COMPARATIVE DIVERSITY, RELATIVE ABUNDANCE, AND FUNCTIONAL GENES OF BACTERIAL COMMUNITY IN A SEMI-CLOSED MARINE FISH HATCHERY BETWEEN DRY AND WET SEASONS

By

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Aquaculture has developed rapidly to become one of the most important food industries in the world. In recent years, increases in demands for local and exports markets of high-value fish species such as groupers (*Epinephelus* sp.) and Asian seabass (*Lates calcarifer*) have encouraged hatcheries to produce more fry to supply the demand from farmers. The increasing trend to develop large-scale production has led to intensive marine aquaculture practices, thus vulnerable to disease outbreaks that affects the quality of fry production. This study investigates the bacterial diversity and its functional genes that present in a semi-closed marine fish hatchery ecosystem between dry and wet seasons, by implementing amplicon metagenomics using Next-Generation Sequencing (NGS). The intestinal samples were collected from different sizes of tiger groupers (n = 9, 16.37 ± 5.46 cm, 54.00 ± 44.03 g and n = 9, 20.86 ± 8.73 cm, 133.43 ± 158.15 g for dry and wet season respectively) and Asian seabass (n = 10, 14.27 ± 4.45 cm, 41.93 ± 35.12 g and n = 13, 16.13 ± 6.83 cm, 60.45 ± 55.07 g for dry and wet season, respectively) as well as the culturing water (10 L), before proceeded to DNA extraction. The extracted DNA were then subjected to 16S rRNA amplicon sequencing using Illumina Miseq platform, followed by determination and comparison of the bacterial diversity, abundance and functional genes between both seasons. Selected physicochemical and seasonal parameters were also determined. Results revealed that gut microbiota of the Asian seabass were dominated by phylum Proteobacteria and order Vibrionales during both seasons. Meanwhile, gut microbiome of tiger groupers shifted from domination of phylum Firmicutes and order Clostridiales during the dry season to Proteobacteria and Lactobacillales during the wet season. Water samples were dominated by phylum Proteobacteria and order Stramenopiles in dry season but shifted to phylum Proteobacteria and order Flavobacteriales in the wet season. Total dissolved solid (TDS), water temperature (T), conductivity (C), salinity (S), ammonia (NH_4), nitrite (NO_2), nitrate (NO_3), sulphate (SO_4), average temperature (AT) and average rainfall (AR) showed significant different between dry and wet season. Proteobacteria, Firmicutes and Fusobacteria showed correlation towards the C, SO_4, T, PO_4, AR, AH and AT during the dry season, meanwhile phylum Bacteroidetes, Fusobacteria, SBR1093, WPS-2 and Verrucomicrobia showed strong correlation towards all environmental parameters during wet season. PICRUSt revealed functions that dominantly present in the tropical marine fish hatchery were metabolism, genetic information processing, environmental information processing, cellular process and human diseases. Remarkably, SIMPER analysis showed several potential metagenomics biomarker genes such as K03406, K06147 and K03088 in comparing microbiota between dry and wet season which can be utilised to be an indicator for the tropical marine fish hatchery environments. This study highlighted the dominant taxa and their putative function that dominated tiger grouper and Asian seabass as well as water samples in dry and wet season with the implementation if 16S amplicon approach better determination of the microbial profiles in the aquaculture systems.

Keywords: 16S rRNA, Asian seabass, hatchery, metagenomics, tiger grouper

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Sektor akuakultur merupakan salah satu sektor yang menyumbang kepada pembangunan industi makanan di Malaysia. Sejak kebelakangan ini, permintaan ikan yang berkualiti tinggi seperti kerapu (*Epihephelus* sp.) dan siakap (*Lates calcarifer*) semakin bertambah secara import mahupun eksport. Ini secara tidak langsung menggalakkan pengusaha pembenihan ikan marin menghasilkan lebih banyak benih untuk memenuhi permintaan daripada penternak ikan. Namun begitu, kesan daripada peningkatan tersebut, hasil marin tersebut semakin terdedah kepada pelbagai penyakit yang memberi kesan kepada kualiti pengeluaran benih ikan. Kajian ini mengkaji tentang perbandingan diversiti bakteria dan gen berfungsi yang wujud di dalam ekosistem pusat pembenihan ikan marin separa tertutup di antara musim kering dan musim tengkujuh dengan mengadaptasikan konsep metagenomik dan penjujukan generasi hadapan *(NGS)*. Sampel usus ikan dari pelbagai saiz diambil daripada ikan kerapu harimau (n = 9, 16.37 ± 5.46 cm, 54.00 ± 44.03 g dan n = 9, 20.86 ± 8.73 cm, 133.43 ± 158.15 g merujuk kepada musim kering dan tengkujuh) dan siakap (n = 10, 14.27 ± 4.45 cm, 41.93 ± 35.12 g dan n = 13, 16.13 ± 6.83 cm, 60.45 ± 55.07 g merujuk kepada musim kering dan tengkujuh) sebelum pengekstrakan DNA dibuat. Sampel air juga diambil daripada tangki ternakan ikan (10 L) dan pengekstrakan DNA juga dijalankan. DNA yang telah diekstrak dihantar untuk proses 16*s Amplicon Sequencing* menggunakan pelantar Illumina Miseq dan seterusnya proses data analisis untuk mengenal pasti dan membandingkan diversiti dan kekerapan bakteria serta gen yang berfungsi. Bacaan parameter fizikokimia dan parameter musim juga turut direkodkan. Dapatan daripada kajian ini menunjukkan mikrobiota di dalam usus ikan siakap semasa musim kering dan tengkujuh didominasi oleh filum Proteobakteria dan order Vibrionales manakala bagi ikan kerapu, ianya didominasi oleh filum *Firmicutes* dan order Clostridiales pada musim kering tetapi pada musim tengkujuh, mikrobiota yang mendominasi adalah filum

Proteobacteria dan Lactobacillales. Untuk sampel air, mikrobiota yang mendominasi adalah daripada filum Proteobacteria dan order Stramenophiles. Pejal terlarut (TDS), suhu air (T), konduktiviti (C), kemasinan (S), ammonia (NH_4), nitrit (NO_2), nitrat (NO_3), sulfat (SO_4), suhu purata (AT), dan purata hujan (AR) menunjukkan terdapat perbezaan ketara dia antara musim kering dan musim tengkujuh. Filum Proteobacteria, Firmicutes dan Fusobacteria menunjukkan hubung kait terhadap C, SO_4, T, PO_4, AR, AH dan AT semasa musim kering, manakala filum Bacteroidetes, Fusobacteria, SBR1093, WPS-2, Verrucomicrobia dan Cyanobacteria menunjukkan hubungkait yang positif terhadap semua parameter semasa musim tengkujuh. Dapatan daripada analisa PICRUSt menunjukkan bahawa gen berfungsi yang mendominasi adalah dikodkan untuk proses informasi alam sekitar, proses selular dan penyakit manusia. Disamping itu, analisis SIMPER menunjukkan beberapa gen yang berpotensi untuk menjadi penunjuk biologi metagenomik iaitu K03406, K06147 and K03088 untuk ekosistem pusat penetasan ikan marin tropika di antara musim kering dan musim tengkujuh. Kajian ini mendedahkan dominan taxa dan gene berfungsi dan kepentingan di musimkering dan musim tengkujuh menggunakan aplikasi metagenomik untuk lebih memahami kepentingan profil mikrobiota di dalam sistem akuakultur.

Keywords:16S rRNA, kerapu, metagenomik, siakap, tempat penetasa

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LIST OF ABBREVIATIONS

PCR	Polymerase Chain Reaction
TiLV	Tilapia Lake Virus
EMS	Early Mortality Syndrome
DO	Dissolved oxygen
PI	Posterior intestinal
MI	Mid-intestinal
NGS	Next-Generation Sequencing
ePCR	Emulsion Polymerase Chain Reaction
PICRUSt	Phylogenetic Investigation of Communities by Reconstruction of Observed States
OTU	Operational taxonomic units
KO	KEGG Orthology
WGS	Whole genome sequencing
QIIME	Quantitative Insights into Microbial Ecology
psu	Practical salinity unit
GI	Gastrointestinal tract
TDS	Total dissolved solid
NH3	Ammonia
NO2	Nitrite
NO3	Nitrate
PO4	Phosphate
SO4	Sulphate
AR	Average rainfall
AT	Average temperature
AH	Average humidity

mM	Milimolar
µm	Micrometer
L	Liter
FLASH	Fast Length Adjustment of Short reads
PEAR	Paired-end read merger
PERMANOVA	Permutational multivariate analysis of variance
PAST	PAleontological Statistics
NMDS	Non-metric multidimensional scaling
LEfSe	Linear discriminant analysis effect size
SIMPER	Similarity percentages

CHAPTER 1

INTRODUCTION

1.1 General Introduction

Fish disease has been a restricting factor in increasing fish production, mainly affecting farmers, traders and customers, resulting in substantial financial losses. The infantile immune response in fish makes the early developmental stages more vulnerable to infectious diseases in the hatchery. In hatcheries and early rearing systems, common fish infections are spread by protozoans, ciliates, myxosporodians, worms, opportunistic bacteria and fungi (Faruk and Anka, 2017). The production and survival rate of healthy fish seeds relies on appropriate approaches for health management, a conservation of good water quality, healthy diet and the implementation of biosecurity controls (Assefa and Abunna, 2017). One of the prerequisites for sustainable agriculture is healthy and disease-free fish seeds. Many systems were used to culture aquatic organism such as open, semi-closed and close system were practiced in aquaculture (Rud et al., 2016). In Malaysia, system that mainly practiced for hatchery production were semi-closed and closed system. This study was focusing on the marine fish hatchery which implement semi-closed system to produce fish seed for local fish farming. Seasonal variation which causes on temperature and other parameter changes can affected the fish gut microbiota in terms of composition and their abundance (Ringo et al., 2016). Metagenomics defined as combination of the technology in genomics and bioinformatics to identify genetic characteristic of the microorganism populations directly from the environment, which contain very complex genome within the microbial niche (Anderson et al., 2013). Metagenomics can be divided into two approaches, i.e. amplicon metagenomics (16S rRNA amplicon) and shotgun metagenomics. Amplicon metagenomics mainly focused on particular genes, which can be amplified by using polymerase chain reaction (PCR) before being sequenced and analyzed. In contrast, shotgun metagenomics gives broader description on potential genes diversity in the microbial world, which is much diverse than the amplicon metagenomics as shotgun sequenced all of the DNA that isolated from the environmental samples (Gilbert and Dupont, 2011). This study utilized 16S amplicon metagenomics to determine the bacterial community in a marine fish hatchery, coupled with metagenome prediction using PICRUSt to understand the microbial functions within the aquaculture environment during dry and wet season. The accelerating advancement of the metagenomics field contributed to the discovery of the timely diseases as control strategies in aquaculture and make it easier for identification and monitoring activities of the microbial communities in different ecosystems. Compared to the use of the classical gene sequencing such as polymerase-chain reaction (PCR), this conventional technique lacks the capacity to provide a general overview of the genome transcript within living organism (Munangandu and Evensen, 2017).

The field of metagenomics is a powerful tool to give information such as biocatalysts of enzymes, linkages of genome for function and phylogeny for cultured organisms, and evolutionary profile functions and structure of the microbial community (Wilmes & Bond, 2008; Gilbert et al., 2010). In comparison to the conventional approaches in which the outbreak agents were detected only after triggering the disease outbreaks, the use of metagenomics in aquaculture has boosted the capacity of researchers to diagnose the evolving pathogen before it causes outbreaks (Alavandi and Poornima, 2012; Munangandu et al., 2017), development of natural diets with less adverse effects (Gajardo et al., 2016) and discover the cellular process which manage different biological responses in aquatic organisms (Xu et al., 2015; Xu et al., 2016).

1.2 Problem statement

Widely known husbandry practices that are being practiced in intensive culture systems are high stocking densities, excess feeding and artificial fertilization. These provide an optimal environmental condition to produce numerous types of fish pathogens, which cause host species to suffer from stress as a consequence. Mohan (2007) found a few pathogenic bacteria triggering morbidity of seedlings in hatcheries and nurseries which include *Aeromonas, Vibrio* and *Pseudomonas.* Either as primary pathogens or as secondary opportunistic invaders, bacteria could lead to disease. The disease can cause mass mortality of the infected seed in a brief period, generate sustained small-scale mortality, decrease growth and result in low quality seeds, depending on the severity and nature (Bondad-Reantaso, 2007). In relation, seed originating from infected hatcheries could have been infected and the pathogens could be transmitted to grow-out systems where epidemic could occur and resulting in stocked seed mortality. Seasonal impact on the environmental parameters such as temperature raising of the seawater during the dry season showed correlation with the increasing of the potentially virulent *Vibrio* spp. And the disappearance of the lactic acid bacteria (LAB) which could have negative impact on the fish health (Neuman et al., 2016). The microbiome diversity of the gastrointestinal is often implemented as a diagnostic tool to monitor the fish health and metabolic process, with least diversity of microbiota and stability closely suggesting that microbial infection may be present (Clarke et al., 2014).

Gastrointestinal microbiota, a strategic function for mitigating emerging aquaculture diseases, may affecting fish physiology, growth, life span, immunity, and prevalent to pathogenic infection (Burns et al. 2016; Nie et al. 2017; Smith et al. 2017). In order to achieve this, a fundamental understanding on the relationship between fish health, microbiota, invading infectious agents and influenced of seasonal parameters are required. For example, higher microbial diversity and variations of *Vibrio* population were observed in Portuguese marine aquaculture in warmer season which involving seasonal factors such as temperature, pH and chemical parameters of the water (Pereira et al., 2011; de Brujin et al., 2017). Microbiome of the gastrointestinal tract therefore play a vital role in fish health. Aquaculture production system also plays an important role for prolong production of marine fish. This study mainly focusing on semi-closed system during dry and wet season which has recirculating water system and microbiota that present in this system potentially have impact on the fish health.

This study will utilize amplicon metagenomic approach using V3 and V4 region of 16 rRNA gene to understand the bacterial community from fish hatchery ecosystem that producing commercial marine fish fry during wet and dry season. The results are important to investigate the diversity patterns of the bacterial flora within the fish hatchery environment, and their functional gene that responsible for their biological processes. The bacteria and gene identification may help in developing microbiome-based diagnostic tool in monitoring and speed up the identification of pathogen in fish, thus help fish farming sectors to avoid massive losses due to pathogen infection in Malaysia. However, the application of metagenomics in aquaculture related study are still scarce in this country.

1.3 Objectives of studies

The objectives of this study are:

i) To determine the diversity and abundance of the bacterial community that present in a water sample, tiger grouper and Asian seabass from tropical marine fish hatchery between dry and wet seasons.
ii) To identify the functional genes of bacterial community that presence in a water sample, tiger grouper and Asian seabass from tropical marine fish hatchery between dry and wet seasons.

1.4 Hypothesis

Objectives 1:

H_0: There is no significant difference in terms of diversity and abundance of the bacterial community that presence in a water sample, tiger grouper and Asian seabass from semi-closed topical marine fish hatchery between dry and wet seasons.

H_a: There is a significant difference in terms of diversity and abundance of the bacterial community that presence in a water sample, tiger grouper and Asian seabass from semi-closed topical marine fish hatchery between dry and wet seasons.

Objectives 2:

H_0: There is no significant difference of functional genes of bacterial community that presence in a water sample, tiger grouper and Asian seabass from semi-closed tropical marine fish hatchery between dry and wet seasons.

H_a: There is a significant difference of functional genes of bacterial community that presence in a water sample, tiger grouper and Asian seabass from semi-closed tropical marine fish hatchery between dry and wet seasons.

REFERENCES

Abia, A. L. K., Alisoltani, A., Keshri, J., and Ubomba-Jaswa, E. (2017). Metagenomics analysis of the bacterial communities and their functional profiles in water and sediments of the Apies River, South Africa, as a function of land use. Science of the Total Environment, *616*: 326-334.

Abubucker, S., Segata, N., Goll, J., Schubert, A. M., Izard, J., Cantarel, B. L., and White, O. (2012). Metabolic reconstruction for metagenomics data and its application to the human microbiome. PLoS Computational Biology, *8*(6), e1002358.

Ahmad, A. K., Amal, M. N. A., Saad, M. Z., Murni, M., Abdullah, A., Mustafa, S., and Yusof, N. H. N. (2019). Prevalence, Risk Factors and Transmission of Nervous Necrosis Virus in A Hatchery Producing Hybrid Grouper (*Epinephelus lanceolatus* _ *Epinephelus fuscoguttatus*) Fry. Pertanika Journal of Tropical Agricultural Science, *42*(1).

Ahmadian, A., Gharizadeh, B., O'Meara, D., Odeberg, J., and Lundeberg, J. (2001). Genotyping by apyrase-mediated allele-specific extension. Nucleic Acids Research, *29*(24): 121.

Alavandi, S. V., and Poornima, M. (2012). Viral metagenomics: a tool for virus discovery and diversity in aquaculture. Indian Journal of Virology, *23*(2): 88-98.

Amal, M. N. A., Koh, C. B., Nurliyana, M., Suhaiba, M., Nor-Amalina, Z., Santha, S., and Zamri-Saad, M. (2018). A case of natural co-infection of Tilapia Lake Virus and Aeromonas veronii in a Malaysian red hybrid tilapia (Oreochromis niloticus× O. mossambicus) farm experiencing high mortality. Aquaculture, *485*: 12-16.

Amann, R. I., Ludwig, W., and Schleifer, K. H. (1995). Phylogenetic identification and in situ detection of individual microbial cells without cultivation. Microbiology and Molecular Biology Reviews *59*(1): 143-169.

Amato, K. R., Yeoman, C. J., Kent, A., Righini, N., Carbonero, F., Estrada, A., and Leigh, S. R. (2013). Habitat degradation impacts black howler monkey (Alouatta pigra) gastrointestinal microbiomes. The ISME Journal, *7*(7), 1344-1353.

Andersen, L. O. B., Nielsen, H. V., and Stensvold, C. R. (2013). Waiting for the human intestinal Eukaryotome. The ISME Journal, *7*(7), 1253- 1255.

Arasu, A. R. T., Kailasam, M., Sundaray, J. K., Subburaj, R., Thiagrajan, G., and Karaiyan, K. (2008). Improved hatchery technology for Asian seabass *Lates calcarifer* (Bloch). Special Publication, (*34*): 38.

Arnold, C. R., Wakeman, J. M., Williams, T. D., and Treece, G. D. (1978). Spawning of red snapper (Lutjanus campechanus) in captivity. Aquaculture, *15*(3), 301-302.

Assefa, A., and Abunna, F. (2018). Maintenance of fish health in aquaculture: review of epidemiological approaches for prevention and control of infectious disease of fish. Veterinary Medicine International, 2018.

Austin, B., and Zhang, X. H. (2006). Vibrio harveyi: a significant pathogen of marine vertebrates and invertebrates. Letters in applied microbiology, *43*(2), 119-124.

Baldani, J. I., Videira, S. S., dos Santos Teixeira, K. R., Reis, V. M., de Oliveira, A. L. M., Schwab, S., and Hartmann, A. (2014). The family rhodospirillaceae. The Prokaryotes: Alphaproteobacteriaand Betaproteobacteria, 533-618.B Retrieved from https://link.springer.com/referenceworkentry/10.1007%2F978-3-642-30197-1_300

Barb, J. J., Oler, A. J., Kim, H. S., Chalmers, N., Wallen, G. R., Cashion, A., and Ames, N. J. (2016). Development of an analysis pipeline characterizing multiple hypervariable regions of 16S rRNA using mock samples. PLoS One, 11(2): e0148047

Bashir, Y., Pradeep Singh, S., and Kumar Konwar, B. (2014). Metagenomics: an application-based perspective. Chinese Journal of Biology, 2014.

Bates, N. R., Best, M. H. P., Neely, K., Garley, R., Dickson, A. G., and Johnson, R. J. (2012). Detecting anthropogenic carbon dioxide uptake and ocean acidification in the North Atlantic Ocean. Biogeosciences, 9(7): 2509-2522.

Bazakos, C., Dulger, A. O., Uncu, A. T., Spaniolas, S., Spano, T., and Kalaitzis, P. (2012). A SNP- based PCR–RFLP capillary electrophoresis analysis for the identification of the varietal origin of olive oils. Food chemistry, 134(4), 2411-2418.

Bentley, D. R., Balasubramanian, S., Swerdlow, H. P., Smith, G. P., Milton, J., Brown, C. G., and Boutell, J. M. (2008). Accurate whole human genome sequencing using reversible terminator chemistry. Nature, 456(7218): 53.

Betancur, L. A., Naranjo-Gaybor, S. J., Vinchira-Villarraga, D. M., Moreno-Sarmiento, N. C., Maldonado, L. A., Suarez-Moreno, Z. R., and Ramos, F. A. (2017). Marine Actinobacteria as a source of compounds for phytopathogen control: An integrative metabolic profiling/bioactivity and taxonomical approach. PLoS One, *12*(2), e0170148.

Bibby, K. (2013). Metagenomics identification of viral pathogens. Trends in Biotechnology, *31*(5): 275-279.

Birtel, J., Walser, J. C., Pichon, S., Bürgmann, H., and Matthews, B. (2015). Estimating bacterial diversity for ecological studies: methods, metrics, and assumptions. PloS One, *10*(4): e0125356.

Blankenberg, D., Kuster, G. V., Coraor, N., Ananda, G., Lazarus, R., Mangan, M., and Taylor, J. (2010). Galaxy: a web-based genome analysis tool for experimentalists. Current protocols in molecular biology, *89*(1), 19-10.

Bolhuis, H., Cretoiu, M. S., and Stal, L. J. (2014). Molecular ecology of microbial mats. FEMS Microbiology Ecology, *90*(2): 335-350.

Bondad-Reantaso, M. G. (Ed.). (2007). Assessment of freshwater fish seed resources for sustainable aquaculture (No. 501). Food and Agriculture Org.

Booth, M. A., Tucker, B. J., Allan, G. L., and Fielder, D. S. (2008). Effect of feeding regime and fish size on weight gain, feed intake and gastric evacuation in juvenile Australian snapper *Pagrus auratus*. Aquaculture, *282*(1-4):104-110.

Bray, J. R., and Curtis, J. T. (1957). An ordination of the upland forest communities of southern Wisconsin. Ecological Monographs, *27*(4): 325-349.

Breitburg, D. L., Steinberg, N., DuBeau, S., Cooksey, C., and Houde, E. D. (1994). Effects of low dissolved oxygen on predation on estuarine fish larvae. Marine Ecology Progress Series, 235-246.

Burns, A. R., Stephens, W. Z., Stagaman, K., Wong, S., Rawls, J. F., Guillemin, K., and Bohannan, B. J. (2016). Contribution of neutral processes to the assembly of gut microbial communities in the zebrafish over host development. The ISME journal, *10*(3), 655-664.

Cabello, F.C. et al. (2013) Antimicrobial use in aquaculture re- examined: its relevance to antimicrobial resistance and to animal and human health. Environmental Microbiology. *15*: 1917–1942

Cahill, M. M. (1990). Bacterial flora of fishes: a review. Microbial ecology, *19*(1), 21-41.

Candela, M., Consolandi, C., Severgnini, M., Biagi, E., Castiglioni, B., Vitali, B., and Brigidi, P. (2010). High taxonomic level fingerprint of the human intestinal microbiota by Ligase Detection Reaction-Universal Array approach. BMC Microbiology, *10*(1): 116.

Cao, Z., Simon, T., Wei, S. E., and Sheikh, Y. (2017). Realtime multi-person 2d pose estimation using part affinity fields. In Proceedings of the IEEE conference on computer vision and pattern recognition (pp. 7291-7299).

Caporaso, J. G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F. D., Costello, E. K., and Knight, R. (2010). QIIME allows analysis of high-throughput community sequencing data. Nature methods, *7*(5), 335-336.

Caporaso, J. G., Lauber, C. L., Walters, W. A., Berg-Lyons, D., Huntley, J., Fierer, N., and Gormley, N. (2012). Ultra-high-throughput microbial community analysis on the Illumina HiSeq and MiSeq platforms. The ISME journal, *6*(8): 1621-1624.

Caporaso, J. G., Lauber, C. L., Walters, W. A., Berg-Lyons, D., Lozupone, C. A., Turnbaugh, P. J., and Knight, R. (2011). Global patterns of 16S rRNA diversity at a depth of millions of sequences per sample. Proceedings of the national academy of sciences, *108*(Supplement 1), 4516-4522.

Ch'ng, C. L., and Senoo, S. (2008). Egg and larval development of a new hybrid grouper, tiger grouper *Epinephelus fuscoguttatus*× giant grouper E. lanceolatus. Aquaculture Science, *56*(4): 505-512.

Chakravorty, S., Helb, D., Burday, M., Connell, N., and Alland, D. (2007). A detailed analysis of 16S ribosomal RNA gene segments for the diagnosis of pathogenic bacteria. Journal of Microbiological Methods, *69*(2): 330-339.

Chao, A. (1984). Nonparametric estimation of the number of classes in a population. Scandinavian Journal of statistics, 265-270.

Chapman, D., and Kimstach, V. (1996). Selection of water quality variables. In: Chapman D(ed) Water quality assessments: a guide to the use of biota, sediments and water in environment monitoring. E. and FN. Spon, 59–126.

Chiew, I., Salter, A. M., and Lim, Y. S. (2019). The Significance of Major Viral and Bacterial Diseases in Malaysian Aquaculture Industry. Pertanika Journal of Tropical Agricultural Science, *42*(3).

Chu, K. B., Abdulah, A., Abdullah, S. Z., and Bakar, R. A. (2013). A case study on the mortality of cobia (Rachycentron canadum) cultured in traditional cages. Tropical life sciences research, *24*(2), 77.

Claesson, M. J., O'Sullivan, O., Wang, Q., Nikkilä, J., Marchesi, J. R., Smidt, H., and O'Toole, P.W. (2009). Comparative analysis of pyrosequencing and a phylogenetic microarray for exploring microbial community structures in the human distal intestine. PloS One, *4*(8).

Claesson, M. J., O'Sullivan, O., Wang, Q., Nikkilä, J., Marchesi, J. R., Smidt, H., and O'Toole, P. W. (2009). Comparative analysis of pyrosequencing and a phylogenetic microarray for exploring microbial community structures in the human distal intestine. PloS One, *4*(8), e6669.

Clarke, S. F., Murphy, E. F., O'Sullivan, O., Ross, R. P., O'Toole, P. W., Shanahan, F., and Cotter, P. D. (2013). Targeting the microbiota to address diet-induced obesity: a time dependent challenge. PloS One, *8*(6): e65790.

Clarke, S. F., Murphy, E. F., O'Sullivan, O., Lucey, A. J., Humphreys, M., Hogan, A., and Cotter, P. D. (2014). Exercise and associated dietary extremes impact on gut microbial diversity. Gut, *63*(12), 1913-1920.

Clarridge, J. E. (2004). Impact of 16S rRNA gene sequence analysis for identification of bacteria on clinical microbiology and infectious diseases. Clinical Microbiology Reviews, *17*(4): 840-862.

Compeau, P. E., Pevzner, P. A., and Tesler, G. (2011). How to apply de Bruijn graphs to genome assembly. Nature Biotechnology, *29*(11), 987-991.

Cornejo-Granados, F., Lopez-Zavala, A. A., Gallardo-Becerra, L., Mendoza-Vargas, A., Sánchez, F., Vichido, R., and Ochoa-Leyva, A. (2017). Microbiome of Pacific whiteleg shrimp reveals differential bacterial community composition between Wild, Aquacultured and AHPND/EMS outbreak conditions. Scientific Reports, *7*(1): 11783.

Dantas, Ê. W., Moura, A. N., and Bittencourt-Oliveira, M. D. C. (2011). Cyanobacterial blooms in stratified and destratified eutrophic reservoirs in semi-arid region of Brazil. Anais da Academia Brasileira de Ciências, *83*, 1327-1338.

Dassi, E., Ballarini, A., Covello, G., Quattrone, A., Jousson, O., De Sanctis, V., and Segata, N. (2014). Enhanced microbial diversity in the saliva microbiome induced by short-term probiotic intake revealed by 16S rRNA sequencing on the IonTorrent PGM platform. Journal of Biotechnology, 190:30-39.

de Bruijn, I., Liu, Y., Wiegertjes, G. F., and Raaijmakers, J. M. (2017). Exploring fish microbial communities to mitigate emerging diseases in aquaculture. FEMS Microbiol Ecology, *94*(1):161.

De Filippo, C., Ramazzotti, M., Fontana, P., and Cavalieri, D. (2012). Bioinformatic approaches for functional annotation and pathway inference in metagenomics data. Briefings in bioinformatics, *13*(6), 696-710.

Debroas, D., Humbert, J. F., Enault, F., Bronner, G., Faubladier, M., and Cornillot, E. (2009). Metagenomics approach studying the taxonomic and functional diversity of the bacterial community in a mesotrophic lake (Lac du Bourget–France). Environmental Microbiology, *11*(9): 2412-2424.

Defoirdt, T. et al. (2007) Alternatives to antibiotics to control bacterial infections: luminescent vibriosis in aquaculture as an example. Trends Biotechnology. *25*: 472–479

Dehler, C. E., Secombes, C. J., and Martin, S. A. (2017). Environmental and physiological factors shape the gut microbiota of Atlantic salmon parr (Salmo salar L.). Aquaculture, *467*:149-157.

Desai, A. R., Links, M. G., Collins, S. A., Mansfield, G. S., Drew, M. D., Van Kessel, A. G., and Hill, J. E. (2012). Effects of plant-based diets on the distal gut microbiome of rainbow trout (Oncorhynchus mykiss). Aquaculture, *350*: 134-142.

DeSantis, T. Z., Hugenholtz, P., Larsen, N., Rojas, M., Brodie, E. L., Keller, K., and Andersen, G. L. (2006). Greengenes, a chimera-checked 16S rRNA gene database and workbench compatible with ARB. Applied and environmental microbiology, *72*(7), 5069-5072.

Dhert, P., Rombaut, G., Suantika, G., and Sorgeloos, P. (2001). Advancement of rotifer culture and manipulation techniques in Europe. Aquaculture, *200*(1-2):129-146.

Di Maiuta, N., Schwarzentruber, P., Schenker, M., and Schoelkopf, J. (2013). Microbial population dynamics in the faeces of wood-eating loricariid catfishes. Letters in Applied Microbiology, *56*(6): 401-407.

Dimitroglou, A., Merrifield, D. L., Carnevali, O., Picchietti, S., Avella, M., Daniels, C., and Davies, S. J. (2011). Microbial manipulations to improve fish health and production–a Mediterranean perspective. Fish and Shellfish Immunology, *30*(1): 1-16.

Doan, Q. K., Vandeputte, M., Chatain, B., Morin, T., and Allal, F. (2017). Viral encephalopathy and retinopathy in aquaculture: a review. Journal of Fish Diseases, *40*(5), 717-742.

Dogan, A., Lasch, P., Neuschl, C., Millrose, M. K., Alberts, R., Schughart, K., and Brockmann, G. A. (2013). ATR-FTIR spectroscopy reveals genomic loci regulating the tissue response in high fat diet fed BXD recombinant inbred mouse strains. BMC Genomics, *14*(1): 386.

Dooley, J. J., Sage, H. D., Clarke, M. A. L., Brown, H. M., and Garrett, S. D. (2005). Fish species identification using PCR− RFLP analysis and lab-on-a-chip capillary electrophoresis: application to detect white fish species in food products and an interlaboratory study. Journal of agricultural and food chemistry, *53*(9), 3348-3357.

Duarte, L. N., Coelho, F. J., Oliveira, V., Cleary, D. F., Martins, P., and Gomes, N. C. (2019). Characterization of bacterioplankton communities from a hatchery recirculating aquaculture system (RAS) for juvenile sole (Solea senegalensis) production. PLoS One, *14*(1): e0211209.

Duncan, S. H., Lobley, G. E., Holtrop, G., Ince, J., Johnstone, A. M., Louis, P., and Flint, H. J. (2008). Human colonic microbiota associated with diet, obesity and weight loss. International Journal of Obesity, *32*(11), 1720-1724.

Dwyer, K. S., Brown, J. A., Parrish, C., and Lall, S. P. (2002). Feeding frequency affects food consumption, feeding pattern and growth of juvenile yellowtail flounder (*Limanda ferruginea*). Aquaculture, *213*(1-4): 279-292.

Edgar, R. C. (2010). Search and clustering orders of magnitude faster than BLAST. Bioinformatics, *26*(19), 2460-2461.

Eliku, T., and Leta, S. (2018). Spatial and seasonal variation in physicochemical parameters and heavy metals in Awash River, Ethiopia. Applied Water Science *8*(6): 177.

Erkus, O., de Jager, V. C., Geene, R. T., van Alen-Boerrigter, I., Hazelwood, L., van Hijum, S. A., and Smid, E. J. (2016). Use of propidium monoazide for selective profiling of viable microbial cells during Gouda cheese ripening. International Journal Of Food Microbiology, *228*, 1-9.

Escobar-Zepeda, A., Vera-Ponce de Leon, A., and Sanchez-Flores, A. (2015). The road to metagenomics: from microbiology to DNA sequencing technologies and bioinformatics. Frontiers in Genetics, *6*: 348.

Estruch, G., Collado, M. C., Peñaranda, D. S., Vidal, A. T., Cerdá, M. J., Martínez, G. P., and Martinez-Llorens, S. (2015). Impact of fishmeal replacement in diets for gilthead sea bream (*Sparus aurata*) on the gastrointestinal microbiota determined by pyrosequencing the 16S rRNA gene. PLoS One, *10*(8): e0136389.

Faith, J. J., Guruge, J. L., Charbonneau, M., Subramanian, S., Seedorf, H., Goodman, A. L., and Rosenbaum, M. (2013). The long-term stability of the human gut microbiota. Science, *341*(6141): 1237439.

Faruk, M. A. R., and Anka, I. Z. (2017). An overview of diseases in fish hatcheries and nurseries. Fundamental and Applied Agriculture, *2*(3): 311-316.

Faruk, M. A. R., and Anka, I. Z. (2017). An overview of diseases in fish hatcheries and nurseries. Fundamental and Applied Agriculture, *2*(3), 311-316.

FDA, Import Alert 16–131. (DWPE). Retrieved 25 August 2017, from USFDA. (2016).

Flimlin, G., Buttner, J., and Webster, D. (2008). Aquaculture systems for the northeast. Northeastern Regional Aquaculture. University of Maryland. Maryland–United States. Publication, (*104*), 1-7.

Franzosa, E. A., Hsu, T., Sirota-Madi, A., Shafquat, A., Abu-Ali, G., Morgan, X. C., and Huttenhower, C. (2015). Sequencing and beyond: integrating molecular'omics' for microbial community profiling. Nature Reviews Microbiology, *13*(6): 360.

Franzosa, E. A., Morgan, X. C., Segata, N., Waldron, L., Reyes, J., Earl, A. M., and Huttenhower, C. (2014). Relating the metatranscriptome and metagenome of the human gut. Proceedings of the National Academy of Sciences, *111*(22), E2329-E2338.

FRI. (2020). Short Term Storage of Groupher's Sperm. https://fri.dof.gov.my/fri.php/database_stores/store_view/83?print=1

Fuellen, G., Spitzer, M., Cullen, P., and Lorkowski, S. (2005). Correspondence of function and phylogeny of ABC proteins based on an automated analysis of 20 model protein data sets. Proteins, *61*(4): 888-899.

Funari, E., and Testai, E. (2008). Human health risk assessment related to cyanotoxins exposure. Critical reviews in toxicology, *38*(2), 97-125.

Gajardo, K., Rodiles, A., Kortner, T. M., Krogdahl, Å., Bakke, A. M., Merrifield, D. L., and Sørum, H. (2016). A high-resolution map of the gut microbiota in Atlantic salmon (*Salmo salar*): A basis for comparative gut microbial research. Scientific Reports, *6*: 30893.

Gao, X., Starmer, J., and Martin, E. R. (2008). A multiple testing correction method for genetic association studies using correlated single nucleotide polymorphisms. Genetic Epidemiology: The Official Publication of the International Genetic Epidemiology Society, *32*(4), 361-369.

Garrett, W. S., Gordon, J. I., and Glimcher, L. H. (2010). Homeostasis and inflammation in the intestine. Cell, *140*(6): 859-870.

Gevers, D., Pop, M., Schloss, P. D., and Huttenhower, C. (2012). Bioinformatics for the human microbiome project. PLoS Computational Biology, *8*(11): e1002779.

Gilbert, J. A., and Dupont, C. L. (2011). Microbial metagenomics: beyond the genome. Annual Reviews of Marine Science *3*: 347-371.

Gilbert, J. A., Meyer, F., Jansson, J., Gordon, J., Pace, N., Tiedje, J., and Knight, R. (2010). The earth microbiome project: meeting report of the “1 st EMP meeting on sample selection and acquisition” at Argonne National Laboratory October 6 th 2010. Standards in Genomic Sciences, *3*(3), 249-253.

Giovannoni, S. J., Britschgi, T. B., Moyer, C. L., and Field, K. G. (1990). Genetic diversity in Sargasso Sea bacterioplankton. Nature, *345*(6270): 60-63.

Givens, C. E., Ransom, B., Bano, N., and Hollibaugh, J. T. (2015). Comparison of the gut microbiomes of 12 bony fish and 3 shark species. Marine Ecology Progress Series, *518*: 209-223.

Goecks, J., Nekrutenko, A., and Taylor, J. (2010). Galaxy: a comprehensive approach for supporting accessible, reproducible, and transparent computational research in the life sciences. Genome Biology, *11*(8): R86.

Gohl, D. M., Vangay, P., Garbe, J., MacLean, A., Hauge, A., Becker, A., and Knights, D. (2016). Systematic improvement of amplicon marker gene methods for increased accuracy in microbiome studies. Nature Biotechnology, *34*(9): 942-949.

Greenwood, P. H. (1976). A review of the family Centropomidae (Pisces, Perciformes). Buletin of the British Museum (Natural History), *29*: 1- 81.

Guo, P., Zhang, K., Ma, X., and He, P. (2020). Clostridium species as probiotics: potentials and challenges. Journal of Animal Science and Biotechnology, *11*(1), 1-10.

Gupta, N., and Verma, V. K. (2019). Next-Generation Sequencing and Its Application: Empowering in Public Health Beyond Reality. In Microbial Technology for the Welfare of Society (pp. 313-341). Springer, Singapore.

Gupta, R. S. (2000). The phylogeny of proteobacteria: relationships to other eubacterial phyla and eukaryotes. FEMS Microbiology Reviews, *24*(4): 367-402.

Haas, B. J., Gevers, D., Earl, A. M., Feldgarden, M., Ward, D. V., Giannoukos, G., and Methé, B. (2011). Chimeric 16S rRNA sequence formation and detection in Sanger and 454-pyrosequenced PCR amplicons. Genome Research, *21*(3): 494-504.

Halpern, B. S., Walbridge, S., Selkoe, K. A., Kappel, C. V., Micheli, F., D'agrosa, C., and Fujita, R. (2008). A global map of human impact on marine ecosystems. Science, *319*(5865): 948-952.

Hammer, Ø., Harper, D. A., and Ryan, P. D. (2001). PAST: paleontological statistics software package for education and data analysis. Palaeontologia Electronica, *4*(1): 9.

Haughland, D. L., Hillman, A., and Azeria, E. T. (2018). Tackling rarity and sample bias with large-scale biodiversity monitoring: a case study examining the status, distribution and ecology of the lichen Cladonia rei in Alberta, Canada. The Lichenologist, *50*(2), 211-230.

Haugland, Ø., Mikalsen, A. B., Nilsen, P., Lindmo, K., Thu, B. J., Eliassen, T. M., and Evensen, Ø. (2011). Cardiomyopathy syndrome of Atlantic salmon (*Salmo salar L.*) is caused by a double-stranded RNA virus of the Totiviridae family. Journal of Virology, *85*(11): 5275-5286.

Hee, Y. Y., and Suratman, S. (2016). Physico-chemical parameters profile during dry and wet seasons in southern South China Sea: Brunei Bay. Chemistry – An Asian Journal *28*(10): 2146 - 2152.

Hennersdorf, P., Kleinertz, S., Theisen, S., Abdul-Aziz, M. A., Mrotzek, G., Palm, H. W., and Saluz, H. P. (2016). Microbial diversity and parasitic load in tropical fish of different environmental conditions. PLoS One, *11*(3), e0151594.

Hennersdorf, P., Mrotzek, G., Abdul-Aziz, M. A., and Saluz, H. P. (2016). Metagenomics analysis between free-living and cultured *Epinephelus fuscoguttatus* under different environmental conditions in Indonesian waters. Marine Pollution Buletin, *110*(2): 726-734.

Heuer, H., and Smalla, K. (1997). Application of denaturing gradient gel electrophoresis and temperature gradient gel electrophoresis for studying soil microbial communities. Modern soil microbiology., 353- 373.

Hewson, I., Paerl, R. W., Tripp, H. J., Zehr, J. P., and Karl, D. M. (2009). Metagenomics potential of microbial assemblages in the surface waters of the central Pacific Ocean tracks variability in oceanic habitat. Limnology and Oceanography, *54*(6): 1981-1994.

Hiergeist, A., Gläsner, J., Reischl, U., and Gessner, A. (2015). Analyses of intestinal microbiota: culture versus sequencing. Institute for Laboratory Animal Research, *56*(2), 228-240.

Holland, I. B., Cole, S. P., Kuchler, K., and Higgins, C. F. (Eds.). (2003). ABC proteins: from bacteria to man. Elsevier.

Huang, F., Pan, L., Lv, N., and Tang, X. (2017). Characterization of novel *Bacillus* strain N31 from mariculture water capable of halophilic heterotrophic nitrification–aerobic denitrification. Journal of Biosciences and Bioengineering, *124*(5): 564-571.

Huang, J., Yu, Z., and Chistoserdova, L. (2018). Lanthanide-dependent methanol dehydrogenases of XoxF4 and XoxF5 clades are differentially distributed among methylotrophic bacteria and they reveal different biochemical properties. Frontiers in microbiology, *9*, 1366.

Huse, S. M., Ye, Y., Zhou, Y., and Fodor, A. A. (2012). A core human microbiome as viewed through 16S rRNA sequence clusters. PloS one, 7(6): e34242.

Huson, D. H., and Weber, N. (2013). Microbial community analysis using MEGAN. Methods in enzymology, *531*, 465-485.

Huttenhower, C., Kostic, A. D., and Xavier, R. J. (2014). Inflammatory bowel disease as a model for translating the microbiome. Immunity, *40*(6), 843-854.

Ina-Salwany, M. Y., Al-saari, N., Mohamad, A., Mursidi, F. A., Mohd-Aris, A., Amal, M. N. A., and Zamri-Saad, M. (2019). Vibriosis in fish: a review on disease development and prevention. Aquatic Animal Health, *31*(1): 3-22.

Ingerslev, H. C., Strube, M. L., von Gersdorff Jørgensen, L., Dalsgaard, I., Boye, M., and Madsen, L. (2014). Diet type dictates the gut microbiota and the immune response against *Yersinia ruckeri* in rainbow trout (*Oncorhynchus mykiss*). Fish and Shellfish Immunology, *40*(2): 624-633.

Ingerslev, H. C., von Gersdorff Jørgensen, L., Strube, M. L., Larsen, N., Dalsgaard, I., Boye, M., and Madsen, L. (2014). The development of the gut microbiota in rainbow trout (*Oncorhynchus mykiss*) is affected by first feeding and diet type. Aquaculture, *424*: 24-34.

Islam, M. S., and Tanaka, M. (2004). Impacts of pollution on coastal and marine ecosystems including coastal and marine fisheries and approach for management: a review and synthesis. Marine Pollution Bulletin, *48*(7-8): 624-649.

Iwamoto, T., Mise, K., Takeda, A., Okinaka, Y., Mori, K. I., Arimoto, M., and Nakai, T. (2005). Characterization of Striped jack nervous necrosis virus subgenomic RNA3 and biological activities of its encoded protein B2. Journal Of General Virology, *86*(10), 2807-2816.

Janda, J. M., and Abbott, S. L. (2007). 16S rRNA gene sequencing for bacterial identification in the diagnostic laboratory: pluses, perils, and pitfalls. Journal of Clinical Microbiology, *45*(9): 2761-2764.

Jensen, A. R. (1965). Scoring the Stroop test. Acta Psychologica, *24*(5), 398-408.

Jovel, J., Patterson, J., Wang, W., Hotte, N., O'Keefe, S., Mitchel, T., and Wong, G. K. S. (2016). Characterization of the gut microbiome using 16S or shotgun metagenomics. Frontiers in Microbiology, *7*: 459.

Kaminski, J., Gibson, M. K., Franzosa, E. A., Segata, N., Dantas, G., and Huttenhower, C. (2015). High specificity targeted functional profiling in microbial communities with ShortBRED. PLoS Computational Biology, *11*(12): e1004557.

Kanehisa, M., Goto, S., Sato, Y., Kawashima, M., Furumichi, M., and Tanabe, M. (2013). Data, information, knowledge, and principle: back to metabolism in KEGG. Nucleic Acids Research, *42*(D1): D199-D205

Kara, H. M., Chaoui, L., Derbal, F., Zaidi, R., De Boisséson, C., Baud, M., and Bigarré, L. (2014). Betanodavirus-associated mortalities of adult wild groupers Epinephelus marginatus (Lowe) and Epinephelus costae (Steindachner) in Algeria. Journal of Fish Diseases, *37*(3): 273- 278.

Klindworth, A., Pruesse, E., Schweer, T., Peplies, J., Quast, C., Horn, M., and Glöckner, F. O. (2013). Evaluation of general 16S ribosomal RNA gene PCR primers for classical and next-generation sequencing-based diversity studies. Nucleic Acids Research, *41*(1): e1e1.

Knight, R., Jansson, J., Field, D., Fierer, N., Desai, N., Fuhrman, J. A., and Bailey, M. J. (2012). Unlocking the potential of metagenomics through replicated experimental design. Nature Biotechnology, *30*(6): 513.

Kohl, K. D., Weiss, R. B., Cox, J., Dale, C., and Denise Dearing, M. (2014). Gut microbes of mammalian herbivores facilitate intake of plant toxins. Ecology Letters, *17*(10), 1238-1246.

Kongtorp, R. T., Kjerstad, A., Taksdal, T., Guttvik, A., and Falk, K. (2004). Heart and skeletal muscle inflammation in Atlantic salmon, Salmo *salar L.*: a new infectious disease. Journal of Fish Diseases, *27*(6):351-358.

Krause, L., Diaz, N. N., Edwards, R. A., Gartemann, K. H., Krömeke, H., Neuweger, H., and Goesmann, A. (2008). Taxonomic composition and gene content of a methane-producing microbial community isolated from a biogas reactor. Journal of Biotechnology, *136*(1-2),91-101.

Kuczynski, J., Costello, E. K., Nemergut, D. R., Zaneveld, J., Lauber, C. L., Knights, D., and Knight, R. (2010). Direct sequencing of the human microbiome readily reveals community differences. Genome biology, *11*(5), 1-9.

Kuczynski, J., Lauber, C. L., Walters, W. A., Parfrey, L. W., Clemente, J. C., Gevers, D., and Knight, R. (2012). Experimental and analytical tools for studying the human microbiome. Nature Reviews Genetics, *13*(1) 47.

Kulabhusan, P. K., Rajwade, J. M., Sugumar, V., Taju, G., Hameed, A. S., and Paknikar, K. M. (2017). Field-usable lateral flow immunoassay for the rapid detection of white spot syndrome virus (WSSV). PloS one, *12*(1): e0169012.

Kumar, J., Kumar, M., Gupta, S., Ahmed, V., Bhambi, M., Pandey, R., and Chauhan, N. S. (2016). An improved methodology to overcome key issues in human fecal metagenomic DNA extraction. Genomics, Proteomics and Bioinformatics, *14*(6), 371-378.

Kumar, P. S., Brooker, M. R., Dowd, S. E., and Camerlengo, T. (2011). Target region selection is a critical determinant of community fingerprints generated by 16S pyrosequencing. PloS one, *6*(6): e20956.

Langille, M. G., Zaneveld, J., Caporaso, J. G., McDonald, D., Knights, D., Reyes, J. A., and Beiko, R. G. (2013). Predictive functional profiling of microbial communities using 16S rRNA marker gene sequences. Nature Biotechnology, *31*(9): 814.

Lazarevic, V., Whiteson, K., Huse, S., Hernandez, D., Farinelli, L., Østerås, M., and François, P. (2009). Metagenomic study of the oral microbiota by Illumina high-throughput sequencing. Journal of Microbiological Methods, *79*(3), 266-271.

Lee, C. S. (2003). Biotechnological advances in finfish hatchery production: a review. Aquaculture, *227*(1-4): 439-458.

Ley, R. E., Hamady, M., Lozupone, C., Turnbaugh, P. J., Ramey, R. R., Bircher, J. S., and Gordon, J. I. (2008). Evolution of mammals and their gut microbes. Science, *320*(5883), 1647-1651.

Ley, R. E., Peterson, D. A., and Gordon, J. I. (2006). Ecological and evolutionary forces shaping microbial diversity in the human intestine. Cell, *124*(4): 837-848.

Li, J., Quinque, D., Horz, H. P., Li, M., Rzhetskaya, M., Raff, J. A., and Stoneking, M. (2014). Comparative analysis of the human saliva microbiome from different climate zones: Alaska, Germany, and Africa. BMC Microbiology, *14*(1): 316.

Li, X. D., Liu, C. Q., Liu, X. L., and Bao, L. R. (2011). Identification of dissolved sulfate sources and the role of sulfuric acid in carbonate weathering using dual-isotopic data from the Jialing River, Southwest China.Journal of Asian Earth Sciences, *42*(3):370-380.

Liu, S., Yu, L. X., Singh, R. P., Jin, Y., Sorrells, M. E., and Anderson, J. A. (2010). Diagnostic and co-dominant PCR markers for wheat stem rust resistance genes Sr25 and Sr26. Theoretical and Applied Genetics, *120*(4), 691-697.

Liu, W. T., Marsh, T. L., Cheng, H., and Forney, L. J. (1997). Characterization of microbial diversity by determining terminal restriction fragment length polymorphisms of genes encoding 16S rRNA. Applied and environmental microbiology, *63*(11), 4516-4522.

Liu, Z., Lozupone, C., Hamady, M., Bushman, F. D., and Knight, R. (2008). Short pyrosequencing reads suffice for accurate microbial community analysis. Nucleic Acids Research, *35*(18), e120.

Llewellyn, M. S., Boutin, S., Hoseinifar, S. H., and Derome, N. (2014). Teleost microbiomes: the state of the art in their characterization, manipulation and importance in aquaculture and fisheries. Frontiers Microbiology, *5*:207.

Looi, C., C. and Senoo, S. (2008). Egg and larval development of a new hybrid grouper, tiger grouper *Epinephelus fuscoguttatus*× giant grouper *E. lanceolatus*. Aquaculture Science, *56*(4), 505-512.

López-Pérez, M., and Rodriguez-Valera, F. (2014). The family Alteromonadaceae. The Prokaryotes: Gammaproteobacteria, 69-92. Retrieved from https://link.springer.com/referenceworkentry/10.1007%2F978-3-642-38922-1_233

Lovatelli, A. (1990). Regional seafarming resources atlas.

Luo, C., Rodriguez-r, L. M., and Konstantinidis, K. T. (2014). MyTaxa: an advanced taxonomic classifier for genomic and metagenomics sequences. Nucleic Acids Research, *42*(8): e73- e73.

Luo, R., Liu, B., Xie, Y., Li, Z., Huang, W., Yuan, J., and Wang, J. (2012). SOAPdenovo2: an empirically improved memory-efficient short-read de novo assembler. Gigascience, 1(1), 2047-217X.

Luo, Y., Wang, T. T., Teng, Z., Chen, P., Sun, J., and Wang, Q. (2013). Encapsulation of indole-3-carbinol and 3, 3′-diindolylmethane in zein/carboxymethyl chitosan nanoparticles with controlled release property and improved stability. Food chemistry, *139*(1-4), 224-230.

Magoč, T., and Salzberg, S. L. (2011). FLASH: fast length adjustment of short reads to improve genome assemblies. Bioinformatics, *27*(21): 2957- 2963.

Mardis, E. R. (2008). Next-generation DNA sequencing methods. Annual Review of Genomics and Human Genetics, *9*: 387-402.

Mardis, E. R. (2008). The impact of next-generation sequencing technology on genetics. Trends in Genetics, *24*(3): 133-141.

Marte, C. L. (2003). Larviculture of marine species in Southeast Asia: current research and industry prospects. Aquaculture, *227*(1-4), 293-304.

Marte, C. L., and Toledo, J. D. (2015). Marine fish hatchery: developments and future trends in resource enhancement and sustainable aquaculture practices in Southeast Asia: challenge in responsible production of aquatic species: proceedings of the international workshop on resource enhancement and sustainable aquaculture practices in Southeast Asia 2014 (RESA) (pp. 189-200).

Mazmanian, S. K. (2008). Capsular polysaccharides of symbiotic bacteria modulate immune responses during experimental colitis. Journal of pediatric gastroenterology and nutrition, *46*(S1), E11-E12.

McHardy, I. H., Goudarzi, M., Tong, M., Ruegger, P. M., Schwager, E., Weger, J. R., and McGovern, D. P. (2013). Integrative analysis of the microbiome and metabolome of the human intestinal mucosal surface reveals exquisite inter-relationships. Microbiome, *1*(1): 17.

McLaughlin, J. B., DePaola, A., Bopp, C. A., Martinek, K. A., Napolilli, N. P., Allison, C. G., and Middaugh, J. P. (2005). Outbreak of Vibrio parahaemolyticus gastroenteritis associated with Alaskan oysters. New England Journal of Medicine, *353*(14), 1463-1470.

Meyer, F., and Paarmann, D. (2008). D0Souza M, Olson R, Glass EM, Kubal M, Paczian T, Rodriguez A, Stevens R, Wilke A, et al: The metagenomics RAST server-a public resource for the automatic phylogenetic and functional analysis of metagenomes. BMC Bioinformatics, *9*: 386.

M'gonigle, R. H. (1941). Acute catarrhal enteritis of salmonid fingerlings. Transactions of the American Fisheries Society, *70*(1): 297-303.

Miller, J. R., Hocking, A. M., Brown, J. D., and Moon, R. T. (1999). Mechanism and function of signal transduction by the Wnt/β-catenin and Wnt/Ca 2+ pathways. Oncogene, *18*(55), 7860-7872.

Moeller, A. H., Li, Y., Ngole, E. M., Ahuka-Mundeke, S., Lonsdorf, E. V., Pusey, A. E., and Ochman, H. (2014). Rapid changes in the gut microbiome during human evolution. Proceedings of the National Academy of Sciences, *111*(46), 16431-16435.

Moffitt CM, Cajas-Cano L. Blue growth: The 2014 FAO state of world fisheries and aquaculture. Fisheries, *39*(11):552-553

Mohamad, N., Amal, M. N. A., Yasin, I. S. M., Saad, M. Z., Shaqinah, N. N., Al-saari, N., and Sawabe, T. (2019). Vibriosis in cultured marine fishes: a review. Aquaculture, *512*, 734289.

Mohamad, N., Mohd Roseli, F. A., Azmai, M. N. A., Saad, M. Z., Md Yasin, I. S., Zulkiply, N. A., and Nasruddin, N. S. (2019). Natural concurrent infection of *Vibrio harveyi* and *V. alginolyticus* in cultured hybrid groupers in Malaysia. Journal of aquatic animal health, *31*(1), 88-96.

Mohan, C. V. (2007). Seed quality in freshwater fish production. FAO Fisheries Technical Paper (FAO).

Mohan, C. V., and Bondad-Reantaso, M. G. (2007). Seed quality in freshwater fish production.

Molinari, L. M., Scoaris, D. D. O., Pedroso, R. B., Bittencourt, N. L. R., Nakamura, C. V., Nakamura, T. U., and Dias, B. P. (2003). Bacterial microflora in the gastrointestinal tract of Nile tilapia, Oreochromis niloticus, cultured in a semi-intensive system. Acta Sci Biol Sci, *25*(2), 267-271.

Morán, A. C., Hengst, M. B., De la Iglesia, R., Andrade, S., Correa, J. A., and González, B. (2008). Changes in bacterial community structure associated with coastal copper enrichment. Environmental Toxicology and Chemistry: An International Journal, *27*(11): 2239-2245.

Morgan, X. C., and Huttenhower, C. (2012). Chapter 12: Human microbiome analysis. PLoS Computational Biology, *8*(12), e1002808.

Morgan, X. C., Tickle, T. L., Sokol, H., Gevers, D., Devaney, K. L., Ward, D. V., and Huttenhower, C. (2012). Dysfunction of the intestinal microbiome in inflammatory bowel disease and treatment. Genome biology, *13*(9), 1-18.

Munang'andu, H. M. (2016). Environmental viral metagenomics analyses in aquaculture: Applications in epidemiology and disease control. Frontiers in Microbiology, *7*:1986.

Munang'andu, H. M. (2016). Environmental viral metagenomics analyses in aquaculture: Applications in epidemiology and disease control. Frontiers in microbiology, *7*, 1986.

Munang'andu, H. M., and Evensen, Ø. (2017). Current advances in functional genomics in aquaculture. Applications of RNA-Seq and Omics Strategies-from Microorganisms to Human Health. IntechOpen, Rijeka, Croatia, 173-212.

Munang'andu, H. M., Mugimba, K. K., Byarugaba, D. K., Mutoloki, S., and Evensen, Ø. (2017). Current advances on virus discovery and diagnostic role of viral metagenomics in aquatic organisms. Frontiers in Microbiology, *8*: 406.

Munday, B. L., Kwang, J., and Moody, N. (2002). Betanodavirus infections of teleost fish: a review. Journal of Fish Diseases, *25*(3): 127-142.

Mustafa, S., Senoo, S., and Luin, M. (2013). Response of pure stock of coral reef tiger grouper and hybrid grouper to simulated ocean acidification. International Journal of Climate Change: Impacts and Responses, *5*(1).

Muyzer, G., De Waal, E. C., and Uitterlinden, A. G. (1993). Profiling of complex microbial populations by denaturing gradient gel electrophoresis analysis of polymerase chain reaction-amplified genes coding for 16S rRNA. Applied and Environmental Microbiology, *59*(3), 695-700.

Nakamura, K., Oshima, T., Morimoto, T., Ikeda, S., Yoshikawa, H., Shiwa, Y., and Altaf-Ul-Amin, M. (2011). Sequence-specific error profile of Illumina sequencers. Nucleic Acids Research, *39*(13): e90-e90.

Neefs, J. M., Van de Peer, Y., De Rijk, P., Chapelle, S., and De Wachter, R. (1993). Compilation of small ribosomal subunit RNA structures. Nucleic Acids Research, *21*(13): 3025-3049.

Neuman, C., Hatje, E., Zarkasi, K. Z., Smullen, R., Bowman, J. P., and Katouli, M. (2016). The effect of diet and environmental temperature on the faecal microbiota of farmed T asmanian Atlantic Salmon (*Salmo salar L*.). Aquaculture Research, *47*(2), 660-672.

Nie, L., Zhou, Q. J., Qiao, Y., and Chen, J. (2017). Interplay between the gut microbiota and immune responses of ayu (*Plecoglossus altivelis*) during *Vibrio anguillarum* infection. Fish and Shellfish Immunology, *68*, 479-487.

Noga, E. J. (2011). Fish disease: diagnosis and treatment. USA, John Wiley and Sons., 211 pp.

Nogales, B., Lanfranconi, M. P., Piña-Villalonga, J. M., and Bosch, R. (2011). Anthropogenic perturbations in marine microbial communities. FEMS Microbiology Reviews, *35*(2): 275-298.

Norman, J. M., Handley, S. A., and Virgin, H. W. (2014). Kingdom-agnostic metagenomics and the importance of complete characterization of enteric microbial communities. Gastroenterology, *146*(6), 1459-1469.

Novais, R. C., and Thorstenson, Y. R. (2011). The evolution of Pyrosequencing® for microbiology: from genes to genomes. Journal of microbiological methods, *86*(1) 1-7.

Nyrén, P. (1987). Enzymatic method for continuous monitoring of DNA polymerase activity. Analytical Biochemistry, *167*(2): 235-238.

Ochman, H., Worobey, M., Kuo, C. H., Ndjango, J. B. N., Peeters, M., Hahn, B. H., and Hugenholtz, P. (2010). Evolutionary relationships of wild hominids recapitulated by gut microbial communities. PLoS Biology, *8*(11), e1000546.

Octavia, S., and Lan, R. (2014). The family enterobacteriaceae. The Prokaryotes: Gammaproteobacteria, 225-286. Retrieved from https://link.springer.com/referenceworkentry/10.1007%2F978-3-642-38922-1_167

Olsen, I. (2014). The family fusobacteriaceae. The Prokaryotes: Firmicutes and Tenericutes, 109-132. Retrieved from https://www.researchgate.net/publication/287127551_The_Family_Fusobacteriaceae

Othman, M., Agüero, R., and Lin, H. C. (2008). Alterations in intestinal microbial flora and human disease. Current Opinion in Gastroenterology, *24*(1), 11-16.

Pakingking, R., Palma, P., and Usero, R. (2015). Quantitative and qualitative analyses of the bacterial microbiota of tilapia (Oreochromis niloticus) cultured in earthen ponds in the Philippines. World Journal of Microbiology and Biotechnology, *31*(2), 265-275.

Palacios, G., Lovoll, M., Tengs, T., Hornig, M., Hutchison, S., Hui, J., and Kristoffersen, A. B. (2010). Heart and skeletal muscle inflammation of farmed salmon is associated with infection with a novel reovirus. PLoS One, *5*(7): e11487.

Papanikos, N., Phelps, R. P., Davis, D. A., Ferry, A., and Maus, D. (2008). Spontaneous spawning of captive red snapper, *Lutjanus campechanus*, and dietary lipid effect on reproductive performance. Journal of the World Aquaculture Society, *39*(3): 324-338.

Pereira, C., Salvador, S., Arrojado, C., Silva, Y., Santos, A. L., Cunha, Â., and Almeida, A. (2011). Evaluating seasonal dynamics of bacterial communities in marine fish aquaculture: a preliminary study before applying phage therapy. Journal of environmental monitoring, *13*(4),1053-1058.

Petricoin III, E. F., Ardekani, A. M., Hitt, B. A., Levine, P. J., Fusaro, V. A., Steinberg, S. M., and Liotta, L. A. (2002). Use of proteomic patterns in serum to identify ovarian cancer. The Lancet, *359*(9306), 572-577.

Porchas-Cornejo, M. A., Martínez-Porchas, M., Vargas-Albores, F., Gollas-Galvan, T., Martínez-Córdova, L. R., Vazquez-Euan, R., and Peña-Messina, E. (2017). High-resolution detection of bacterial profile of ocean water, before and after being used by shrimp farms. Aquaculture International, *25*(5), 1833-1843.

Poretsky, R., Rodriguez-R, L. M., Luo, C., Tsementzi, D., and Konstantinidis, K. T. (2014). Strengths and limitations of 16S rRNA gene amplicon sequencing in revealing temporal microbial community dynamics. PloS One, *9*(4), e93827.

Port, J. A., Wallace, J. C., Griffith, W. C., and Faustman, E. M. (2012). Metagenomics profiling of microbial composition and antibiotic resistance determinants in Puget Sound. PloS One, *7*(10): e48000.

Programme UNE. Marine and coastal ecosystems and human well-being: A synthesis report based on the findings of the Millennium Ecosystem Assessment. UNEP. 2006:76

Quast, S., Berger, A., and Eberle, J. (2013). ROS-dependent phosphorylation of Bax by wortmannin sensitizes melanoma cells for TRAIL-induced apoptosis. Cell death and disease, 4(10), e839-e839.

Ram, R. J., VerBerkmoes, N. C., Thelen, M. P., Tyson, G. W., Baker, B. J., Blake, R. C., and Banfield, J. F. (2005). Community proteomics of a natural microbial biofilm. Science, *308*(5730): 1915-1920.

Ray, C. (2016). Characterization of the gut and skin microbiomes of wild-caught fishes from Lake Guntersville (Master's thesis). Auburn University, Alabama, United States.

Rhodes, M. A., and Phelps, R. P. (2008). Evaluation of the ciliated protozoa, *Fabrea salina* as a first food for larval red snapper, *Lutjanus campechanus* in a large-scale rearing experiment. Journal of Applied Aquaculture, *20*(2): 120-133.

Riesenfeld, C. S., Schloss, P. D., and Handelsman, J. (2004). Metagenomics: genomic analysis of microbial communities. Annual Reviews of Genetics, *38*: 525-552.

Ringø, E., Strøm, E., and Tabachek, J. A. (1995). Intestinal microflora of salmonids: a review. Aquaculture Research, *26*(10): 773-789.

Ringø, E., Zhou, Z., Vecino, J. G., Wadsworth, S., Romero, J., Krogdahl, Å., and Merrifield, D. L. (2016). Effect of dietary components on the gut microbiota of aquatic animals. A never-ending story? Aquaculture Nutrition, *22*(2), 219-282.

Ringø, E., Zhou, Z., Vecino, J. G., Wadsworth, S., Romero, J., Krogdahl, Å., and Owen, M. (2016). Effect of dietary components on the gut microbiota of aquatic animals. A never-ending story? Aquaculture Nutrition, *22*(2): 219-282.

Rodríguez Ayala, F., Bartolini, M., and Grau, R. (2020). The stress-responsive alternative sigma factor SigB of Bacillus subtilis and its relatives: An old friend with new functions. Frontiers in Microbiology, *11*, 1761.

Roeselers, G., Mittge, E. K., Stephens, W. Z., Parichy, D. M., Cavanaugh, C. M., Guillemin, K., and Rawls, J. F. (2011). Evidence for a core gut microbiota in the zebrafish. ISME Journal, *5*(10): 1595.

Roux, S., Enault, F., Robin, A., Ravet, V., Personnic, S., Theil, S., and Debroas, D. (2012). Assessing the diversity and specificity of two freshwater viral communities through metagenomics. PloS One, *7*(3): e33641.

Rud, I., Kolarevic, J., Holan, A. B., Berget, I., Calabrese, S., and Terjesen, B. F. (2017). Deep-sequencing of the bacterial microbiota in commercial- scale recirculating and semi-closed aquaculture systems for Atlantic salmon post-smolt production. Aquacultural Engineering, *78*, 50-62.

Rusch, D. B., Halpern, A. L., Sutton, G., Heidelberg, K. B., Williamson, S., Yooseph, S.,and Beeson, K. (2007). The Sorcerer II global ocean sampling expedition: northwest Atlantic through eastern tropical Pacific. PLoS Biology, *5*(3): e77.

Sadovy, Y. (2000). Regional survey for fry/fingerling supply and current practices for grouper mariculture: evaluating current status and long- term rospects for grouper mariculture in South East Asia. Final report to the collaborative APEC Grouper Research and Development Network (FWG 01/99).

Salinas, I., Myklebust, R., Esteban, M. A., Olsen, R. E., Meseguer, J., and Ringø, E. (2008). In vitro studies of Lactobacillus delbrueckii subsp. lactis in Atlantic salmon (*Salmo salar L.*) foregut: tissue responses and evidence of protection against *Aeromonas salmonicida* subsp. *salmonicida* epithelial damage. Veterinary Microbiology, *128*(1- 2): 167-177.

Salipante, S. J., Kawashima, T., Rosenthal, C., Hoogestraat, D. R., Cummings, L. A., Sengupta, D. J., and Hoffman, N. G. (2014). Performance comparison of Illumina and ion torrent next-generation sequencing platforms for 16S rRNA-based bacterial community profiling. Applied and Environmental Microbiology, *80*(24): 7583-7591.

Sanger, F., and Coulson, A. R. (1975). A rapid method for determining sequences in DNA by primed synthesis with DNA polymerase. Journal of Molecular Biology, *94*(3): 441-448.

Schaubeck, M., Clavel, T., Calasan, J., Lagkouvardos, I., Haange, S. B., Jehmlich, N., and Bleich, A. (2016). Dysbiotic gut microbiota causes transmissible Crohn's disease-likeileitis independent of failure in antimicrobial defence. Gut, *65*(2): 225-237.

Schloss, P. D., Westcott, S. L., Ryabin, T., Hall, J. R., Hartmann, M., Hollister, E. B., and Weber, C. F. (2009). Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. Applied and environmental microbiology, *75*(23), 7537-7541.

Schmidt, V. T., Smith, K. F., Melvin, D. W., and Amaral-Zettler, L. A. (2015). Community assembly of a euryhaline fish microbiome during salinity acclimation. Molecular Ecology, *24*(10): 2537-2550.

Segata, N., Izard, J., Waldron, L., Gevers, D., Miropolsky, L., Garrett, W. S., and Huttenhower, C. (2011). Metagenomics biomarker discovery and explanation. Genome Biology, *12*(6): R60.

Senanan, W., Pechsiri, J., Sonkaew, S., Na-Nakorn, U., Sean-In, N., and Yashiro, R. (2015). Genetic relatedness and differentiation of hatchery populations of Asian seabass (*Lates calcarifer*) (Bloch, 1790) broodstock in Thailand inferred from microsatellite genetic markers. Aquaculture Research, *46*(12), 2897-2912.

Seng, L. T. (2002). Practical approaches to health management for cage cultured marine fishes. Aquaculture Asia, *7*(3), 42-48.

Shannon, C. E. (1948). A mathematical theory of communication. The Bell System Technical Journal, *27*(3), 379-423.

Shields, R. J., Kotani, T., Molnar, A., Marion, K., Kobashigawa, J., and Tang, L. (2005). Intensive cultivation of a subtropical paracalanid copepod, *Parvocalanus* sp., as prey for small marine fish larvae. Copepods in Aquaculture, 209-223.

Silva, C. R., Gomes, L. C., and Brandão, F. R. (2007). Effect of feeding rate and frequency on tambaqui (Colossoma macropomum) growth, production and feeding costs during the first growth phase in cages. Aquaculture, *264*(1-4): 135-139.

Simpson, E. H. (1949). Measurement of diversity. Nature, *163*(4148), 688-688.

Sims, D., Sudbery, I., Ilott, N. E., Heger, A., and Ponting, C. P. (2014). Sequencing depth and coverage: key considerations in genomic analyses. Nature Reviews Genetics, *15*(2): 121.

Smith, P., Willemsen, D., Popkes, M., Metge, F., Gandiwa, E., Reichard, M., and Valenzano, D. R. (2017). Regulation of life span by the gut microbiota in the short-lived African turquoise killifish. Elife, *6*, e27014.

Spaniolas, S., May, S. T., Bennett, M. J., and Tucker, G. A. (2006). Authentication of coffee by means of PCR-RFLP analysis and lab-on-a-chip capillary electrophoresis. Journal of Agricultural And Food Chemistry, *54*(20), 7466-7470.

Sullam, K. E., Essinger, S. D., Lozupone, C. A., O'Connor, M. P., Rosen, G. L., Knight, R. O. B., and Russell, J. A. (2012). Environmental and ecological factors that shape the gut bacterial communities of fish: a meta-analysis. Molecuar Ecology, *21*(13): 3363-3378.

Sun, Y., Yang, H., Ling, Z., Chang, J., and Ye, J. (2009). Gut microbiota of fast and slow growing grouper *Epinephelus coioides*. African Journal of Microbiology Research, *3*(11): 637-640.

Sundquist, A., Bigdeli, S., Jalili, R., Druzin, M. L., Waller, S., Pullen, K. M., and Ronaghi, M. (2007). Bacterial flora-typing with targeted, chip-based Pyrosequencing. BMC Microbiology, *7*(1), 1-11.

Tanner, A., Maiden, M. F., Paster, B. J., and Dewhirst, F. E. (1994). The impact of 16S ribosomal RNA-based phylogeny on the taxonomy of oral bacteria. Periodontology 2000, *5*(1): 26-51.

Tarnecki, A. M., Burgos, F. A., Ray, C. L., and Arias, C. R. (2017). Fish intestinal microbiome: diversity and symbiosis unravelled by metagenomics. Journal of Applied Microbiology, *123*(1): 2-17.

Thomas, F., Hehemann, J. H., Rebuffet, E., Czjzek, M., and Michel, G. (2011). Environmental and gut bacteroidetes: the food connection. Frontiers in Microbiology, *2*, 93.

Thomas, F., Hehemann, J. H., Rebuffet, E., Czjzek, M., and Michel, G. (2011). Environmental and gut bacteroidetes: the food connection. Frontiers Microbiology, *2*: 93.

Thukral, R., Shrivastav, K., Mathur, V., Barodiya, A., and Shrivastav, S. (2017). Actinomyces: A deceptive infection of oral cavity. Journal of the Korean Association of Oral and Maxillofacial Surgeons, *43*(4), 282-285.

Toranzo, A. E., Magariños, B., and Romalde, J. L. (2005). A review of the main bacterial fish diseases in mariculture systems. Aquaculture, *246*(1-4): 37-61.

Tothill, R. W., Tinker, A. V., George, J., Brown, R., Fox, S. B., Lade, S., and Australian Ovarian Cancer Study Group. (2008). Novel molecular subtypes of serous and endometrioid ovarian cancer linked to clinical outcome. Clinical Cancer Research, *14*(16), 5198-5208.

Tremblay, J., Singh, K., Fern, A., Kirton, E. S., He, S., Woyke, T., and Tringe, S. G. (2015). Primer and platform effects on 16S rRNA tag sequencing. Frontiers in Microbiology, *6*: 771.

Tseng, C. H., Chiang, P. W., Shiah, F. K., Chen, Y. L., Liou, J. R., Hsu, T. C., and Tang, S. L. (2013). Microbial and viral metagenomes of a subtropical freshwater reservoir subject to climatic disturbances. The ISME Journal, *7*(12): 2374.

Tucker, B. J., Booth, M. A., Allan, G. L., Booth, D., and Fielder, D. S. (2006). Effects of photoperiod and feeding frequency on performance of newly weaned Australian snapper *Pagrus auratus*. Aquaculture, *258*(1-4) 514-520.

Tuomisto, H. (2010). A diversity of beta diversities: straightening up a concept gone awry. Part 1. Defining beta diversity as a function of alpha and gamma diversity. Ecography, *33*(1), 2-22.

Tuomisto, H., and Ruokolainen, K. (2006). Analyzing or explaining beta diversity? Understanding the targets of different methods of analysis. Ecology, *87*(11), 2697-2708.

Turnbaugh, P. J., Hamady, M., Yatsunenko, T., Cantarel, B. L., Duncan, A., Ley, R. E., and Gordon, J. I. (2009). A core gut microbiome in obese and lean twins. Nature, *457*(7228), 480-484.

van Elsas, J. D., Costa, R., Jansson, J., Sjöling, S., Bailey, M., Nalin, R., and Van Overbeek, L. (2008). The metagenomics of disease-suppressive soils–experiences from the METACONTROL project. Trends in Biotechnology, *26*(11): 591-601.

Vasconcelos, V. (2006). Eutrophication, toxic cyanobacteria and cyanotoxins: when ecosystems cry for help. Limnetica, *25*(1-2): 425-432.

Verschuere, L., Rombaut, G., Sorgeloos, P., and Verstraete, W. (2000). Probiotic bacteria as biological control agents in aquaculture. Microbiology and Molecular Biology Reviews, *64*(4), 655-671.

Wagner, A. O., Malin, C., Knapp, B. A., and Illmer, P. (2008). Removal of free extracellular DNA from environmental samples by ethidium monoazide and propidium monoazide. Applied and Environmental Microbiology, *74*(8), 2537-2539.

Walsh, A. M., Crispie, F., O'Sullivan, O., Finnegan, L., Claesson, M. J., and Cotter, P. D. (2018). Species classifier choice is a key consideration when analysing low-complexity food microbiome data. Microbiome, *6*(1): 50.

Wang, Q., Garrity, G. M., Tiedje, J. M., and Cole, J. R. (2007). Naive Bayesian classifier for rapid assignment of rRNA sequences into the new bacterial taxonomy. Applied and Environmental Microbiology, *73*(16), 5261-5267.

Wang, X., Sun, G., Feng, T., Zhang, J., Huang, X., Wang, T., and Geng, M. (2019). Sodium oligomannate therapeutically remodels gut microbiota and suppresses gut bacterial amino acids-shaped neuroinflammation to inhibit Alzheimer's disease progression. Cell Research, *29*(10), 787-803.

Wen, L., Ley, R. E., Volchkov, P. Y., Stranges, P. B., Avanesyan, L., Stonebraker, A. C., and Chervonsky, A. V. (2008). Innate immunity and intestinal microbiota in the development of Type 1 diabetes. Nature, *455*(7216), 1109-1113.

Wilmes, P., Wexler, M., and Bond, P. L. (2008). Metaproteomics provides functional insight into activated sludge wastewater treatment. PLoS One, *3*(3), e1778.

Wolf, K., Snieszko, S. F., Dunbar, C. E., and Pyle, E. (1960). Virus nature of infectious pancreatic necrosis in trout. Proceedings of the Society for Experimental Biology and Medicine, *104*(1): 105-108.

Wu, S., Wang, G., Angert, E. R., Wang, W., Li, W., and Zou, H. (2012). Composition, diversity, and origin of the bacterial community in grass carp intestine. PLoS One, *7*(2): e30440.

Xia, J. H., Lin, G., Fu, G. H., Wan, Z. Y., Lee, M., Wang, L., and Yue, G. H. (2014). The intestinal microbiome of fish under starvation. BMC Genomics, *15*(1): 266.

Xu, H., Yang, J., Gao, W., Li, L., Li, P., Zhang, L., and Shao, F. (2014). Innate immune sensing of bacterial modifications of Rho GTPases by the Pyrin inflammasome. Nature, *513*(7517), 237-241.

Xu, J., Zhu, L., He, S., Wu, Y., Jin, W., Yu, T., and Wen, Z. (2015). Temporal-spatial resolution fate mapping reveals distinct origins for embryonic and adult microglia in zebrafish. Developmental cell, *34*(6), 632-641.

Xu, Q., Black, W. P., Nascimi, H. M., and Yang, Z. (2011). DifA, a methyl-accepting chemoreceptor protein-like sensory protein, uses a novel signaling mechanism to regulate exopolysaccharide production in *Myxococcus xanthus*. Journal of Bacteriology, *193*(3): 759-767.

Xu, S., Panikker, P., Iqbal, S., and Elefant, F. (2016). Tip60 HAT action mediates environmental enrichment induced cognitive restoration. PloS One, *11*(7), e0159623.

Xu, Z., Jiang, Y., Te, S., He, Y., and Gin, K. (2018). The Effects of Antibiotics on Microbial Community Composition in an Estuary Reservoir during Spring and Summer Seasons. Water, *10*(2): 154.

Ye, L., Amberg, J., Chapman, D., Gaikowski, M., and Liu, W. T. (2014). Fish gut microbiota analysis differentiates physiology and behavior of invasive Asian carp and indigenous American fish. The ISME Journal, *8*(3): 541.

Yeh SL, Kuo CM, Ting YY, Chang CF (2003). Androgens stimulate sex change in protogynous grouper, *Epinephelus coioides*: spawning performance in sex-changed males. Comparative biochemistry and Physiology – Part C: Toxicology, *135C*: 375-382.

Yokoyama, S., Koshio, S., Takakura, N., Oshida, K., Ishikawa, M., Gallardo-Cigarroa, F. J.and Teshima, S. I. (2006). Effect of dietary bovine lactoferrin on growth response, tolerance to air exposure and low salinity stress conditions in orange spotted grouper *Epinephelus coioides*. Aquaculture, *255*(1): 507-513.

Yoon, S. S., Kim, E. K., and Lee, W. J. (2015). Functional genomic and metagenomic approaches to understanding gut microbiota–animal mutualism. Current Opinion in Microbiology, *24*, 38-46.

Yue, G. H., Xia, J. H., Liu, F., and Lin, G. (2012). Evidence for female-biased dispersal in the protandrous hermaphroditic Asian seabass, *Lates calcarifer*. PloS One, *7*(6), e37976.

Yusoff, A. (2015). Status of resource management and aquaculture in Malaysia.

Zaneveld, J. R., Lozupone, C., Gordon, J. I., and Knight, R. (2010). Ribosomal RNA diversity predicts genome diversity in gut bacteria and their relatives. Nucleic acids research*, 38*(12), 3869-3879.

Zhang, J., Kobert, K., Flouri, T., and Stamatakis, A. (2013). PEAR: a fast and accurate Illumina Paired-End reAd mergeR. Bioinformatics, *30*(5): 614-620.

Zhang, M., Sun, Y., Liu, Y., Qiao, F., Chen, L., Liu, W. T., and Li, E. (2016). Response of gut microbiota to salinity change in two euryhaline aquatic animals with reverse salinity preference. Aquaculture, *454*: 72-80.

Zoetendal, E. G., Akkermans, A. D., Akkermans-van Vliet, W. M., de Visser, J. A. G., and de Vos, W. M. (2001). The host genotype affects the bacterial community in the human gastronintestinal tract. Microbial Ecology in Health and Disease, *13*(3), 129-134.