

Preliminary Test of Fish Respiratory and Locomotive Signal Using Multispecies Freshwater Bio indicator (MFB)

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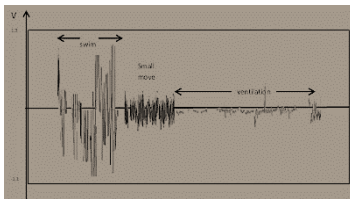
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Graphical abstract



Abstract

Fish produces many types of behavior as response to stress that cause by pollution. Respiration and locomotion are two main responses that normally produced. As such, study on these responses is important especially for pollution monitoring. Study on fish respiratory and locomotive behaviors was undertaken using multispecies freshwater bio indicator (MFB). Signal produced by fish determines the specific frequency range for respiratory and locomotive activities. This study aims to produce unstressed and stress signals (ventilation and locomotion) as a respond to TSS contamination. Three common species namely guppy (*Poecilia reticulata*), fighting fish (*Trichopsis vittatus*) and Malaysian masher (*Tor tambroides*) were used and test was conducted for 24-hour period. Result of the study indicates that ventilation and locomotion activities were clearly separated by different wavelength for all species but each species produced similar wavelength for each activity. A paired t-test confirmed that wavelength for each activity from all species was not differ significantly ($p > 0.05$, $\alpha = 0.05$). Only ventilation produce significant respond to TSS load and ventilation percentage signal was increase as TSS concentration increase. Similar respond was observed for all species. This study demonstrates that TSS contamination can be detected at early stage and maximum TSS load into the river system could be estimated.

Keywords: Freshwater pollution; Total suspended solids; Biological indicator; Stress signal, Biological monitoring

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1.0 INTRODUCTION

Water is a fundamental constituent of life and is essential to a wide range of economic activities. It is also a limited resource, as we have to take an action to preserve and continual monitor. Water pollution is caused by point and non-point sources. Point sources include sewage treatment plants, manufacturing and agro-based industries and animal farms. Non-point sources are defined as diffused sources such as agricultural activities and surface runoffs. In 2006, the Malaysian Department of Environment (DOE) registered 18,956 water pollution point sources comprising mainly sewage treatment plants (48% inclusive of 601 Network Pump Stations), manufacturing industries (45%), animal farms (5%) and agro-based industries (2%). There are 1,064 monitoring stations in 143 river basin monitored by DOE with status 60% were found to be clean, 35% slightly polluted and 5% polluted. The most major pollutant detected was biochemical oxygen demand (BOD5) from untreated or partially treated sewage and ammoniacal nitrogen (NH3-N) from agro-based and manufacturing industries. The main sources from domestic sewage, livestock farming, earthworks and land clearing activities is suspended solids (TSS) [1].

Total suspended solids (TSS) are fine matters in water that can be trapped by a filter. Total suspended solids can include wide variety of material, such as silt, decaying plant and animal matter, industrial wastes, and sewage. Suspended sediment consists primarily of silt and clay-size particles that may be rapidly transported downstream and locally deposited on floodplains and overbank storage locations or may infiltrate into gravel interstices of the bed [5]. High concentrations of suspended solids cause various kind of problems for stream health and aquatic life [6]. The decrease in water clarity caused by TSS can affect the ability of fish vision reduce feeding capability. Suspended sediment is associated with negative effects on the spawning, growth, and reproduction of salmonids [7, 8]. Suspended sediment reported could clog fish [9], reduce growth rates [10, 11], decrease resistance to disease, and prevent egg and larval development. Precipitate suspended solids at the bottom of a water body, could smother the fish eggs and aquatic insects, as well as suffocate newly hatched insect larvae. Settling sediments can fill in spaces between rocks which could have been used by aquatic organisms for inhabitant.

The suspended solid is the main pollutant released into the water body after any land clearance for developments. Without good management practice TSS could overloaded and harm

aquatic population especially fish. Many studies proves impacts of TSS on fishes, but mostly from Europe Country and in Malaysia, no information on TSS toxic concentration been published. As such, study on determining minimum level of TSS that could effects fishes in in freshwater is crucial. This could avoid fish stress and fish kill especially endangered species. The study was conducted to produce baseline wavelength (Hz) for ventilation and locomotion of local fishes and this benchmark will be used to determine minimum TSS level that initiate fish stress.

2.0 MATERIALS AND METHODS

Fish samples were collected from nearby streams and acclimated in the aquarium in the laboratory. Three fish species collected were guppy fish (*Poecilia reticulata*), fighting fish (*Trichopsis vittatus*) and Malaysian masher (*Tor tambroides*). Pre-mature fish were collected and used in the experiment since only small size fish can be placed in testing chamber. The multispecies freshwater biomonitor, MFB (Limco International, Germany) with eight measurement chambers was used for ventilation and locomotion test. This hardware is connected to the computer and ventilation and locomotion signal is detected by quadrapole that fixed in the chamber. The signal is recorded and plotted in electricity flows and then translated into histogram frequency (Hz) by fast furrier transformation (FFT) by MFB software [2]. The MFB allows for the investigation of behavioural and closely related physiological changes due to alterations in water quality [3]. Major advantages of such system include greater sensitivity relative to chemical indicators, their non-destructive nature, their ecological relevance and ability to perform long-term biomonitoring [4]. The chamber of MFB was placed inside the folded aquarium with aluminium foil to reduce noise level during the experiment. For the best observation of the fish responses, a digital camera was placed in front of the aquarium to record fish activities inside the chamber during the test. Quadrapole impedance conversion technique was applied. Movements of each fish sample that was placed in a cylindrical test chamber alter the impedance across a pair of electrodes which is opposite each other along the chamber walls. The other pair of non-current generating electrodes measures the changes in impedance.

Since no information available for local species ventilation and locomotion frequency, preliminary test was undertaken to identify the range of frequency (Hz) produced by each species in normal conditioned. As a result, specific range of frequency for ventilation and locomotion in natural condition were established and used for further TSS stress test. The test was undertaken in normal condition in which without TSS stressor and natural water was used. The behavioral pattern was recorded continuously for a period of 24 hrs and movement was recorded by the MFB unit at every 10 minutes. As a result 144 measurements were recorded for each fish and 10 fish were tested for each species.

A similar test was conducted for TSS effects and five different concentration were used (50 mg/L, 100 mg/L, 150 mg/L, 200 mg/L, 250 mg/L and 300 mg/L. Specific ranges of frequency for ventilation and locomotion gained from previous test were used and percentage of time spending for both activities was calculated. A one sample t-test was used to measure percentage variation from the normal condition. A significant variance recorded was considered to produce adverse effect to the

fish. A statistical package for social science (SPSS ver 20) was used for one sample t-test.

3.0 RESULTS AND DISCUSSION

3.1 Ventilation and Locomotion Standardization

Fish behavior was observed simultaneously using quadrapole and video camera to confirm type of behavior produced at specific wavelength. Two major activities were exhibited by the fish, which are locomotion (swimming and fin movement) and ventilation (operculum opening and closing). Swimming activity produces high amplitude and low frequency waveform and small fin movement, primarily pectoral and end of tail fin was of a lower amplitude and higher frequency and was seen to be less regular than swimming or ventilation waveforms. Ventilation activity produces the lowest amplitude but highest frequency (Figure 1). Although three activities produced, small movement was excluded due to irregular signal produced. Ventilation and locomotion contribute more than 80% of total activity recorded and therefore selected for assessment.

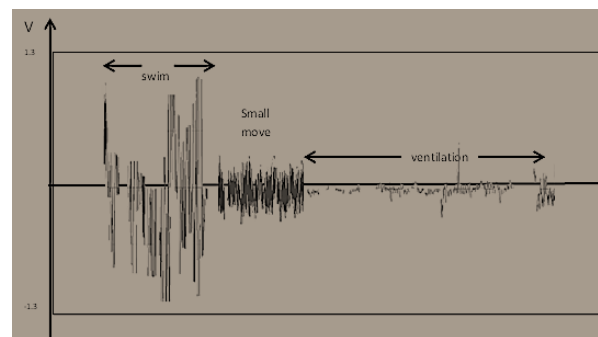


Figure 1 The amplitude graph shown three behaviours of fish sample: swimming, small fin and tail movement and ventilation

From 144 testing in normal conditions (unstressed), range of frequency for ventilation and locomotion for the three species were obtained. Each species exhibited large range of frequency. Behavioral signal responses of guppy fish (*Poecilia reticulata*), masher (*Tor tambroides*) and fighting fish (*Trichopsis vittatus*) show that all species were active during the entire one hour period in the control treatment.

Poecilia reticulata exhibits small range of ventilation but large for locomotion (Figure 2). As viewed using video recorder, ventilation range for this species was recorded as in the range of 0.5 Hz to 1.5 Hz. The locomotion range was recorded from 2 Hz to 4.5 Hz. During 24 hrs testing period, this species consumed 50.6% of time ventilation activity and rest for locomotion. Malaysian masher (*Tor tambroides*) exhibited different pattern. It's produced larger ventilation range that was 0.5 Hz to 2.5 Hz, but slim locomotion range (3 Hz to 3.5 Hz). They consume more than 70% time for ventilation (Figure 3). The fighting fish (*Trichopsis vittatus*) displayed moderate ventilation range that is 0.5 Hz to 1.5 Hz and 2.0 Hz to 3.0 Hz for locomotion (Figure 4).

Preliminary result indicates that all three species has their own behavior for ventilation and locomotion. However, one-way ANOVA test indicates that none of the species exhibits significant ventilation pattern. For locomotion activity, all species

exhibits large variation and significantly different ($p > 0.05$, $\alpha = 0.05$) [12, 13, 14]. Preliminary results show every species have almost similar ventilation frequency which generally in the range of 0.5 Hz to 2.5 Hz. However, locomotion is vary that could be due to species characteristics. Masheer is active fish compared to others and therefore exhibited more high frequency locomotion. This shows that ventilation activity is more applicable and accurate to be used for assessment.

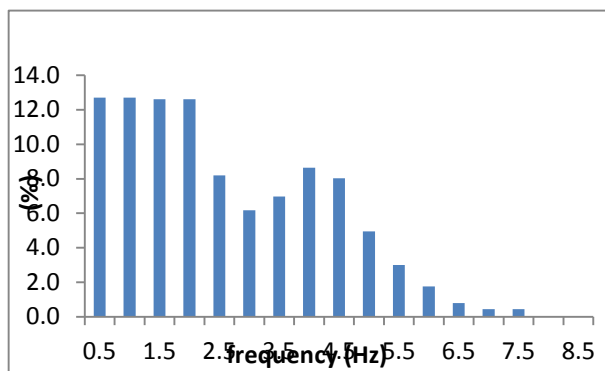


Figure 2 Percentage frequency ventilation and locomotion for *Poecilia reticulata*

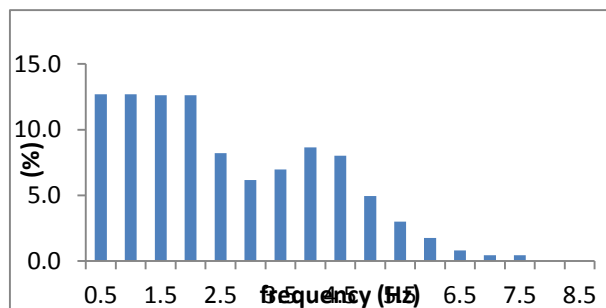


Figure 3 Percentage frequency ventilation and locomotion for *Tor tambroides*

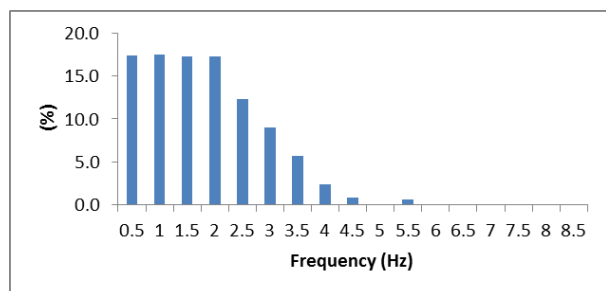


Figure 4 Percentage frequency ventilation and locomotion for *Trichopsis vittatus*

3.2 Stress Test with TSS

Six series of TSS concentration were used for stress test and each concentration was tested with similar treatment for each fish species. The TSS concentrations were chosen based on Malaysian national water standard to represents class I to class v. Result shows that locomotion activity does not affected by the TSS even for the highest concentration (300 mg/L). Each concentration

produce insignificant percentage variance when compared to the normal condition percentage. This observed occurs for all three species. As ventilation and locomotion were detected from minimum to maximum ranges, the stress test also undertaken at each frequency (Hz) with interval 0.5. The one-way ANOVA test indicates insignificant differences between TSS concentration and also between locomotion frequencies ($p > 0.05$, $\alpha = 0.05$). Similar insignificant results were produced by guppy and fighting fish. This explains that locomotion is less sensitive to TSS test and probably only suitable for higher concentration.

Ventilation respond to TSS effects was vary depends to frequency used and species tested. However, all species were exhibited stress level according to TSS gradient. Fishes increase their ventilation rates as TSS concentration increased.

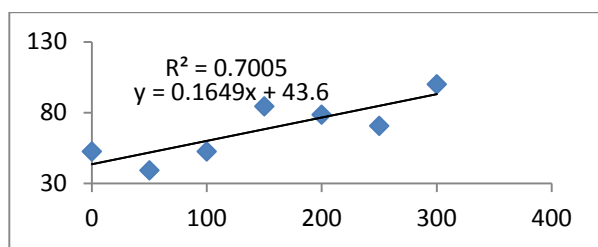
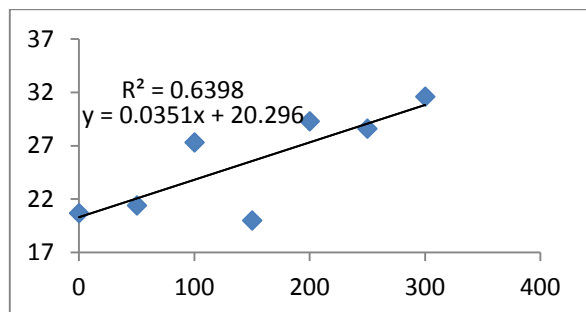
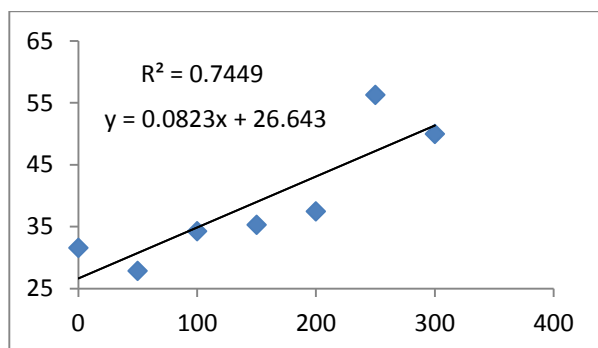
Due to large range of frequency available for ventilation, only the highest percentage of occurrence that recorded in preliminary test was selected. Based on preliminary result, the highest percentage for masheer, guppy and fighting fish were 3 Hz, 2 Hz and 2 Hz respectively. Result for TSS test is shown in Figure 5. Each species exhibits increment gradually in ventilation as TSS is increase and this true where fish breathe more in stress condition. As sediment begins to accumulate in the gill filaments, fish excessively open and close their gills to expunge the silt [15]. At 0 mg/L TSS (control) every species produce frequency approximately similar with the preliminary test (natural condition). This proves that selected frequency for the testing is suitable and representative. The Pearson correlation test indicates ventilation rate for all three species has strong positive correlation with TSS concentration. This study confirms the potential use of ventilation frequency as detector for TSS contaminations. The presence of TSS has been shown to produce gill flaring in response to short term sediment pulses [15, 16], and increased coughing frequency [17]. Fish gills are delicate and easily damaged by abrasive particles. If irritation continues, mucus is produced to protect the gill surface, which may impede the circulation of water over gills and interfere with fish respiration. This study will help to produce a base line data for TSS stress to local species. Indeed, it has been suggested that behaviour, as a summation of physiological activity, offers a more sensitive and ecologically relevant measure of environmental degradation [18].

4.0 CONCLUSION

Preliminary test has produced a baseline frequency for ventilation and locomotion and this is very useful as no information yet available. Ventilation is more sensitive than locomotion and suitable to detect low concentration of TSS. All tested species show adverse effect due to TSS contamination and exhibited positive respond to TSS concentration. This study was conducted in the laboratory and under controlled environment. Field experiment should be undertaken as other environmental factors could affect the fish responds.

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*Tor tambroides**Poecilia reticulata**Trichopsis vittatus***Figure 5** Correlation coefficient ventilation rate with TSS concentration

References

[1] Department of Environment's website. <http://www.doe.gov.my/webportal/en/info-umum/> 1 March 2014.

- [2] Gerhardt A. 2000. Recent Trends in Online Biomonitoring for Water Quality Control. In: Gerhardt A. Editor. Biomonitoring of Polluted Water. Zurich: Trans Tech Publications Ltd.
- [3] Gerhardt. A. Clostermann M., Fridlund B., Svensson, E. 1994. Monitoring of Behavioural Patterns of Aquatic Organisms with an Impedance Conversion Technique. *Environ Int.* 20: 209–219.
- [4] Craig, S. and P. Laming, 2004. Behaviour of the Three-spined Stickleback, *Gasterosteus aculeatus* (Gasterosteidae, Teleostei) In The Multispecies Freshwater Biomonitor: A Validation of Automated Recordings At Three Levels of Ammonia Pollution. *Water Research.* 38: 2144–2154.
- [5] Everest, F. H., R. L. Beschta, J. C. Scrivener, K. V. Koski, J. R. Sedell, and C. J. Cederholm. 1987. Fine Sediment and Salmonid Production: A Paradox. In E. O. Salo and T. W. Cundy [ed.]. Streamside Management: Forestry and Fishery Interactions. University of Washington Institute of Forest Resources, Seattle, Washington.
- [6] Rabeni, C. F., Doisy, K. E. and Zweigh, L. D. 2005. Stream Invertebrate Community Functional Responses to Deposited Sediment. *Aquatic Science.* 67: 395–402. from <www.rsl.psw.fs.fed.us/people/lreid.html>
- [7] Lloyd, D. S., J. P. Koenings, J. D. LaPerriere. 1987. Effects of Turbidity in Fresh Waters of Alaska. *North American Journal of Fisheries Management.* 7: 18–33.
- [8] Reid, L. M. 1998. Forest Roads, Chronic Turbidity, and Salmon. *EOS, Transactions, American Geophysical Union.* 79(45): F285.
- [9] Bo, F. T., S. G., Malacarne M. Pessino, and F. Sgariboldi 2007. Effects of Clogging on Stream Macroinvertebrates: An Experimental Approach. *Limnologia-Ecology and Management of Inland Waters.* 37: 186–192.
- [10] Billota, G. S. and R. E. Braizer, 2008. Understanding the Influence of Suspended Solids on Water Quality and Aquatic Biota. *Water Research.* 42(12): 2849–2861.
- [11] Kemp, P., Sear, D., Collins, A., Nadn, P. and Jones, I. 2011. The Impact of Fine Sediments on Riverine Fish. *Hydrological.* 25(11): 1800–1821.
- [12] Qureshi, M. I., Rasli, A. M., Awan, U., Ma, J., Ali, G., Alam, A., & Zaman, K. 2014. Environment and Air Pollution: Health Services Bequeath to Grotesque Menace. *Environmental Science and Pollution Research.* 1–10.
- [13] Rasli, A. M., Norhalim, N., Kowang, T. O., & Qureshi, M. I. 2014. Applying Managerial Competencies to Overcome Business Constraints and Create Values Evidence from Small Technology-based Firms in Malaysia. *Journal of Management Info.* 3(1): 99–121.
- [14] Qureshi, M. I., Rasli, A. M., & Zaman, K. 2014. A New Trilogy to Understand the Relationship among Organizational Climate, Workplace Bullying and Employee Health. *Arab Economic and Business Journal.* 9(2): 133–146.
- [15] Berg, L. 1982. The Effect of Exposure to Short-term Pulses of Suspended Sediment on the Behavior of Juvenile Salmonids. P. 177–196 in G. F. Hartman *et al.* [eds.]. *Proceedings of the Carnation Creek Workshop: A Ten-year Review.* Department of Fisheries and Oceans, Pacific Biological Station, Nanaimo, Canada.
- [16] Berg, L. and T. G. Northcote. 1985. Changes in Territorial, Gill-flaring, and Feeding Behaviour in Juvenile Coho Salmon (*Oncorhynchus kisutch*) Following Short-term Pulses of Suspended Sediment. *Canadian Journal of Fisheries and Aquatic Sciences.* 42: 1410–1417.
- [17] Servizi, J. A. and D.W. Martens. 1992. Sublethal Responses of Coho Salmon (*Oncorhynchus kisutch*) to Suspended Sediments. *Can. J. Fish. Aquat. Sci.* 49: 1389–1395.
- [18] Scherer E. 1992. Behavioural Responses as Indicators of Environmental Alterations: Approaches, Results, Developments. *J Ichthyol.* 8: 122–31.