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Rapid bioassessment of macroinvertebrate communities is suitable for monitoring the impacts of fish farm effluents

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ABSTRACT

Development of the fish farming industry in Iran in an environmentally and economically sustainable manner requires an effective and low-cost means of regularly monitoring receiving environments. Biomonitoring using macroinvertebrates is known to be effective for assessing water quality. The problem, however, is that biomonitoring can be labour intensive and analyses can have a long turnaround time. Rapid bioassessment methods have been developed to overcome these limitations, but it is not known whether they are as sensitive to changes in water quality as are their more time-consuming counterparts. To answer this question, we compared three methods for sampling and measuring macroinvertebrates. We refer to these as the quantitative method, semi-quantitative method, and qualitative method respectively. The quantitative method was a single habitat method with taxonomic identification of macroinvertebrates to genus level that counted all taxa. The semi-quantitative method involved multi-habitat sampling with identification to family level and quantification as relative abundance. The qualitative method was the same as the semi-quantitative method except that incidence (presence/absence) was recorded instead of abundance. The study was carried out at three fish farms in Iran with sampling done once per season for a year from the outfall of each farm as well as from the receiving rivers, with one sample taken upstream of the effluent discharge and two samples downstream. Analysis by permutational multivariate analysis of variance (PERMANOVA) revealed that the effects of three variables of season, farm, and site on macroinvertebrate communities were significant for all three methods. Qualitative sampling was the only method that showed a statistically significant interaction between farm and season as well as a difference among the sites within each farm. Although the results of a BEST (Bio-Bio) analysis showed that different families were responsible for the differences between the sites, all three methods were able to detect the differences between the sites within each farm. However, pairwise comparisons between sites within farms indicated some differences between the three methods. The quantitative method revealed fewer differences than did the other two methods. The qualitative method did not lose any important information and had the added advantage of saving considerable time and effort in sampling and enumerating. These results suggest that rapid bioassessment could be used to effectively monitor the receiving waters of fish farm effluents.

Statement of relevance: This manuscript compares three methods of sampling which are quantitative, semi-quantitative and qualitative methods in order to find the most efficient and cost-effective method of sampling. There is no apparent consensus on the appropriate method of collecting and measuring macroinvertebrates, in particular for investigating the effect of fish farms on the rivers. Our manuscript revealed that rapid bioassessment method as a cost-efficient and effective method can be used in order to develop aquaculture in a sustainable manner, both environmentally and economically. Therefore, authors believe that this manuscript is appropriate for publication by the Journal of Aquaculture.

1. Introduction

Aquaculture, once considered an environmentally friendly practice, is now recognised as a potential polluter of aquatic environments (Pillay, 2008). As fish farming intensifies and the size of farms increases, so does the potential impact on aquatic ecosystems (Boyd, 2003; F.A.O., 2000). Higher production rates result in an elevated production of waste materials including uneaten feed and metabolic waste products (Viadero et al., 2005) that may have a negative impact on the quality of the receiving water (Camargo, 1992; Schulz et al., 2003). In order to develop aquaculture in a sustainable manner, both environmentally and economically, cost-efficient

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methods for monitoring the impact of farm effluents need to be developed.

Benthic macroinvertebrates are widely used as indicators of the ecological condition of streams and rivers (Camargo, 1992; Rosenberg and Resh, 1993), but there is no apparent consensus on a single "best" method of collecting and enumerating macroinvertebrates. Collection methods have included quantitative sampling (e.g. Surber samples), where a fixed and repeatable area of sediment is sampled (Camargo and Gonzalo, 2007; Soofiani et al., 2012), or rapid bioassessment (RBA) methods that involve semi-quantitative sampling from a variety of habitats (e.g. kick-net samples from bottom sediments and sweep-net samples from edges) and qualitative assessment of the incidence of taxa (i.e. presence/absence) (Loch et al., 1996; Melo, 2005). Some researchers believe that quantitative sampling of sediments is required for the detection of moderate or subtle impacts (Kerans et al., 1992; Kilgour et al., 2005; Wright et al., 1995), while others suggest that the multi-habitat approach is more sensitive to disturbance gradients (Metzeling et al., 2003; Stark, 1993). The choice of method would seem to depend on which habitats are primarily affected, and it could be expected that trout farm effluents mainly affect the benthic region due to high suspended organic matter loads. Enumeration methods have included identification to family, genus or lower taxonomic resolutions, and quantification by counting of individuals or the recording of presence/absence. Sensitivity has been reported to be higher with species-level data (Jones, 2008; Taylor, 1997), but most researchers believe that family-level data needed less time, cost, and expertise while providing no less information than species-level data (Heino, 2008; Hewlett, 2000; Metzeling et al., 2003). With regard to quantification, numerous authors have demonstrated that presence/absence data is sufficient for most purposes (Hewlett, 2000; Marchant et al., 1995; Rutt et al., 1990).

While RBA methods that use qualitative sampling and enumeration of macroinvertebrate families using presence/absence potentially sacrifice information that could otherwise help to reveal important environmental impacts, these methods are quicker and cheaper to use and there is reason to believe they are sufficiently sensitive to the potential adverse effects of aquaculture effluents (Loch et al., 1996). To determine whether RBA methods are suitable for monitoring the impacts of fish farm effluents, we assessed samples taken from three fish farms adjacent to the Zayandeh-Roud River in Iran at sites and times that would likely differ in their macroinvertebrate community composition in order to compare three methods for collecting and enumerating macroinvertebrates. We refer to these methods as the quantitative method, semi-quantitative method, and qualitative method respectively. The quantitative method was a single habitat method with taxonomic identification of macroinvertebrates to genus level that counted all taxa. The semi-quantitative method involved multi-habitat sampling with identification to family level and quantification as relative abundance. The qualitative method was the same as the semi-quantitative method except that incidence (presence/absence) was recorded instead of abundance.

2. Methods

2.1. Site descriptions

The Zayandeh-Roud River, located in the centre of Iran, is spring-fed and drains approximately 41,500 km^2 of agricultural and urban land. It is characterised by low water velocities and stagnant water conditions, and receives effluent from aquaculture (mainly trout) farms. In order to compare the three methods of collecting and enumerating macroinvertebrates, samples were collected from three different fish farms over a period of one year (see Fig. 1). Farm 1, Dimeh farm, has a maximum production capacity of 250 tonnes per annum. It is located in the upper reaches of the river (32°50′ N 50°21′ E) and is spring-fed. Farm 2, Hojatabad farm, (32°71′ N 50°79′ E) has a much smaller production capacity of 25 tonnes and uses a natural lagoon for effluent treatment before discharging into the river. Farm 3, Takab farm with a production capacity of 70 tonnes, (32°37′ N 51°52′ E) discharges effluent directly to the river. Farms 2 and 3 are both river-fed. All facilities are flow-through, active year round, and maintain grower and brood stocks with the exception of farm 2 which engages only in rearing fingerlings to a marketable size.

Four sampling stations (sites) were assigned at each farm: site 1, outfall (where effluent discharges into the river); site 2, approximately 100 m upstream of the outfall; and sites 3 and 4 located approximately 100 m and 1 km downstream of the outfall respectively — referred to as downstream 1 and downstream 2. At each site, samples were selected systematically at each season, both for convenience and to reflect a range of communities.

2.2. Macroinvertebrate collection, identification, and enumeration

Benthic samples were taken at each of the sites quarterly during autumn and winter of 2007 and spring of 2008. For the quantitative sampling method, triplicate samples of the benthic macroinvertebrates were collected with a Surber sampler by using 2–3 min kicks from a riffle habitat. The sampler was placed on the substrate to delineate a 0.062-m^2 sampling area. The dislodged organisms were captured in the attached net with 60-μm mesh size (Barbour et al., 1999). For the purpose of this study, all three replicates were combined. Therefore, a total area of 0.186 m^2 was covered for this method.

Samples for the semi-quantitative and qualitative methods were collected as described by Lenat (1988) except for a few modifications. Samples were taken without any replication. In addition, to be consistent with the quantitative method, only 2–3 min was spent on each sample collection. This sampling method attempts to collect most, if not all, available microhabitats. The macroinvertebrate samples were collected using a multi-habitat method comprising four samples: a kick-net collection (from riffle habitat); a sweep-net (collection from bank area); a leaf pack and a collection made by visual inspection of large rocks (5–10), depending on their size; and logs (Eaton and Lenat, 1991; Lenat, 1988; Loch et al., 1996; Metzeling et al., 2003). The four habitat samples were combined to give an overall assessment of the macroinvertebrate communities at each site.

All samples were preserved in 70% ethanol. The macroinvertebrates were identified to genus level for the quantitative method and to family level for the semi-quantitative and qualitative methods with the aid of a dissecting microscope. Individuals were counted for the quantitative and semi-quantitative methods, whereas taxa were recorded as present/absent for the qualitative method. Taxonomic keys were used for the identification of macroinvertebrates (Elliott et al., 1988; Hynes, 1977; Pescador et al., 1995). Benthic taxa found in the present study are presented in Appendix A.

2.3. Statistical analyses

All analyses were performed with the software package PRIMER v6 and PERMANOVA+ v1.0.3 (Anderson et al., 2008; Clarke and Gorley, 2006). Exploratory analyses were undertaken using principal coordinates analysis (PCO). To determine whether the three methods provide the same level of information concerning changes in macroinvertebrate communities and to test for the main effects of season, fish farm, and site, as well as their interactions, a three-factor (season, farm, and site) PERMANOVA was used. The highest-order interaction term (season × farm × site) was excluded from the model to compensate for the lack of replication within seasons. Exclusion of the third-order interaction term was justified on the grounds that while it could be expected that there will be an interaction between farm and site (i.e. that dissimilarities among sites will vary across farms), there is no reason to believe that this interaction would differ between seasons. Hypothesis testing was performed using 9999 permutations. In all cases except for the qualitative method, which was based on the presence/absence dichotomy, the data were square-root transformed and all multivariate