Chapter 8

8.0 General Discussion and Conclusions

Understanding various stages of fish development coupled with their nutrition requirements would give the aquaculture practitioners a good viewpoint in developing a closed aquaculture system (Pinto *et al.*, 2010). Very little research has been devoted to the development of diet formulation based on insect especially super worm, *Zophobas morio* meal although their availability was quite common in Malaysia. Due to lack of nutrient information on this insect, its application to fish diet formulation was not researched upon. Indeed, generalization of nutritional information of alternative protein sources particularly insect in fish diets can be problematic due to wide variation of feeding habits, digestive capabilities and tolerance of insect-derived feedstuffs among fish. Thus, identification of alternative sources for fish meal replacement is specific-species.

The aim of this research was to investigate the nutritive value of selected insect and their suitability as alternative protein source to dietary fish meal in the diet of red tilapia *Oreochromis* spp. Although current tilapia diets contain less of fish meal and a large proportion being replaced by soybean meal (SBM) which is still considered as the best readily available protein source. However, it does contain endogenous antinutitional factors (ANF) that affects health and fish growth performance. It is important to note that the dietary inclusion of SBM in tilapia feeds is affected by dietary protein level. Shiau *et al.* (1987) showed that FM could be partially replaced by SBM within tilapia hybrid feeds containing sub optimal protein level (24%) whereas optimum protein level (32%) significantly depressed fish performances. In this research, insects are used to replace fish meal proportionately to be used in tilapia diets in order to make tilapia aquaculture more profitable and sustainable in the future. Importance of insect protein as food and feed has gained attention from FAO recently (FAO, 2012).

Super worm meals are most promising insect based alternatives to fish meal based on nutritional profile. They are a good source of protein, mineral, lipid and fatty acid and provide adequate energy (Bukkens, 1997). The use of meal worm as an alternative protein source has been studied in catfish and showed that catfish fed diets with up to 80% replacement of fish meal with the worm meal still displayed good growth and feed utilization efficiency (Ng *et al.*, 2001).

Major conclusions of this study are as follows:

1) The present study demonstrated that protein content of SWM was valued at 43.83% which can be considered as good protein source although not as high as FM. Generally, SWM has a good essential amino acid profile since this future substitute of FM appeared to be comparable and applicable to be incorporated into the new formulated diet. Arginine (21.91 mg/g of protein) revealed a high content while the rest were within the comparable values needed by fish. Besides, SWM was recommended as a potential candidate because all seventeen amino acids were available. The availability of higher lipid content (40.01%) compared to FM has been analysed to reveal content of fatty acids profile. The present study also revealed that SWM contained polyunsaturated fatty acids mostly linoleic (C18:2n-6, 23.42%) which was higher than polyunsaturated fatty acid content in FM.
2) Evaluation of SWM as a partial substitution of FM at various levels (25%, 50%, 75% and 100%) in red tilapia diets showed that up to 50% replacement of FM can be accepted without any adverse effects on fish performances. In the present study, higher level of SWM inclusion has led to a reduction in size and weight of fish (Table 5.2). Normally, fish will grow up to 400% in weight if they were cultured in a good environment. There was no sign of mortalities and the study showed that fish survived well (100% in all

treatments) even though fish seemed to be stressed especially at the beginning of the experiments.

3) A combination or supplementation attempt has been carried out to compensate any deficiencies in essential amino acid particularly methionine that may play a major role in growth. Fish fed with Diet 3 supplemented with 1.0% DL-Methionine has effectively showed a significant growth response (135.98%) compared to diet without supplementation. This is evidence showing that supplementation with a chemical substance such as DL-Methionine into fish feed formulation can make the nutrient contents closely resembling FM. The present results showed that red tilapia could well utilize crystalline methionine leading to growth improvements.

4) The best performing diet in terms of weight gain was the diet with 10% (w/w) supplementation level of MSM. Weight gain value of fish fed with Diet 2 (165.11%) was highly significant compared to Diet 3 and Diet 4. Fish fed with Diet 2 still had high survival rate at 93.33%. This may be attributed to adequate level of MSM supplementation that could stimulate its immune system against viral infections which could lead to a decrease in mortality rate.

Dietary protein is always considered to be of primary importance to fish feeding (Jauncey and Ross, 1982). In the present study, the optimum dietary protein level is 30-34% for red tilapia fry (>5g / fish). This is in agreement with Balarin and Halfer (1982) who reported that dietary protein level is 25-35% for 5-25g tilapia fingerlings. However, De Silva *et al*. (1989) demonstrated that the suitable dietary protein requirement for tilapia from 1g to 5g was 28%. Ahmad *et al*. (2004) concluded that the variations in optimum dietary protein requirement for maximum growth might be due to variations in size, stocking density and protein quality of protein feedstuffs.

Kalsoom *et al.* (2009) mentioned FCR as a value obtained from the number of kg feed used to produce one kg fish. Low FCR is usually required in feeding practice. In the present study, average FCR recorded ranged between 1.15 – 1.86. Al- Hafedh (1999) found that FCR ranged from 1.5 - 2.6 for 0.51g fry. However the experiment was conducted during the winter season (18 – 25 ˚C). In this study, water temperature range was 26 – 27 ˚C which represents the optimum range of temperature for tilapia is 26 – 29 ˚C (Chervinski, 1982).

Another cause for differences among diets apart from the nutritional value of protein feedstuffs can be due to the inability to digest fiber. Fiber can have a physical effect on feed digestion and can reduce the time available for nutrient absorption, further decreasing gut retention time. However, most fish that contains fiber below 3% showed good performances in this current study. Results suggest that fiber content effects were likely minimal in this study. In addition, fiber was regarded to be one of the major factors for poor performance (Ali *et al.*, 2008; Krogdahl *et al.*, 2010). The protein to energy (P/E) ratios in all diets was within the range 16.26 – 19.43 mg/Kj suggested by De Silva *et al.* (1989). In this study, lipid content in all diets increased with increasing SWM inclusion or supplementation level of MSM and DL-Methionine. Thus, future studies are needed to optimize the dietary lipid when fish diets are formulated. Mohanta *et al.* (2007) revealed that providing adequate non protein from lipid can minimize the use of protein as source of energy. Red tilapia juveniles in the present study gained little form dietary supplementation of crystalline amino acid due to leaching factor. Coated or protected EAA should be used leading to minimum EAA leaching (Viola *et al*., 1988).

The whole body compositions of tilapia were not affected in the DL-Methionine and MSM supplementation experiments. However, the whole body protein compositions were affected by FM replacement of SWM diet treatments. The

experimental diets were formulated isonitrogeneously based on tilapia nutrient requirement for the juvenile stage (32% of crude protein). Any difference in the performance of fish growth was probably due to quality of tested ingredients used and levels applied (Soltan *et al.,* 2008). It is reported in this current research that the whole body lipid composition is reduced with the increasing amount of the level of dietary lipid in fish diet. This result is in agreement with Nandeesha *et al.* (2000) findings who suggested that silkworm pupae fat as an energy source in common carp diets. In addition, insect derived oil such as super worm and silkworm pupae is likely rich in short chain unsaturated fatty acids, as similarly found in this current study and has been found to be an excellent energy source. However, the high lipid content in super worm can be solved by providing the enriched wastes to be consumed. Insect has the ability to convert lost nutrients in their biomass by consuming the wastes. Sealey *et al.* (2011) showed that *Hermetia illucens* larvae have the potential to convert lost nutrients by incorporating residual amino acids in wastes. Teshima *et al.* (1985) also added that *O. niloticus* utilized lipid more efficiently as an energy source when the dietary protein was insufficient. Our previous studies indicated that the fish oil inclusion may also be reduced by SWM inclusion due to its high lipid content. In fish diets, fish oil like fish meal is also expensive.

The present study has demonstrated that SWM could be used as partial substitutes for fish meal in red tilapia diets at 50% level of inclusion without sacrificing growth and feed utilization. However, the use of supplements such as DL-Methionine in the diets can add to the final cost of diet production and may even be more expensive than fish meal based diets.

Future Studies and Expectations

This research can be further expanded to include the following:

1) Future studies need to expand on the effect of super worm based diet and fresh super worm solely fed to tilapia juveniles. Other potential insect such as black soldier larvae which is also readily available and can be found in abundance locally should be studied to develop more awareness about insect protein as a novel protein source among researchers in fish nutrition.
2) Since the utilization of purified protein and crystalline amino acid are costly and time consuming, development of efficient methods and standardization of the quantitative dietary EAA requirement is needed to give a maximal effect on fish nutrition.
3) This present study was carried out on a laboratory scale. Since this study was managed under laboratory condition, it could be practically conducted in semi intensive culture as this practice was commonly used by farmers in Malaysia. Therefore, long term studies should be managed with fish growing at maximum level at each phase of life cycle (fry, fingerlings, juveniles, growers) using insect-based diet.
4) More holistic economic analysis should be conducted taking into account all factors in fish production under realistic farming conditions. It is suggested that overall production cost should be considered such as labour, electricity cost and so on.
5) Some further studies were suggested to improve on the current result including research on influence of super worm meal substitution on intestinal histology and activity of digestive enzymes determination. An assessment of fish quality needs to be evaluated.

Continued research on sustainable and economical diets for red tilapia will help the development of tilapia aquaculture in Malaysia. Although this is just a small contribution to the vast fish nutrition aspect, this first attempt will be a key factor to the development of a balanced and nutritionally sound aquafeed that are economical and profitable.