different ages, namely less than 1 year (Station A), 5-10 years (Station B), and more than 15 years (Station C). The coordinates of the research station were: Station A - 01°59'S, 106°06' E; Station B - 01°59'S, 106°06'E; Station C - 01°55'S, 106°06'E (Figure 1) (Kurniawan 2019; Kurniawan et al 2019). Water sampling was carried out in these pits located in Bangka Regency, Bangka Belitung Islands Province, Indonesia, in 2017-2018.

There were 4 water samples, 1.5 L each, collected from depths lower than 4 m and higher than 4 m, with five sampling points for repetitions in each station. The collected water samples were placed into sample containers, transported in a cool box, and analyzed in a laboratory. Analysis of the metal content was carried out using X-Ray Fluorescence (XRF) instruments Rigaku NEX CG (Kodom et al 2012), with cross-section specifications of 3 refracting metals, namely copper (Cu), molybdenum (Mo), and aluminum (Al). The presence of bacteria was identified with Next Generation Sequencing (NGS). Data analysis was performed descriptively with Microsoft Excel 2010 and Origin 8 to explain the concentration of metal oxides identified in the samples and their pattern in the research location.

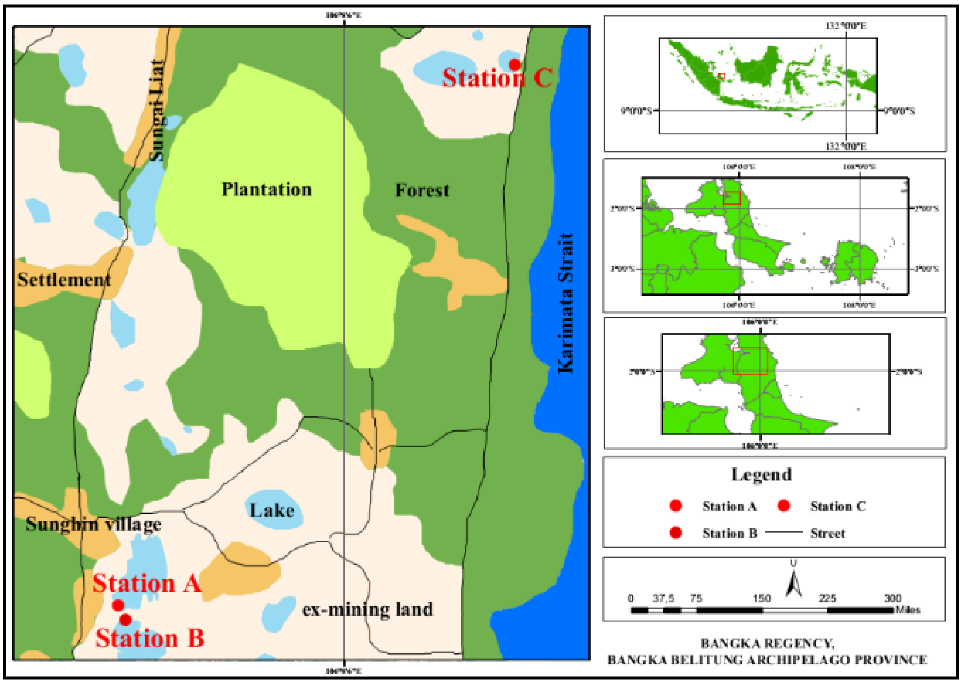

Figure 1. Research stations in ex-tin mining pits in Bangka Regency, Bangka Belitung Archipelago Province, Indonesia.

Results and Discussion. This reseach has identified some heavy metals from abandoned tin mining pits. There were 16 heavy metals were identified: As, Co, Cr, Cu, Fe, Ga, Hf, Mn, Ni, Pb, Sn, Ta, Te, Th, V, and Zn. These heavy metals showed oxide forms by XRF analysis (Table 1). The functional properties of metal oxides are strongly dependent on the crystal structure of the oxide, composition, native defects, doping, etc., which govern their optical, electrical, chemical and mechanical characteristics (Grilli 2020).

Table 1

Concentrations of metal oxides in abandoned tin mining pit waters

Metal form	*Metal oxide form*	*Name of oxide form*	*Average metal oxide concentration in the each station (ppm)*		
			A	*B*	*C*
As	As_2O_3	Arsenic (III) oxide	5.7	8.49	3.04
Co	Co_2O_3	Cobalt (III)) oxide	14.2	ND	9.34
Cr	Cr_2O_3	Chromium (III) oxide	13.8	14.5	1.96
Cu	CuO	Cupric (II) oxide	7.03	7.83	6.91
Fe	Fe_2O_3	Iron (III) oxide	1424.3	2307.67	1571.93
Ga	Ga_2O_3	Gallium (III) oxide	11.5	12	12.65
Hf	HfO_2	Hafnium (IV) oxide	7.74	8.76	11.07
Mn	MnO	Manganese (II) oxide	33.7	35.1	39.05
Ni	NiO	Nickel (II) oxide	10.4	7.32	4.64
Pb	PbO	Lead (II) oxide	14.5	13	9.4
Sn	SnO_2	Tin (IV) oxide	89.73	64.77	74.53
Ta	Ta_2O_5	Tantalum (V) oxide	987.33	1373.33	888.73
Te	TeO_2	Tellurium (II) oxide	13	9.48	14.5
Th	ThO_2	Thorium (II) oxide	10.5	9.97	15.75
V	V_2O_5	Vanadium (V) oxide	3	ND	2.38
Zn	ZnO	Zinc oxide	3	ND	ND

Note: ND - not detected; Station A (age of pit < 1 year), Station B (age of pit between 5-10 years), and Station C (age of pit > 15 years).

The form of oxides and their concentrations (Table 1) indicated that there was potential of contamination with heavy metals in abandoned tin mining pit waters. The highest concentration found was Fe_2O_3, which was identified at Station B. This oxide form was also found with the highest concentration in all abandoned tin mining pits. Other oxide forms such as Ta_2O_5, SnO_2, and MnO also presented high concentrations in all stations, while others had low values.

Fe_2O_3, Ta_2O_5, SnO_2, MnO also showed their distribution patterns in the waters. Fe_2O_3 and Ta_2O_5 had increasing patterns of concentration in abandoned tin mining pit waters with age between 5-10 years compared those with age less than 1 year. The values decreased in pits with ages above 15 years. SnO_2 had a decreasing pattern of concentration in abandoned tin mining pit waters with age between 5-10 years compared to those with ages less than 1 year, and the values increased in pits with ages above 15 years. MnO showed an increasing pattern of concentration among the chronosequence of abandoned tin mining pit waters. The presence of these oxides indicates a high potential for heavy metal contamination, although the chronosequences of abandoned tin mining pits were more than 15 years. In addition, other heavy metals also had high values in pits with ages above 15 years, namely Ga_2O_3, HfO_2, TeO_2, and ThO_2. As_2O_3, Cr_2O_3, CuO, NiO, PbO, and ZnO had decreasing concentration patterns. The values indicate that the concentrations decrease during chronosequences above 15 years.

The presence of heavy metals is correlated with pH conditions. According to Kurniawan et al (2019) in a previous study in this research location, a number of metals and their oxides contribute to water quality parameters such as pH. In water with an age below 15 years, a low pH value (3) occurred, while water with an age above 15 years had a neutral pH value (7). A number of minerals undergoing chemical reactions that form acidic pH are categorized as potentially acid forming: Cu, Fe, Pb, and Zn (Celebi & Oncel 2016). The acidic condition also can be formed by associations of Al, As, Cd, Co, Cr, and Mn with environment material (Campaner et al 2014). Oxidation and hydrolysis reactions of elements such as S, Fe, Cu, Zn, Ni, Cr, and Pb cause cation formations of Cu^{2+}, Zn^{2+}, Ni^{2+}, Cr^{3+}, and Pb^{2+}. The increase in proton H^+ contributes to the increase in acidity. An increasing number of H^+ ions can cause more acidic pH conditions in these environments (Gaikwad & Gupta 2008; Hatar et al 2013). The existence of these metals can directly or indirectly affect the pH value (De Saedeleer et al 2010; Zhao et al 2010; Fernandes et al 2011; Strom et al 2011; Huang et al 2012; Sadeghi et al 2012; Zhang et al 2014) and form acidity in mining waters known as acid mine drainage (Bigham & Nordstrom 2000; Kolmert & Johnson 2001; Tan et al 2007; Ashraf et al 2011b; Kurniawan 2019).

The consequences of the acidity of waters include the disruption of the life of some organisms. Organisms at a microscopic level such as bacteria and archaea that are acidophilic have the ability to survive and live optimally in extreme acidic conditions, including in acid mine waters (Navarro et al 2013). Acidophilic groups have the capacity to modify the physical and chemical conditions of waters by detoxifying or exploiting their metabolism and play an important role in the biogeochemical cycles of iron and sulfur (Fashola et al 2015). Acidophilic bacteria can be true acidophil (extreme acidophiles), which live at pH lower than 2.7, even lower than 1, with optimum growth under a pH of 3, and moderate acidophil that live in a pH range of 3-7.2, with optimum growth in pH of 3-5 (Johnson & Hallberg 2008; Mendez et al 2008; Oren 2010). Various studies explain that some bacteria found in mining areas are derived from the following phyla: Proteobacteria, Acidobacteria, Chloroflexi, Cyanobacteria, Actinobacteria, Nitrospirae, Firmicutes, Planctomycetes, Bacteroidetes, and Chlorobi (Gupta 2000; Lefebvre et al 2010; Hua et al 2015; Mesa et al 2017; Teng et al 2017; Cesario Fernandes et al 2018). In these research locations, the presence of bacteria was identified and presented in Table 2.

Some species of these phylum can be grouped as acidophilic bacteria and they have the ability to reduce and oxidize metals, sulfur and other minerals (Islam et al 2004; Hallberg 2010; Harahuc et al 2000; Yli-Hemminki et al 2014). They also help the carbon cycle flow (Wegner & Liesack 2017; Hausmann et al 2018; Sun et al 2018), are nitrogen retarders (Gargaud et al 2011; Sun et al 2015), and play a role in decomposing organic matter (Khare & Arora 2015). This capability is generally used in the process of detoxification of contaminated waters (Davis-Belmar & Norris 2009; Johnson et al 2009; Murali et al 2014; Shivlata & Satyanarayana 2015; Hu et al 2018).

The presence and biological activity of microbial groups in environments containing sulfide minerals can accelerate the formation of acidic conditions in the environment (Rawlings 2005). Acidic pH conditions involve oxidation processes and complex chemical reactions to produce H^+ ions, sulfates (SO_4^{2-}), Mn^{3+}, and other ions. More and more of these ions that form in an environment cause an increase in acidity (Gaikwad & Gupta 2008; Hatar et al 2013; Nurofiq et al 2016). The acceleration of the formation of acidic conditions can involve biological interactions such as microbial metabolic activity (Violante et al 2010).

The oxidation processes of iron and sulfur in acidic waters produce an energy reaction used by acidophilic microbes for growth and metabolic functions. The acidophilic bacteria use sulfur metabolic enzymes to oxidize sulfur (sulfur dioxygenase, sulfur oxygenase reductase, and Hdr-like complex). They also use thiosulfate oxidizing enzymes such as sulfuroxidizing enzyme and thiosulfate dehydrogenase, sulfide oxidizing enzymes such as sulfide quinone oxidoreductase (Wang et al 2019). They present iron oxidizing enzymes (Li et al 2017) to utilize the iron cycle under acidic pH conditions for ferrous iron ions (Fe^{2+}) as electron donors and ferric iron ions (Fe^{3+}) as electron acceptors (Lei et al 2016).

The presence of acidophilic bacteria in low pH values occurs due to its capability to survive and use sulfide minerals such as sulfur and iron as their energy source for growth by oxidation processes (Korehi et al 2013). Some examples are $S^0 + O_2 + H_2O \rightarrow SO_4^{2-} + 2H^+$ or $Fe^{2+} + O_2 + 2H^+ \rightarrow Fe^{3+} + H_2O$ (Rawlings 2005). The relative abundance of acidophilic bacteria in long chronosequences can contribute to the decomposition of organic material such as water plants, metabolism products of water organisms, and dead organisms. The decomposing processes can increase CO_2 levels, which can interact with H_2O to form carbonic acid (H_2CO_3) by the following reaction $CO_2 + H_2O \rightarrow H_2CO_3 \rightarrow H^+ + HCO_3^-$ (Loerting & Bernard 2010; Ghoshal & Hazra 2015). The dissociation of H_2CO_3 to ion carbonate (HCO_3^-) can neutralize ion hydrogens (H^+) so it can increase the pH value to neutral (Andersen 2002). The decomposition product of organic acid from organic materials has the functional group R-COOH as dissociated organic anion and it can use H^+ cations to make the concentration of H^+ ions decrease in environment, and the pH to reach neutral condition (Rukshana et al 2010).

Table 2

The presence of bacteria in abandoned tin mining pits

No	*Phylum*	*The presence of phylum*		
		Station A	*Station B*	*Station C*
1	Proteobacteria	+	+	+
2	Actinobacteria	+	+	+
3	Chloroflexi	+	+	+
4	Firmicutes	+	+	+
5	Acidobacteria	+	+	+
6	Planctomycetes	+	+	+
7	Bacteroidetes	+	+	+
8	Chlorobi	+	+	+
9	Cyanobacteria	+	+	+
10	Gemmatimonadetes	+	+	+
11	OD1 (candidate of Phylum Parcubacteria)	+	+	+
12	Spirochaetes	+	+	+
13	Thermi	-	+	+
14	Nitrospirae	+	+	+
15	Verrucomicrobia	+	+	+
16	Armatimonadetes	+	-	+
17	Chlamydiae	+	+	+
18	Elusimicrobia	+	-	+
19	Caldiserica	-	-	+
20	Chaldithrix	-	-	+
21	Lentisphaerae	-	-	+
22	Fibrobacteres	-	-	+

The presence of heavy metals and their oxide, and the pH change in environment can impact to life of macroorganisms such as fish. There were some fish found in the abandoned tin mining pits (Table 3). The presence of fish in abandoned tin mining pits indicated that some species of fish can survive in the environment. Their metabolism products are decomposed by Bacteriodetes to produce CO_2 and carboxylate functional groups ($COOH^-$), bringing the pH values to neutral (Andersen 2002; Loerting & Bernard 2010; Rukshana et al 2010; Ghoshal & Hazra 2015).

In addition, the presence of CO_2 in the water can be changed into complex organic molecules and oxygen (O_2) in aerobic photosynthesis by the following reaction $H_2O + CO_2 \rightarrow CH_2O + O_2$ (Johnson 2016). The organic molecules can be feed for organisms and the optimum O_2 also can supported their life. The CO_2 can also be used as a carbon source for some bacteria in anaerobic photosynthesis, such as Green Sulfur Bacteria like *Chlorobium*, Green Non-Sulfur Bacteria like *Chloroflexus*, Purple Sulfur Bacteria like *Thiospirillum*, and Purple Non-Sulfur Bacteria like *Rhodobacter*. The anorganic photosynthesis uses H_2S, H_2, and S as electron donors with the following reaction: $6CO_2 + 12H_2S \rightarrow C_6H_{12}O_6 + 6H_2O + 12S$ (Nisbet & Fowler 2003). The organic materials resulting from this reaction are used as nutrients in an environment and can improve water quality.

Table 3
The presence of fish in abandoned tin mining pits

No	Genus	The presence of fish		
		Station A	Station B	Station C
1	*Aplocheilus* sp.	-	-	+
2	*Rasbora* sp.	+	+	+
3	*Betta* sp.	+	+	+
4	*Puntius* sp.	+	+	-
5	*Channa* sp.	+	+	+
6	*Oreochromis* sp.	-	+	-
7	*Belontia* sp.	+	+	+
8	*Anabas* sp.	+	+	-
9	*Trichopodus* sp.	+	+	+

The information about the environmental conditions from abandoned tin mining pits showed there was an interaction between the presence heavy metals and their oxides, pH values, acidophilic bacteria and fish life. The capability of acidophilic bacteria and some fish in acid mine waters can be optimized by studying their biochemical characteristics as an effort to improve the condition of the aquatic environment.

Conclusions. The presence of a number of metals and their oxides in ex-tin mining or abandoned tin mining pit waters showed an association with the pH in the environment. The change in pH, especially acidic pH causes the presence of a number of acidophilic groups. Acidophilic groups have the ability to live in this extreme environment and then move biogeochemically. The potential ability of these acidophilic bacteria can be utilized to carry out detoxification processes for inorganic and organic contamination found in some waters. The changes of environment quality can impact organism life such as fish. Furthermore, the presence of fish can produce organic materials as a product of their metabolism and it can support biogeochemical processes for chronosequences in this environment.

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