ASSESSMENT OF Piper betle L. METHANOLIC EXTRACT AS A POTENTIAL GROWTH AND HEALTH PROMOTER FOR JUVENILE RED HYBRID TILAPIA (Oreochromis sp.)

SYAHIDAH AHMAD

FPV 2019 16

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By

SYAHIDAH AHMAD

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirements for the Degree of Doctor of Philosophy

July 2018

DEDICATION

To my supportive husband Muhamad Amir,
who has sacrificed so much and comprehend my emotions

To my beloved parents and mother in-law,
who always kept praying for me to achieve my goal

To my lovely sister and brothers,
who always believed in me and loved me unconditionally

To my kind-hearted supervisor,
who has encouraged me all the way of my study

Abstract of the thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Doctor of Philosophy

ASSESSMENT OF *Piper betle* L. METHANOLIC EXTRACT AS A POTENTIAL GROWTH AND HEALTH PROMOTER FOR JUVENILE RED HYBRID TILAPIA (*Oreochromis* sp.)

By

SYAHIDAH BINTI AHMAD

July 2018

Chairman : Hassan Hj. Mohd Daud, PhD
Faculty : Veterinary Medicine

Tilapia is listed as the most widely cultured fish in many parts of the world. The rising global demand of tilapia has driven significant efforts for the improvement of the growth and health of this species. Various antibiotics, growth promoters, and chemotherapeutic agents have been used for the above purposes. However, they tend to bring undesirable side effects. The use of herbs has been suggested as an alternative strategy to the use of conventional agents. Therefore, the present study aims to select safe herbal extracts with potent antibacterial properties as dietary supplementation for juvenile red hybrid tilapia (*Oreochromis* sp.). Five Malaysian local herbal extracts of methanol and aqueous i.e. *Cosmos caudatus* (ulam Raja), *Curcuma mangga* (temumangga), *Justicia gendarussa* (gandarusa), *Piper betle* (sireh), and *Zingiber zerumbet* (lempoyang) were screened for antibacterial activities against nine common fish pathogenic bacteria using agar-well diffusion method. The assay showed methanolic extracts could inhibit all of bacteria tested namely *Bacillus* sp., *Enterococcus feacalis*, *Staphylococcus aureus*, *Streptococcus agalactiae*, *Aeromonas hydrophila*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Vibrio alginolyticus* with higher inhibitions compared to aqueous extracts. The aqueous extracts showed inhibition only towards *S. aureus*, *S. agalactiae*, *A. hydrophila*, and *V. alginolyticus.* Due to low antibacterial activities, aqueous extracts were omitted for further evaluation, which included the methanolic extracts of *C. mangga* and *J. gendarussa*. Comparative inhibitory activities between selected methanolic extracts of herbs and standard antibiotics revealed the ability of the herbs as potent antibacterial agent are presented in the following descending order of effectiveness: enrofloxacin, *P. betle*, oxytetracycline, *Z. zerumbet*, tetracycline, *C. caudatus*, and erythromycin. Methanolic extracts of *P. betle*, *Z. zerumbet*, and *C. caudatus* were investigated for *in vivo* toxicity. The median lethal doses (LD_{50}) of the extracts administered intraperitoneally to *Oreochromis* sp. juveniles were determined to be 310.97 mg/mL (*P. betle*), 227.53 mg/mL (*Z. zerumbet*), and 234.42 mg/mL (*C. caudatus*). Although all of herbal extracts yielded low mortality, organ histology revealed moderate pathological changes induced by *C. caudatus* and *Z. zerumbet* that increased proportionally with the dose given. Consequently, *C. caudatus* and *Z. zerumbet* were discarded for next assessment due to their toxicity effects. Qualitative phytochemical

screening was carried out for *P. betle* methanolic extract (PBME) prior to the identification and quantification of its major antibacterial active compounds. The phytochemical analysis showed the presence of alkaloids, phenols, flavonoids, tannins, saponins, glycosides, terpenoids, and steroids. The TLC-agar-overlay bioautography assay identified two major compounds i.e. hydroxychavicol and eugenol as the responsible compounds for antibacterial activities. The content of hydroxychavicol and eugenol in PBME were found to be 374.72±2.79 mg/g and 49.67±0.16 mg/g as quantitatively analysed by HPLC. Feeding trial was done in order to determine the effects of PBME on growth performance, blood parameters, and histology of organs of *Oreochromis* sp. PBME was supplemented in the diet at 0.20 g/kg (T1), 0.78 g/kg (T2), and 3.13 g/kg (T3) containing 37.47% and 4.97% of hydroxychavicol and eugenol, and fed to *Oreochromis* sp. for 12 weeks. The fish fed with 0.20 g/kg PBME had the best performance, and showed significant improvement in weight gain (616.23%), body length gain (6.87 cm), specific growth rate (2.34%/day), protein efficiency ratio (1.97), and feed conversion ratio (1.48). Haematological analysis showed higher trend in RBC, Hb, and Hct values, with lower WBC value in PBME-fed fish compared to control. Total serum protein, albumin, and globulin contents were improved particularly with the supplementation of PBME at 0.78 g/kg. Histological findings also revealed no adverse effects of the PBME supplementations on fish spleen and intestine. The effects of dietary supplementation of PBME in *Oreochromis* sp. were then assessed on survival and resistance to *S. agalactiae*. In 14 days of *S. agalactiae* post-challenge, survival and RPS of *Oreochromis* sp. fed with PBME was higher than untreated-control group, but not significantly different compared to OTC-treated group. The results showed PBME-treated fish had a high resistance to disease, and the efficacy of PBME as prophylaxis was equivalent to OTC antibiotic. The supplementation of PBME at 0.78 g/kg gave the best result, and it could restore the altered haematological parameters and serum protein contents in infected fish to near normal values. Histological analysis showed the degrees of damage were lesser in infected organs of PBME-treated fish compared to untreated-control fish. The results revealed the potential of PBME to ameliorate the organ damages induced by *S. agalactiae*. In conclusion, the current study suggests that *Piper betle* methanolic extract is effective as a growth and health promoter in *Oreochromis* sp.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

PENILAIAN EKSTRAK METANOL *Piper betle* L. SEBAGAI POTENSI PENGGALAK PERTUMBUHAN DAN KESIHATAN BAGI JUVENIL TILAPIA HIBRID MERAH (*Oreochromis* sp.)

Oleh

SYAHIDAH BINTI AHMAD

Julai 2018

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Tilapia disenaraikan sebagai ikan yang paling banyak dikultur di kebanyakan tempat di dunia. Permintaan tilapia yang semakin meningkat secara global telah mendorong usaha-usaha penting bagi meningkatkan pertumbuhan dan kesihatan spesies ini. Pelbagai antibiotik, penggalak pertumbuhan, dan agen kemoterapi telah digunakan untuk tujuan tersebut. Walau bagaimanapun, ia cenderung membawa kepada kesan-kesan sampingan yang tidak diingini. Penggunaan herba disyorkan sebagai strategi alternatif kepada penggunaan agen konvensional. Oleh itu, kajian ini bertujuan untuk memilih ekstrak-ekstrak herba yang selamat dengan ciri-ciri antibakteria yang kuat untuk dijadikan penambah makanan untuk juvenil tilapia hibrid merah (*Oreochromis* sp.). Lima ekstrak metanol dan ekstrak berair herba tempatan Malaysia iaitu *Cosmos caudatus* (ulam Raja), *Curcuma mangga* (temumangga), *Justicia gendarussa* (gandarusa), *Piper betle* (sireh), dan *Zingiber zerumbet* (lempoyang) telah disaring untuk aktiviti antibakteria terhadap bakteria ikan patogenik yang lazim dengan menggunakan kaedah resapan telaga-agar. Asai menunjukkan bahawa ekstrak metanol telah merencatkan kesemua bakteria yang diuji iaitu *Bacillus* sp., *Enterococcus feacalis*, *Staphylococcus aureus*, *Streptococcus agalactiae*, *Aeromonas hydrophila*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, dan *Vibrio alginolyticus* dengan perencatan yang lebih tinggi berbanding dengan ekstrak berair. Ekstrak berair hanya menunjukkan perencatan terhadap *S. aureus*, *S. agalactiae*, *A. hydrophila*, dan *V. alginolyticus*. Disebabkan oleh aktiviti antibakteria yang rendah, ekstrak berair disingkirkan untuk penilaian lanjut, termasuklah ekstrak metanol *C. mangga* dan *J. gendarussa*. Perbandingan aktiviti perencatan di antara ekstrak metanol herba terpilih dan antibiotik piawai mennunjukkan keupayaan herba sebagai agen antibakteria yang kuat, diperlihatkan di dalam urutan menurun berikut: enrofloxacin, *P. betle*, oxytetracycline, *Z. zerumbet*, tetracycline, *C. caudatus*, dan erythromycin. Ekstrak metanol *P. betle*, *Z. zerumbet*, dan *C. caudatus* dikaji untuk ketoksikan *in vivo*. Nilai dos kematian purata (LD_{50}) ekstrak yang diberikan secara suntikan intraperitoneum pada juvenil *Oreochromis* sp. bersamaan dengan 310.97 mg/mL (*P. betle*), 227.53 mg/mL (*Z. zerumbet*), dan 234.42 mg/mL (*C. caudatus*). Walaupun kesemua ekstrak herba memberikan hasil kematian yang rendah, histologi organ menunjukkan terdapat perubahan patologi sederhana yang disebabkan oleh *C. caudatus*

dan *Z. zerumbet*, yang mana berkadar langsung dengan peningkatan dos. Oleh itu, *C. caudatus* dan *Z. zerumbet* dikecualikan untuk penilaian seterusnya kerana kesan ketoksikan yang wujud. Pemeriksaan fitokimia kualitatif telah dijalankan terhadap ekstrak metanol *P. betle* (PBME) sebelum pengenalpastian dan kuantifikasi sebatian antibakteria aktif utama. Analisis fitokimia menunjukkan kehadiran alkaloid, fenol, flavonoid, tanin, saponin, glikosid, terpenoid, dan steroid. Asai TLC-bioautografi agar terlapis telah mengenalpasti dua sebatian utama iaitu hydroxychavicol dan eugenol sebagai sebatian yang bertanggungjawab untuk aktiviti antibakteria. Kandungan hydroxychavicol dan eugenol dalam PBME didapati sebanyak 374.72±2.79 mg/g dan 49.67±0.16 mg/g, seperti yang dianalisis secara kuantitatif oleh HPLC. Ujian pemakanan telah dijalankan bagi menentukan kesan penambahan PBME terhadap prestasi pertumbuhan, parameter darah, dan histologi organ juvenil *Oreochromis* sp. PBME ditambah dalam diet pada 0.20 g/kg (T1), 0.78 g/kg (T2), dan 3.13 g/kg (T3) mengandungi 37.47% dan 4.97% sebatian hydroxychavicol dan eugenol, dan seterusnya diberi makan kepada *Oreochromis* sp. selama 12 minggu. Ikan yang diberi makan dengan 0.20 g/kg PBME menunjukkan prestasi terbaik, dan perubahan yang signifikan pada pertambahan berat badan (616.23%), panjang badan (6.87 cm), kadar pertumbuhan spesifik (2.34%/hari), nisbah kecekapan protein (1.97), dan nisbah penukaran makanan (1.48). Analisis hematologi menunjukkan kadar yang lebih tinggi dalam nilai RBC, Hb, dan Hct, dengan nilai WBC yang rendah dalam ikan diberi makan PBME berbanding kawalan. Kandungan protein serum, albumin, dan globulin didapati meningkat terutamanya dengan penambahan PBME pada 0.78 g/kg. Penemuan histologi juga menunjukkan tiada kesan sampingan buruk penambahan PBME terhadap limpa dan usus ikan. Kesan penambahan makanan PBME dalam *Oreochromis* sp. kemudian dinilai terhadap kemandirian dan rintangan terhadap *S. agalactiae*. Dalam 14 hari pasca-cabaran *S. agalactiae*, kemandirian dan RPS *Oreochromis* sp. diberi makan PBME adalah lebih tinggi daripada kumpulan kawalan yang tidak dirawat, tetapi tidak berbeza berbanding dengan kumpulan yang menerima OTC. Keputusan ini menunjukkan ikan yang menerima PBME mempunyai rintangan yang tinggi terhadap penyakit, dan keberkesanan PBME sebagai profilaksis adalah setara dengan antibiotik OTC. Penambahan PBME pada 0.78 g/kg memberikan hasil yang terbaik, dan dapat mengembalikan parameter haematologi dan kandungan protein serum dalam ikan yang dijangkiti kepada nilai hampir normal. Analisis histologi menunjukkan tahap kerosakan adalah lebih rendah dalam organ-organ ikan yang dijangkiti dirawat-PBME berbanding dengan ikan kawalan yang tidak dirawat. Keputusan ini menunjukkan PBME berpotensi untuk memulihkkan kerosakan organ yang disebabkan oleh *S. agalactiae*. Kesimpulannya, kajian ini mencadangkan bahawa ekstrak methanol *Piper betle* adalah berkesan sebagai penggalak pertumbuhan dan kesihatan kepada *Oreochromis* sp.

ACKNOWLEDGEMENTS

بِسْمِ ٱللَّهِ ٱلرَّحْمَٰنِ ٱلرَّحِيمِ

First and foremost, praises and gratitude to Allah S.W.T., the Almighty, for giving me the strength and courage to complete this thesis. Without His blessing, this achievement would be impossible. In this opportunity, I would like to express my gratitude to all people whom in one way or another contributed in the completion of this thesis.

This feeling of grateful and appreciation goes to my supervisor, Assoc. Prof. Dr. Hassan Hj. Mohd. Daud for his great supervision, constant guidance, suggestions, immense knowledge, and understanding. Without his good guidance and assistance, this thesis would not have been in this form. Thanks for the patience, unwavering support both morally and financially, motivation and encouragement.

Deepest thanks to my former main supervisor, Assoc. Prof. Dr. Che Roos Saad for accepting me as his last Ph.D student. I am so glad to have a chance to be guided by the renowned fish nutritionist in Malaysia. He has given me invaluable guidance, warm encouragement, and suggestions in my quest for knowledge, as well as the freedom to pursue my research. Although I could not end up the journey with you, all your kindness, understanding, concern, and friendship are extremely appreciated.

I am also thankful to my co-supervisors, Assoc. Prof. Dr. Yaya Rukayadi and Prof. Dr. Mohd. Salleh Kamarudin for their guidance, encouragement, and valuable suggestions throughout my study. I feel so proud and honored for their willingness to be part of the committee members and providing assistance whenever required. To Dr. Yasser Abdel Hadi, thank you for your involvement in our committee previously, and sharing the expertise. Sincere appreciation and thanks are also extended to Assoc. Prof. Dr. Faridah Abas and Dr. Norazian Mohd. Hassan for their advices regarding HPLC work.

I wish to thank all the staffs and post-graduate students from Department of Aquaculture (Agriculture), Microbiology Laboratory (IBS), Aquatic Animal Health Unit (Veterinary), and Bacteriology Laboratory (Veterinary) UPM for their assistance, cooperation, and friendship especially to Mrs. Shafika, Mr. Jasni, Mr. Azmi, Mr. Zainal, and Mrs. Latifah. My heartiest appreciation also goes to my close friends, Farhana, Dr. Husna, Sairatul, Intan, Ayuni, Mursyida, Kak Azrin, Hazma, Fatihah, Saufinas, Nordiah, Faizul, Zamri, Clement, and Azim for the support, credible ideas, help, and all the memories.

I owe my loving thanks to my soul-mate, Muhamad Amir for his boundless support, encouragement, and for keeping me strong throughout the long and winding road of my

Ph.D journey. All your sacrifices, understanding, and patience in handling our situation of being apart for this Ph.D is undeniably a motivation and inspiration for me to keep going until the finish line. May Allah give you all the best in return. Also to my beloved parents, mother in-law, and siblings particularly my sister, for their love, concern, support, and prayers.

Lastly, I thank Ministry of Higher Education and Universiti Putra Malaysia for the funding under MyBrain15 Scholarship, Graduate Research Fellowship, and Research University Grant Scheme (RUGS 3).

I certify that a Thesis Examination Committee has met on 5 July 2018 to conduct the final examination of Syahidah Ahmad on her thesis entitled "Assessment of *Piper betle* L. Methanolic Extract as a Potential Growth and Health Promoter for Juvenile Red Hybrid Tilapia (*Oreochromis* sp.)" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Doctor of Philosophy.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Doctor of Philosophy. The members of the Supervisory Committee were as follows:

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LIST OF ABBREVIATIONS

ANOVA	One-Way Analysis of Variance
AOAC	Association of Official Analytical Chemist
β-C	β-caryophyllene
CCME	*C. caudatus* methanolic extract
cfu	Colony forming unit
CLSI	Clinical and Laboratory Standard Institute
D.DH_2O	Deionized distilled water
DMRT	Duncan Multiple Range Test
DO	Dissolved oxygen
dpi	Days of post-infection
DWG	Daily weight gain
EU	Eugenol
FAO	Food and Agriculture Organization of the United Nations
FCR	Feed conversion ratio
H&E	Haematoxylin and eosin
Hb	Haemoglobin
HC	Hydroxychavicol
Hct	Haematocrit
hpi	Hours of post-injection
HPLC	High Pressure Liquid Chromatography
LD_{50}	Median Lethal Dose
LG	Length gain
MBC	Minimum Bactericidal Concentration

MCHC	Mean corpuscular haemoglobin concentration
MCV	Mean corpuscular volume
MeOH	Methanol
MHA	Muller Hinton Agar
MHB	Muller Hinton Broth
MIC	Minimum Inhibition Concentration
NFE	Nitrogen free extract
NIC	No Inhibition Concentration
OTC	Oxytetracycline
PBME	*P. betle* methanolic extract
PER	Protein efficiency ratio
PWG	Percent weight gain
RBC	Red blood cell
R_f	Retention factor
RPS	Relative percentage survival
SE	Standard error
SGR	Specific growth rate
sp. or spp.	Species (for singular or plural term)
SPSS	Statistical Package for Social Science
TLC	Thin Layer Chromatography
WBC	White blood cell
WG	Weight gain
wkpf	Weeks of post-feeding
ZZME	*Z. zerumbet* methanolic extract

CHAPTER 1

GENERAL INTRODUCTION

1.1 Background of Study

Fish and other aquatic animals are vital sources of food for humans. They are nutritionally excellent since they have high quality animal protein apart from other essential nutrients. Also, they provide high bioavailability even in minor quantities in the diet. At a cheaper price, fish is the most frequently-consumed animal meat and thereby contributes very significantly to the variety in the daily human diet (Thilsted et al., 2013). Globally, over 4.3 billion people rely on fish as their main source of animal protein, with translates into at least 15% of their average per capita animal protein intake. Of this amount, it is approximated that the world consumed 128 million tonnes of fish or about 19.7 kg per individual in all time, as stated by Food and Agriculture Organisation of the United Nations (FAO, 2016).

Since the wild stocks of most commercially-important fish in the oceans have declined, the aquaculture industry has grown considerably in recent years to fulfil the market's demand. In 2014, the global total capture fishery production was reported to be 93.4 million tonnes, a 2.6% decrease as compared with the previous year. In contrast, the aquaculture industry has continued to show strong growth, where the average annual production rate has increased by 6.1% from 36.8 million tonnes in 2002 to 73.8 million tonnes in 2014. If the current trends continue, the aquaculture industry will surpass fisheries production in a few years' time (FAO, 2016).

Among the cultured species, tilapia is listed as the most widely-farmed fish in many parts of the world (FAO, 2014). The percentage of its production has been exponentially growing year by year. The world's total tilapia aquaculture production in 2015 was estimated to be 5 million tonnes, an increase of about 6% in the yearly production (FAO, 2016). The species that are most widely reared are those in the genus of *Oreochromis*. These comprise the Nile tilapia (*O. niloticus*), the Mozambique tilapia (*O. mossambicus*), and the blue tilapia (*O. aureus* and *O. urolepishornorum*). In recent times, fish farmers around the world have produced numerous strains from these parent species along with many successful hybrid strains, including red hybrid tilapia (*Oreochromis* sp.). This strain has become popular and is an important reason for its large-scale production for domestic consumption and exports (Watanabe et al., 2002). It is also the dominant (more than 90%) commercial species cultured in Malaysia (Department of Fisheries, 2013).

The rising global importance of cultured tilapia as a food fish has driven significant efforts for the improvement of the growth and health of this species. The intensive production of fish usually generates a stressful environment as a result of overcrowding, transport, handling, grading, and deterioration of water quality, leading to immune system depression, lower growth performance, and increased susceptibility

of the fish to infectious diseases (Harikrishnan et al., 2011a). Hence, various antibiotics, growth promoters, and chemotherapeutic agents are applied to address these problems. Overuse or misuse of the aforementioned agents lead to other problems such as the development of antibiotic resistance in various bacterial pathogens, accumulation of noxious chemical residues in fish, and environmental pollution (Ringø et al., 2010; Defoirdt et al., 2011; Romero et al., 2012). Thus, to overcome these drawbacks, researches nowadays focus on the use of natural products to replace synthetic agents.

Herbal plants are one of the interesting options as they have a variety of biological activities such as antimicrobial, antioxidant, appetite-stimulating, growth-promoting, and immunomodulatory activities owing by the presence of various active compounds. Besides, they are considered to be safe (Citarasu et al., 2010; Chakraborty et al., 2014; Hashimoto et al., 2016). Their applications are known to be able to improve the performance of animals like poultry and ruminants due to their effective role as broad spectrum prophylactic and growth-promoting agents (Brenes & Roura, 2010; Franz et al., 2010). Although the scientific data on herbs in fish is still limited, a number of studies have also reported beneficial biological effects of the herbs and their derivatives (Rahman et al., 2009; Direkbusarakom, 2011; Talpur & Ikhwanuddin, 2012, 2013). On top of that, herbal preparations can be given orally as feed additives to cultured fish of all sizes, rather than via injection or parental routes (through baths/immersion) that are commonly employed by vaccines, probiotics, and immunostimulants. Oral administration via feeds is considered to be the simplest and practical way as it does not induce handling-related stress. Thus, herbal extracts can be mass-administered.

1.2 Problem Statement

Until today, the occurrence of disease is a major constraint to the efficient production in aquaculture, resulting in serious economic losses. Emerging and re-emerging diseases are frequently reported, especially bacterial infections which are a main threat in the fish-farming industry. Even though tilapia is reported to be hardy, tilapia constantly succumbs to bacterial pathogens. At present, streptococcosis, which is caused by *Streptococcus agalactiae*, is the most important disease which affects tilapia production globally (Pereira et al., 2010; Ye et al., 2011). In Malaysia, streptococcosis can cause 40 – 50% mortality in the farmed tilapia, with an estimated annual loss of RM3 million (FAO, 2011).

Previous studies have documented that improper and continuous use of antibiotics and chemicals in aquaculture have induced numerous adverse side effects, which in turn have given rise to many risks to the human health and environment. Due to this reason, most government authorities around the world impose stringent regulations that limit the use of a number of antibiotics and chemicals in aquaculture. For example, antibiotics such as chloramphenicol, fluoroquinolones, nitrofurans, and quinolones, as well as chemical compounds like malachite green are antimicrobial agents that were recently banned for use in aquaculture by the U.S. Food and Drug Administration (USFDA) after revised testing programmes were done (Collette, 2006). Moreover, many countries such as the European Union (EU) and Singapore refused to import

aquaculture products tainted with antibiotics and chemicals, including the use of whole sub-therapeutic antibiotics as growth-promoting agents. In order to reduce or avoid the dependence of aquaculture on these conventional agents, attempts to use herbs to become an alternative have been intensified.

Therefore, the present study was aimed to assess the antibacterial properties and the safety of some easily-available Malaysian herbs – namely *Cosmos caudatus* (ulam Raja), *Curcuma mangga* (temumangga), *Justicia gendarussa* (gandarusa), *Piper betle* (sireh), and *Zingiber zerumbet* (lempoyang) – in developing fish feed supplement. Subsequently, the effects of selected herbal supplemented diet were investigated in terms of the juvenile red hybrid tilapia's (*Oreochromis* sp.) growth and health as well as disease resistance.

1.3 Hypothesis

1. Local herbal extracts have broad spectrum antibacterial activities and have promising potential to be antibacterial agents.
2. Local herbal extracts have high safety margins and are non-toxic to fish tissues.
3. Selected herbal extract need contains bioactive compounds in certain concentrations to effect antibacterial activities.
4. Supplementation of selected herbal extract in diet can improve the growth and health of *Oreochromis* sp.
5. The supplementation of selected herbal extract in diet can confer protection against *Streptococcus agalactiae* in *Oreochromis* sp.

1.4 Objectives

The general objective of this study was to investigate the potential of local herbal extracts to be diet supplement for juvenile *Oreochromis* sp. to promote better growth and health. The specific objectives of this study were:

1. to screen and evaluate the potential antibacterial activities of local herbal extracts against fish pathogenic bacteria.
2. to evaluate the toxicity of local herbal extracts and determine their safety level in *Oreochromis* sp.
3. to identify and quantify the major antibacterial active compounds of selected herbal extract.
4. to determine the growth performance, survival, feed utilisation, and body composition of *Oreochromis* sp. fed with selected herbal extract supplemented diet.
5. to clarify the effects of selected herbal extract supplemented diet on the haematological and histological of important tissues of *Oreochromis* sp.
6. to determine the disease resistance of *Oreochromis* sp. fed with selected herbal extract supplemented diet when challenged with *Streptococcus agalactiae*.

REFERENCES

Abas, F., Lajis, N. H., & Kalsom, Y. U. (2003). Antioxidative and radical scavenging properties of the constituents isolated from *Cosmos caudatus* Kunth. *Natural Product Sciences*, *9*(4), 245-248.

Abas, F., Lajis, N. H., Israf, D. A., Khozirah, S., & Kalsom, Y. U. (2006). Antioxidant and nitric oxide inhibition activities of selected Malay traditional vegetables. *Food Chemistry*, *95*(4), 566-573.

Abas, F., Lajis, N. H., Shaari, K., Israf, D. A., Stanslas, J., Yusuf, U. K., & Raof, S. M. (2005). A labdane diterpene glucoside from the rhizomes of *Curcuma mangga*. *Journal of Natural Products*, *68*(7), 1090-1093.

Abd Aziz, S. M., Low, C. N., Chai, L. C., Abd Razak, S. S. N., Selamat, J., Son, R., Sarker, M. Z. I., & Khatib, A. (2011). Screening of selected Malaysian plants against several food borne pathogen bacteria. *International Food Research Journal*, *18*(3), 1195-1201.

Abdallah, E. M., Khalid, A. S., & Ibrahim, N. (2009). Antibacterial activity of oleo-gum resins of *Commiphora molmol* and *Boswellia papyrifera* against methicillin resistant *Staphylococcus aureus* (MRSA). *Scientific Research and Essays*, *4*(4), 351-356.

Abdel-Tawwab, M., Ahmad, M. H., Seden, M. E., & Sakr, S. F. (2010). Use of green tea, *Camellia sinensis* L., in practical diet for growth and protection of Nile tilapia, *Oreochromis niloticus* (L.), against *Aeromonas hydrophila* infection. *Journal of the World Aquaculture Society*, *41*(2), 203-213.

Abdelwahab, S. I., Abdul, A. B., Zain, Z. N. M., & Hadi, A. H. A. (2012). Zerumbone inhibits interleukin-6 and induces apoptosis and cell cycle arrest in ovarian and cervical cancer cells. *International Immunopharmacology*, *12*(4), 594-602.

Abdullah, N. F., & Mohamad Hussain, R. (2015). Isolation of allylpyrocatechol from *Piper betle* L. leaves by using high-performance liquid chromatography. *Journal of Liquid Chromatography and Related Technologies*, *38*(2), 289-293.

Abou Zeid, E. N. (1989). *Aromatic plants and its pharmaceutical and agricultural products*. Egypt: Academic Press.

Abuseliana, A., Daud, H., Saleha, A. A., Khairani-Bejo, S., & Alsaid, M. (2010). *Streptococcus agalactiae* the aetiological agent of mass mortality in farmed red tilapia (*Oreochromis* sp.). *Journal of Animal and Veterinary Advances*, *9*(20), 2640-2646.

Abuseliana, A. F., Daud, H. H. M., Aziz, S. A., Bejo, S. K., & Alsaid, M. (2011). Pathogenicity of *Streptococcus agalactiae* isolated from a fish farm in Selangor to juvenile red tilapia (*Oreochromis* sp.). *Journal of Animal and Veterinary Advances*, *10*(7), 914-919.

Abu-Shanab, B., Adwan, G. M., Abu-Safiya, D., Jarrar, N., & Adwan, K. (2005). Antibacterial activities of some plant extracts utilized in popular medicine in Palestine. *Turkish Journal of Biology*, *28*(2-4), 99-102.

Acar, Ü., Kesbiç, O. S., Yılmaz, S., Gültepe, N., & Türker, A. (2015). Evaluation of the effects of essential oil extracted from sweet orange peel (*Citrus sinensis*) on growth rate of tilapia (*Oreochromis mossambicus*) and possible disease resistance against *Streptococcus iniae*. *Aquaculture*, 437, 282-286.

Adams, M. A., Johnsen, P. B., & Hong Qi, Z. (1988). Chemical enhancement of feeding for the herbivorous fish *Tilapia zillii*. *Aquaculture*, *72* (1-2): 95-107.

Adedeji, O. B., Taiwo, V. O., & Agbede, S. A. (2000). Comparative haematology of five Nigerian freshwater fish species. *Nigerian Veterinary Journal*, *21*, 75-84.

Adekunle, A. D. (2012). Effects of herbal growth promoter feed additive in fish meal on the performance of Nile tilapia *Oreochromis niloticus* (L.). *Egypt Academic Journal of Biology Sciences*, *4*(1), 111-117.

Adeoye, A. A., Jaramillo-Torres, A., Fox, S. W., Merrifield, D. L., & Davies, S. J. (2016). Supplementation of formulated diets for tilapia (*Oreochromis niloticus*) with selected exogenous enzymes: overall performance and effects on intestinal histology and microbiota. *Animal Feed Science and Technology*, *215*, 133-143.

Adewole, A. M. (2014). Effects of roselle as dietary additive on growth performance and production economy of *Clarias gariepinus*. *Journal of Emerging Trends in Engineering and Applied Sciences*, *5*(7), 1-8.

Agius, C., & Roberts, R. J. (2003). Melano-macrophage centres and their role in fish pathology. *Journal of Fish Diseases*, *26*(9), 499-509.

Ahmad, I., Mehmood, Z., & Mohammad, F. (1998). Screening of some Indian medicinal plants for their antimicrobial properties. *Journal of Ethnopharmacology*, *62*(2), 183-193.

Ahmad, M. H., & Abdel-Tawwab, M. (2011). The use of caraway seed meal as a feed additive in fish diets: Growth performance, feed utilization, and whole-body composition of Nile tilapia, *Oreochromis niloticus* (L.) fingerlings. *Aquaculture*, *314*(1), 110-114.

Akhila, J. S., Shyamjith, Deepa, & Alwar, M. C. (2007). Acute toxicity studies and determination of median lethal dose. *Current Science*, *93*(7), 917-920.

Akinjogunla, O. J., Yah, C. S., Eghafona, N. O., & Ogbemudia, F. O. (2010). Antibacterial activity of leave extracts of *Nymphaea lotus* (Nymphaeaceae) on methicillin resistant *Staphylococcus aureus* (MRSA) and vancomycin resistant *Staphylococcus aureus* (VRSA) isolated from clinical samples. *Annals of Biological Research*, *1*(2), 174-184.

Akinwole, A. O., & Faturoti, E. O. (2007). Biological performance of African Catfish (*Clarias gariepinus*) cultured in recirculating system in Ibadan. *Aquacultural Engineering*, *36*(1), 18-23.

Akter, K. N., Karmakar, P., Das, A., Anonna, S. N., Shoma, S. A., & Sattar, M. M. (2014). Evaluation of antibacterial and anthelmintic activities with total phenolic contents of *Piper betel* leaves. *Avicenna Journal of Phytomedicine*, *4*(5), 320-329.

Al Somal, N., Coley, K. E., Molan, P. C., & Hancock, B. M. (1994). Susceptibility of *Helicobacter pylori* to the antibacterial activity of manuka honey. *Journal of the Royal Society of Medicine*, *87*(1), 9-12.

Albert, V., & Ransangan, J. (2013). Antibacterial potential of plant crude extracts against Gram negative fish bacterial pathogens. *International Journal of Research in Pharmaceutical and Biosciences*, *3*, 21-27.

Al-Daihan, S., Al-Faham, M., Al-Shawi, N., Almayman, R., Brnawi, A., & Shafi Bhat, R. (2013). Antibacterial activity and phytochemical screening of some medicinal plants commonly used in Saudi Arabia against selected pathogenic microorganisms. *Journal of King Saud University-Science*, *25*(2), 115-120.

Allam, S. M., El-Hossieny, H. M., Abdel-Gawad, A. M., El-Saadany, S. A., & Zeid, A. M. M. (1999). Medicinal herbs and plants as feed additives for ruminants. 1-Effect of using some medicinal herbs and plants as feeds additives on Zaraibi goat performance. *Egyptian Journal of Nutrition and Feeds*, *2*(Special issue), 349-365.

Alsaid, M., Abuseliana, A. F., Daud, H. H., Mustapha, N. M., Bejo, S. K., Abdelhadi, Y. M., & Hamdan, R. H. (2014). Haematological, biochemical and clinical signs changes following experimental infection of *Streptococcus agalactiae* in red hybrid tilapia (*Oreochromis* sp.). *Aquacultura Indonesiana*, *15*(2), 86-93.

Alsaid, M., Daud, H., Bejo, S. K., & Abuseliana, A. (2010). Antimicrobial activities of some culinary spice extracts against *Streptococcus agalactiae* and its prophylactic uses to prevent streptococcal infection in red hybrid tilapia (*Oreochromis* sp.). *World Journal of Fish and Marine Sciences*, *2*(6), 532-538.

Alsaid, M., Mohd, H., Mohamed, N., Khairani, S., Mohamed, Y., Farag, A., & Hayati, R. (2013, December). Pathological findings of experimental *Streptococcus agalactiae* infection in red hybrid tilapia (*Oreochromis* sp.). In *International Conference on Chemical, Agricultural and Medical Sciences* (pp. 29-30). Kuala Lumpur, Malaysia.

Alwan, S. F., Hadi, A. A., & Shokr, A. E. (2009). Alterations in hematological parameters of fresh water fish, *Tilapia zillii*, exposed to aluminum. *Journal of Science and Its Applications*, *3*(1), 12-19.

Aly, S. M., & Mohamed, M. F. (2010). *Echinacea purpurea* and *Allium sativum* as immunostimulants in fish culture using Nile tilapia (*Oreochromis niloticus*). *Journal of Animal Physiology and Animal Nutrition*, *94*(5), 31-39.

Aly, S. M., Ahmed, Y. A. G., Ghareeb, A. A. A., & Mohamed, M. F. (2008). Studies on *Bacillus subtilis* and *Lactobacillus acidophilus*, as potential probiotics, on the immune response and resistance of Tilapia nilotica (*Oreochromis niloticus*) to challenge infections. *Fish and Shellfish Immunology*, *25*(1), 128-136.

Amal, M. N. A., & Zamri-Saad, M. (2011). Streptococcosis in tilapia (*Oreochromis niloticus*): a review. *Pertanika Journal of Tropical Agricultural Science*, *34*(2), 195-206.

Amal, M. N. A., Nur-Nazifah, M., Siti-Zahrah, A., Sabri, M. Y., & Zamri-Saad, M. (2008, October). Determination of LD50 for *Streptococcus agalactiae* infections in red tilapia and GIFT. In *8th International Symposium on Tilapia in Aquaculture* (pp. 1245-1251).

Amal, M. N. A., Zamri-Saad, M., Zulkafli, A. R., Siti-Zahrah, A., Misri, S., Ramley, B., & Sabri, M. Y. (2010). Water thermocline confirms susceptibility of tilapia

cultured in lakes to *Streptococcus agalactiae*. *Journal of Animal and Veterinary Advances*, *9*(22), 2811-2817.

Amna, O. F., Nooraain, H., Noriham, A., Azizah, A. H., & Husna, R. N. (2013). Acute and oral subacute toxicity study of ethanolic extract of *Cosmos caudatus* leaf in Sprague Dawley rats. *International Journal of Bioscience, Biochemistry and Bioinformatics*, *3*(4), 301-305.

Amonkar, A. J., Nagabhushan, M., D'souza, A. V., & Bhide, S. V. (1986). Hydroxychavicol: a new phenolic antimutagen from betel leaf. *Food and Chemical Toxicology*, *24*(12), 1321-1324.

Andarwulan, N., Kurniasih, D., Apriady, R. A., Rahmat, H., Roto, A. V., & Bolling, B. W. (2012). Polyphenols, carotenoids, and ascorbic acid in underutilized medicinal vegetables. *Journal of Functional Foods*, *4*(1), 339-347.

Anderson, D. P. (1992). Immunostimulants, adjuvants, and vaccine carriers in fish: applications to aquaculture. *Annual Review of Fish Diseases*, *2*, 281-307.

Annegowda, H. V., Tan, P. Y., Mordi, M. N., Ramanathan, S., Hamdan, M. R., Sulaiman, M. H., & Mansor, S. M. (2013). TLC–bioautography-guided isolation, HPTLC and GC–MS-assisted analysis of bioactives of *Piper betle* leaf extract obtained from various extraction techniques: in vitro evaluation of phenolic content, antioxidant and antimicrobial activities. *Food Analytical Methods*, *6*(3), 715-726.

Anokwuru, C. P., Anyasor, G. N., Ajibaye, O., Fakoya, O., & Okebugwu, P. (2011). Effect of extraction solvents on phenolic, flavonoid and antioxidant activities of three Nigerian medicinal plants. *Nature and Science*, *9*(7), 53-61.

Arambewela, L. S. R., Arawwawala, L. D. A. M., & Ratnasooriya, W. D. (2005). Antidiabetic activities of aqueous and ethanolic extracts of *Piper betle* leaves in rats. *Journal of Ethnopharmacology*, *102*(2), 239-245.

Ardó, L., Yin, G., Xu, P., Váradi, L., Szigeti, G., Jeney, Z., & Jeney, G. (2008). Chinese herbs (*Astragalus membranaceus* and *Lonicera japonica*) and boron enhance the non-specific immune response of Nile tilapia (*Oreochromis niloticus*) and resistance against *Aeromonas hydrophila*. *Aquaculture*, *275*(1), 26-33.

Arya, V., Yadav, S., Kumar, S., & Yadav, J. P. (2010). Antimicrobial activity of *Cassia occidentalis* L (leaf) against various human pathogenic microbes. *Life Sciences and Medicine Research*, *9*, 1-11.

Ashraf, M. A. S., (2008). Effect of dietary Ginseng herb (Ginsana® G115) supplementation on growth, feed utilization, and hematological indices of Nile Tilapia, *Oreochromis niloticus* (L.), fingerlings. *Journal of the World Aquaculture Society*, *39*(2), 205-214.

Association of Official Analytical Chemists. (1990). *Official methods of analysis of AOAC international* (15th ed.). Washington, DC: AOAC.

Ates, B., Orun, I., Talas, Z. S., Durmaz, G., & Yilmaz, I. (2008). Effects of sodium selenite on some biochemical and hematological parameters of rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) exposed to Pb2+ and Cu2+. *Fish physiology and Biochemistry*, *34*(1), 53-59.

Attimarad, M., Ahmed, K. M., Aldhubaib, B. E., & Harsha, S. (2011). High-performance thin layer chromatography: A powerful analytical technique in pharmaceutical drug discovery. *Pharmaceutical Methods*, *2*(2), 71-75.

Atz, J. W. (1954). The peregrinating tilapia. *Animal Kingdom*, *50*(5):148-155.

Austin, B. (2006). The bacterial microflora of fish, revised. *The Scientific World Journal*, *6*, 931-945.

Awad, E. S. (2010). *Studies on plant based dietary supplements for control of Aeromonas hydrophila infections in rainbow trout (Oncorhynchus mykiss Walbaum)* (Doctoral dissertation). Heriot-Watt University.

Ayob, Z., Mohd Bohari, S. P., Abd Samad, A. & Jamil, S. (2014). Cytotoxic activities against breast cancer cells of local *Justicia gendarussa* crude extracts. *Evidence-Based Complementary and Alternative Medicine, 2014*, 732980. doi:10.1155/2014/732980

Ayob, Z., Samad, A. A., & Bohari, S. M. (2013). Cytotoxicity activities in local *Justicia gendarussa* crude extracts against human cancer cell lines. *Jurnal Teknologi (Sciences and Engineering)*, *64*(2), 45-52.

Ayoola, S. O. (2011). Haematological characteristics of *Clarias gariepinus* (Buchell, 1822) juveniles fed with poultry hatchery waste. *Iranica Journal of Energy and Environment*, *2*(1), 18-23.

Ayotunde, E. O., & Ofem, B. O. (2008). Acute and chronic toxicity of pawpaw (*Carica papaya*) seed powder to adult Nile tilapia (*Oreochromis niloticus* Linne 1757) fingerlings. *Advance in Environmental Biology*, *2*(3), 103-107.

Ayotunde, E. O., Fagbenro, O. A., Adebayo, O. T., & Amoo, A. I. (2004). Toxicity of aqueous extracts of drumstick, *Moringa oleifera* Seeds to Nile tilapia, *Oreochromis niloticus* fingerlings and adults. In *Proceedings of the 6th International Symposium on Tilapia in Aquaculture* (pp. 200-208).

Aziz, Z., & Tey, N. P. (2009). Herbal medicines: prevalence and predictors of use among Malaysian adults. *Complementary Therapies in Medicine, 17*(1), 44-50.

Azmir, J., Zaidul, I. S. M., Rahman, M. M., Sharif, K. M., Mohamed, A., Sahena, F., Jahurul, M. H. A., Ghafoor, K., Norulaini, N. A. N., & Omar, A. K. M. (2013). Techniques for extraction of bioactive compounds from plant materials: a review. *Journal of Food Engineering*, *117*(4), 426-436.

Bajpai, V., Pandey, R., Negi, M. P., Kumar, N., & Kumar, B. (2012). DART MS based chemical profiling for therapeutic potential of *Piper betle* landraces. *Natural Product Communications*, *7*(12), 1627-1629.

Bajpai, V., Sharma, D., Kumar, B., & Madhusudanan, K. P. (2010). Profiling of *Piper betle* Linn. cultivars by direct analysis in real time mass spectrometric technique. *Biomedical Chromatography*, *24*(12), 1283-1286.

Balcázar, J. L., De Blas, I., Ruiz-Zarzuela, I., Cunningham, D., Vendrell, D., & Muzquiz, J. L. (2006). The role of probiotics in aquaculture. *Veterinary microbiology*, *114*(3), 173-186.

Banso, A., & Adeyemo, S. O. (2007). Evaluation of antibacterial properties of tannins isolated from *Dichrostachys cinerea*. *African Journal of Biotechnology*, *6*(15), 1785-1787.

Barham, W. T., Schoonbee, H., & Smit, G. L. (1979). The occurrence of Aeromonas and Streptococcus in rainbow trout, *Salmo gairdneri* Richardson. *Journal of Fish Biology*, *15*(4), 457-460.

Basri, D. F., & Fan, S. H. (2005). The potential of aqueous and acetone extracts of galls of *Quercus infectoria* as antibacterial agents. *Indian Journal of Pharmacology*, *37*(1), 26-29.

Bauer, A. W., Kirby, W. M., Sherris, J. C., & Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. *American Journal of Clinical Pathology*, *45*(4), 493–496.

Behrends, L. L., Nelson, R. G., Smitherman, R. O., & Stone, N. M. (1982). Breeding and culture of the red-gold color phase of tilapia. *Journal of the World Aquaculture Society*, *13*(1-4), 210-220.

Bento, M. H. L., Ouwehand, A. C., Tiihonen, K., Lahtinen, S., Nurminen, P., Saarinen, M. T., & Fischer, J. (2013). Essential oils and their use in animal feeds for monogastric animals--Effects on feed quality, gut microbiota, growth performance and food safety: a review. *Veterinarni Medicina*, *58*(9), 449-458.

Bernet, D., Schmidt, H., Meier, W., Burkhardt-Holm, P., & Wahli, T. (1999). Histopathology in fish: proposal for a protocol to assess aquatic pollution. *Journal of Fish Diseases*, *22*(1), 25-34.

Betoni, J. E. C., Mantovani, R. P., Barbosa, L. N., Di Stasi, L. C., & Fernandes Junior, A. (2006). Synergism between plant extract and antimicrobial drugs used on *Staphylococcus aureus* diseases. *Memorias do Instituto Oswaldo Cruz*, *101*(4), 387-390.

Bhalerao, S. A., Verma, D. R., Gavankar, R. V., Teli, N. C., Rane, Y. Y., Didwana, V. S., & Trikannad, A. (2013). Phytochemistry, pharmacological profile and therapeutic uses of *Piper betle* Linn-An overview. Research and Reviews: *Journal of Pharmacognosy and Phytochemistry*, *1*(2), 10-19.

Bhosale, S. V., Bhilave, M. P., & Nadaf, S. B. (2010). Formulation of fish feed using ingredients from plant sources. *Research Journal of Agricultural Sciences*, *1*(3), 284-287.

Bissa, S., Songara, D., & Bohra, A. (2007). Traditions in oral hygiene: chewing of betel (*Piper betle* L.) leaves. *Current Science*, *92*(1), 26-28.

Blaxhall, P. C. (1972). The haematological assessment of the health of freshwater fish. *Journal of Fish Biology*, *4*(4), 593-604.

Bodeker, G. (2009). *Health and Beauty from the Rainforest: Malaysian Traditions of Ramuan.* Kuala Lumpur: Didier Millet.

Bomba, A., Nemcova, R., Gancarcikova, S., Herich, R., Guba, P., & Mudronova, D. (2002). Improvement of the probiotic effect of micro-organisms by their combination with maltodextrins, fructo-oligosaccharides and polyunsaturated fatty acids. *British Journal of Nutrition*, *88*(1), 95-99.

Borchardt, J. K. (2002). The beginnings of drug therapy: Ancient mesopotamian medicine. *Drug News Perspect*, *15*(3), 187-192.

Brenes, A., & Roura, E. (2010). Essential oils in poultry nutrition: Main effects and modes of action. *Animal Feed Science and Technology*, *158*(1), 1-14.

Bromage, E. S., & Owens, L. (2002). Infection of barramundi *Lates calcarifer* with *Streptococcus iniae*: effects of different routes of exposure. *Diseases of Aquatic Organisms*, *52*(3), 199-205.

Burkill, I. H. (1966). *A Dictionary of the Economic Products of the Malay Peninsula* (vol. II, I–Z). Kuala Lumpur, Malaysia: Ministry of Agriculture and Co-operatives.

Burt, S. (2004). Essential oils: their antibacterial properties and potential applications in foods-a review. *International Journal of Food Microbiology*, *94*(3), 223-253.

Bustamam, A., Ibrahim, S., Al-Zubairi, A. S., Met, M., & Syam, M. M. (2008). Zerumbone: a natural compound with anti-cholinesterase activity. *American Journal of Pharmacology and Toxicology*, *3*(3), 209-211.

Cabello, F. C. (2006). Heavy use of prophylactic antibiotics in aquaculture: a growing problem for human and animal health and for the environment. *Environmental Microbiology*, *8*(7), 1137-1144.

Cai, Z., Lee, F. S. C., Wang, X. R., & Yu, W. J. (2002). A capsule review of recent studies on the application of mass spectrometry in the analysis of Chinese medicinal herbs. *Journal of Mass Spectrometry*, *37*(10), 1013-1024.

Cannell, R. J. P. (1998). *Natural Products Isolation.* New Jersey, USA: Humana Press Inc.

Camargo, M. M. P., & Martinez, C. B. R. (2007). Histopathology of gills, kidney and liver of a Neotropical fish caged in an urban stream. *Neotropical Ichthyology*, *5*(3), 327-336.

Castro, R., Lamas, J., Morais, P., Sanmartín, M. L., Orallo, F., & Leiro, J. (2008). Resveratrol modulates innate and inflammatory responses in fish leucocytes. *Veterinary Immunology and Immunopathology*, *126*(1), 9-19.

Cencic, A., & Chingwaru, W. (2010). The role of functional foods, nutraceuticals, and food supplements in intestinal health. *Nutrients*, *2*(6), 611-625.

Cetin-karaca, H. (2011). *Evaluation of natural antimicrobial phenolic compounds against foodborne pathogens* (Master's thesis, University of Kentucky). Retrieved from https://uknowledge.uky.edu/cgi/viewcontent.cgi?article=1163&context=gradschool_theses

Chakraborty, S. B., & Hancz, C. (2011). Application of phytochemicals as immunostimulant, antipathogenic and antistress agents in finfish culture. *Reviews in Aquaculture*, *3*(3), 103-119.

Chakraborty, S. B., Horn, P., & Hancz, C. (2014). Application of phytochemicals as growth-promoters and endocrine modulators in fish culture. *Reviews in Aquaculture*, *6*(1), 1-19.

Chan, E. W. C., Lim, Y. Y., Wong, S. K., Lim, K. K., Tan, S. P., Lianto, F. S., & Yong, M. Y. (2009). Effects of different drying methods on the antioxidant properties of leaves and tea of ginger species. *Food Chemistry*, *113*(1), 166-172.

Chan, E. W. C., Tan, Y. P., Chin, S. J., Gan, L. Y., Kang, K. X., Fong, C. H., Chang, H. Q., & How, Y. C. (2014). Antioxidant properties of selected fresh and processed herbs and vegetables. *Free Radicals and Antioxidants*, *4*(1), 39.

Chan, E. W. C., Wong, S. K., & Chan, H. T. (2017). Ulam Herbs of *Oenanthe javanica* and *Cosmos caudatus*: an overview on their Medicinal Properties. *Journal of Natural Remedies*, *16*(4), 137-147.

Chan, K. L., & Choo, C. Y. (2002). The toxicity of some quassinoids from *Eurycoma longifolia*. *Planta Medica*, *68*(07), 662-664.

Chang, C. C., Tan, H. C., & Cheng, W. (2013). Effects of dietary administration of water hyacinth (*Eichhornia crassipes*) extracts on the immune responses and disease resistance of giant freshwater prawn, *Macrobrachium rosenbergii*. *Fish and Shellfish Immunology*, *35*(1), 92-100.

Chang, C. J., Tzeng, T. F., Liou, S. S., Chang, Y. S., & Liu, I. M. (2012). Acute and 28-day subchronic oral toxicity of an ethanol extract of *Zingiber zerumbet* (L.) Smith in rodents. *Evidence-Based Complementary and Alternative Medicine, 2012*, 608284. doi:10.1155/2012/608284

Chapman, F. A. (2015). *Culture of hybrid tilapia: A reference profile*. Retrieved from https://edis.ifas.ufl.edu/pdffiles/FA/FA01200.pdf

Chauhan, A., Goyal, M. K., & Chauhan, P. (2014). GC-MS technique and its analytical applications in science and technology. *Jounal of Analytical and Bioanalytical Techniques*, *5*(6), 222.

Chen, C. Y., Wooster, G. A., Getchell, R. G., Bowser, P. R., & Timmons, M. B. (2003). Blood chemistry of healthy, nephrocalcinosis-affected and ozone-treated tilapia in a recirculation system, with application of discriminant analysis. *Aquaculture*, *218*(1), 89-102.

Chen, C., Chao, C., & Bowser, P. R. (2007). Comparative histopathology of *Streptococcus iniae* and *Streptococcus agalactiae*-infected tilapia. *Bulletin-European Association of Fish Pathologists*, *27*(1), 2-9.

Cheng, S. H., Ismail, A., Anthony, J., Ng, O. C., Hamid, A. A., & Barakatun-Nisak, M. Y. (2015). Eight weeks of *Cosmos caudatus* (ulam Raja) supplementation improves glycemic status in patients with type 2 diabetes: a randomized controlled trial. *Evidence-Based Complementary and Alternative Medicine, 2015*, 405615. doi:10.1155/2015/405615

Chervinski, J. (1982). Environmental physiology of tilapias. In Pullin, R. S. V., & Lowe-McConnell, R. H. (Eds.), *The Biology and Culture of Tilapias*: *ICLARM Conference Proceedings* (vol. 7, pp. 119–128).

Chesson, A. (1987) Supplementary enzymes to improve the utilization of pig and poultry diets. In Haresign W, & Cole, D. J. A. (Eds.), *Recent Advances in Animal Nutrition.* Butterworths, London: Anchor Brandon Ltd.

Chiao-Wei, C., Siew-Ling, H., & Ching-Lee, W. (2011). Antibacterial activity of *Sargassum polycystum* C. Agardh and *Padina australis* Hauck (Phaeophyceae). *African Journal of Biotechnology*, *10*(64), 14125-14131.

Chitmanat, C., Tongdonmuan, K., Khanom, P., Pachontis, P., & Nunsong, W. (2005). Antiparasitic, antibacterial, and antifungal activities derived from a *Terminalia catappa* solution against some tilapia (*Oreochromis niloticus*) pathogens. *Acta Horticulturae*, *678*(25), 179-182.

Chitra, S. (1995). *Effect of feeding supplemented stresstol bioencapsulated Artemia franciscana on growth and stress tolerance in Penaeus indicus postlarvae*. (M. Phil dissertation). MS University, Tirunelveli.

Chopra, R. N, Nayyar, S. L., & Chopra, I. C. (1956). *Glossary of Indian medicinal plants.* New Delhi, India: Council of Scientific and Industrial Research.

Choudhary, D., & Kale, R. K. (2002). Antioxidant and non-toxic properties of *Piper betle* leaf extract: *in vitro* and *in vivo* studies. *Phytotherapy Research*, *16*(5), 461-466.

Citarasu, T. (2010). Herbal biomedicines: a new opportunity for aquaculture industry. *Aquaculture International*, *18*(3), 403-414.

Clauss, T. M., Dove, A. D., & Arnold, J. E. (2008). Hematologic disorders of fish. *Veterinary Clinics of North America: Exotic Animal Practice*, *11*(3), 445-462.

Clinical and Laboratory Standards Institute. (2006). *Methods for antimicrobial disk susceptibility testing of bacteria isolated from aquatic animals* (Approved guideline M2-A9). Wayne, PA, USA: CLSI.

Coles, E. H. (1986). *Veterinary clinical pathology* (4th ed.). Philadelphia, PA: Saunders.

Collette, B. (2006). *Industry efforts reduce use of unapproved drugs*. Global Aquaculture Advocate (pp. 38-39). Rockville, MD, USA: USFDA.

Collins, C. H., Lyne, P. M., & Grange, J. (1989). *Microbiology methods* (6th ed.). London, UK: Butterworth & Co. Ltd.

Cook, M. T., Hayball, P. J., Hutchinson, W., Hayball, J. D. & Nowak, B. (2001). The efficiency of a commercial β-glucan preparation on stimulating respiratory burst activity of head kidney macrophages from pink snapper (*Pagrus auratus*). *Fish and Shellfish Immunology*, *11*(8): 661-672.

Cowan, M. M. (1999). Plant products as antimicrobial agents. *Clinical Microbiology Reviews*, *12*(4), 564-582.

Coz-Rakovac, R., Strunjak-Perovic, I., Hacmanjek, M., Lipej, Z., & Sostaric, B. (2005). Blood chemistry and histological properties of wild and cultured sea bass (*Dicentrarchus labrax*) in the North Adriatic Sea. *Veterinary Research Communications*, *29*(8), 677-687.

Cragg, G. M., & Newman, D. J. (2013). Natural products: a continuing source of novel drug leads. *Biochimica et Biophysica Acta-General Subjects*, *1830*(6), 3670-3695.

Dada, A. A. (2015). Improvement of tilapia (*Oreochromis niloticus* Linnaeus, 1758) growth performance fed three commercial feed additives in diets. *Journal of Aquaculture Research and Development*, *6*(4), 1-3.

Dada, A. A. (2017). Use of fluted pumpkin (*Telfairia occidentalis*) leaf powder as feed additive in African catfish (*Clarias gariepinus*) fingerlings. *Journal of Applied Animal Research*, *45*(1), 566-569.

Dada, A. A., & Olugbemi, B. D. (2013). Dietary effects of two commercial feed additives on growth performance and body composition of African catfish *Clarias gariepinus* fingerlings. *African Journal of Food Science*, *7*(9), 325-328.

Da Rocha, A. M., de Freitas, D. S., Burns, M., Vieira, J. P., de La Torre, F. R., & Monserrat, J. M. (2009). Seasonal and organ variations in antioxidant capacity, detoxifying competence and oxidative damage in freshwater and estuarine fishes from Southern Brazil. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, *150*(4), 512-520.

Dar, S. A., Ganai, F. A., Yousuf, A. R., Bhat, T. M., & Bhat, F. A. (2012). Bioactive potential of leaf extracts from *Urtica dioica* L. against fish and human pathogenic bacteria. *African Journal of Microbiology Research*, *6*(41), 6893-6899.

Das, K., Tiwari, R. K. S., & Shrivastava, D. K. (2010). Techniques for evaluation of medicinal plant products as antimicrobial agents: current methods and future trends. *Journal of Medicinal Plants Research*, *4*(2), 104-111.

Davies, J. (1994). Inactivation of antibiotics and the dissemination of resistance genes. Science, *264*(5157), 375-381.

Department of Fisheries. (2013). Annual fisheries statistics 2013. Retrieved from https:// www.dof.gov.my/en/fishery.

De Padua, L. S., Bunyapraphatsara, N., & Lemmens, R. H. M. J. (1999). *Plant resources of South-East Asia* (Vol. 12, pp. 210-218). Netherlands: Backhuys Publishers.

De Silva, S. S. (2004). *Tilapias as alien aquatics in Asia and the Pacific: a review*. FAO Fisheries Technical Paper (No. 453, p 65). Rome, Italy.

Defoirdt, T., Sorgeloos, P., & Bossier, P. (2011). Alternatives to antibiotics for the control of bacterial disease in aquaculture. *Current Opinion in Microbiology*, *14*(3), 251-258.

Delaquis, P. J., Stanich, K., Girard, B., & Mazza, G. (2002). Antimicrobial activity of individual and mixed fractions of dill, cilantro, coriander and eucalyptus essential oils. *International Journal of Food Microbiology*, *74*(1), 101-109.

Dewanjee, S., Gangopadhyay, M., Bhattacharya, N., Khanra, R., & Dua, T. K. (2015). Bioautography and its scope in the field of natural product chemistry. *Journal of Pharmaceutical Analysis*, *5*(2), 75-84.

Din, A. R. J. M., Razak, S. A., & Sabaratnam, V. (2012). Effect of mushroom supplementation as a prebiotic compound in super worm based diet on growth performance of red tilapia fingerlings. *Sains Malaysiana*, *41*(10), 1197-1203.

Direkbusarakom, S. (2011). Application of medicinal herbs to aquaculture in Asia. *Walailak Journal of Science and Technology*, *1*(1), 7-14.

Divyagnaneswari, M., Christybapita, D., & Michael, R. D. (2007). Enhancement of nonspecific immunity and disease resistance in *Oreochromis mossambicus* by *Solanum trilobatum* leaf fractions. *Fish and Shellfish Immunology*, *23*(2), 249-259.

Douglass, J. W., & Jane, K. W. (Eds.). (2010). *Schalm's Veterinary Hematology*. UK: John Wiley and Sons, Blackwell Publishing Ltd.

Duthie, G. G., & Tort, L. (1985). Effects of dorsal aortic cannulation on the respiration and haematology of mediterranean living *Scyliorhinus canicula* L. *Comparative Biochemistry and Physiology Part A: Physiology*, *81*(4), 879-883.

Dutta, H. M. (1996). A composite approach for evaluation of the effects of pesticides on fish. *Fish morphology, JSD Munshi and HM Dutta, editors. Lebanon, Science Publisher Inc*.

Dwivedi, V., & Tripathi, S. (2014). Review study on potential activity of *Piper betle*. *Journal of Pharmacognosy and Phytochemistry*, *3*(4), 93-98.

Ebana, R. U. B., Madunagu, B. E., Ekpe, E. D., & Otung, I. N. (1991). Microbiological exploitation of cardiac glycosides and alkaloids from *Garcinia kola*, *Borreria ocymoides*, *Kolanitida* and *Citrus aurantifolia*. *Journal of Applied Microbiology*, *71*(5), 398-401.

Edeoga, H. O., Okwu, D. E., & Mbaebie, B. O. (2005). Phytochemical constituents of some Nigerian medicinal plants. *African Journal of Biotechnology*, *4*(7), 685-688.

El-Dakar, A. Y., Hassanien, G. D. I., Seham, S., Gad & Sakr, S. E. (2004). Use of medical and aromatic plants in fish diets: I. Effect of dried marjoram leaves on performance of hybrid tilapia *Oreochromis niloticus* x *Oreochromis aureus*, fingerlings. *Journal of the Egyptian Academic Society for Environment Development*, (*B. Aquaculture*), *5* (1): 67-83.

El-Dakar, A. Y., Hassanien, G. D., Gad, S. S., & Sakr, S. E. (2008). Use of dried basil leaves as a feeding attractant for hybrid tilapia, *Oreochromis niloticus* X *Oreochromis aureus*, fingerlings. *Mediterranean Aquaculture Journal*, *1*(1), 35-44.

Eldar, A., Frelier, P. F., Assenta, L., Varner, P. W., Lawhon, S., & Bercovier, H. (1995a). *Streptococcus shiloi*, the name for an agent causing septicemic infection in fish, is a junior synonym of *Streptococcus iniae*. *International Journal of Systematic and Evolutionary Microbiology*, *45*(4), 840-842.

Eldar, A., Bejerano, Y., Livoff, A., Horovitcz, A., & Bercovier, H. (1995b). Experimental streptococcal meningo-encephalitis in cultured fish. *Veterinary Microbiology*, *43*(1), 33-40.

El-Hawarry, W. N. (2012). Biochemical and non-specific immune parameters of healthy Nile tilapia (*Oreochromis niloticus*), blue tilapia (*Oreochromis aureus*) and their interspecific hybrid (male *O. aureus* × female *O. niloticus*) maintained in semi-intensive culture system. *Online Journal of Animal and Feed Research*, *2*(1), 84-88.

Ellis, A. E. (1988). *General principles of fish vaccination*. London: Academic Press.

Ellis, A. E. (2001). Innate host defense mechanisms of fish against viruses and bacteria. *Developmental and Comparative Immunology*, *25*(8), 827-839.

Eloff, J. N. (1998). Which extractant should be used for the screening and isolation of antimicrobial components from plants? *Journal of Ethnopharmacology*, *60*(1), 1-8.

El-Sayed, A. F. M. (2006). *Tilapia culture.* Willingford, Oxfordshire, UK: CABI Publishing.

Etim, N. N., Williams, M. E., Akpabio, U., & Offiong, E. E. (2014). Haematological parameters and factors affecting their values. *Agricultural Science*, *2*(1), 37-47.

Evans, J. J., Klesius, P. H., Gilbert, P. M., Shoemaker, C. A., Al Sarawi, M. A., Landsberg, J., Duremdez, R., Al Marzouk, A., & Al Zenki, S. (2002). Characterization of β-haemolytic Group B *Streptococcus agalactiae* in cultured seabream, *Sparus auratus* L., and wild mullet, *Liza klunzingeri* (Day), in Kuwait. *Journal of Fish Diseases*, *25*(9), 505-513.

Fakurazi, S., Hairuszah, I., Lip, J. M., & Shanthi, G. (2008). The effect of pretreatment of zerumbone on fatty liver following ethanol induced hepatotoxicity. *Journal of Biological Sciences*, *8*(8), 1348-1351.

Fakurazi, S., Hairuszah, I., Lip, J. M., Shanthi, G., Nanthini, U., Shamima, A, Roslida, H., & Tan, Y. (2009). Hepatoprotective action of zerumbone against paracetamol induced hepatotoxicity. *Journal of Medical Sciences*, *9*(3), 161-164.

Farouk, A. E., & Benafri, A. (2007). Antibacterial activity of *Eurycoma longifolia* Jack. *Saudi Medical Journal*, *28*(9), 1422-1424.

Fauconneau, B. (1984). The measurements of whole body protein synthesis in larval and juvenile carp (*Cyprinus carpio* L.). *Comparative Biochemistry and Physiology*, *78*, 845–850.

Fazio, F., Marafioti, S., Arfuso, F., Piccione, G., & Faggio, C. (2013). Comparative study of the biochemical and haematological parameters of four wild Tyrrhenian fish species. *Veterinární medicína*, *58*(11), 576-581.

Ferguson, H. W., Morales, J. A., & Ostland, V. E. (1994). Streptococcosis in aquarium fish. *Diseases of Aquatic Organisms*, *19*(1), 1-6.

Filho, C. I., Müller, E. E., Pretto-Giordano, L. G., & Bracarense, A. P. F. R. L. (2009). Histological findings of experimental *Streptococcus agalactiae* infecion in Nile tilapias (*Oreochromis niloticus*). *Brazilian Journal of Veterinary Pathology*, *2*(1), 12-15.

Finney, D. J. (1952). *Probit analysis*. Cambridge, England: Cambridge University Press.

Finney, D. J. (1971). *Probit analysis* (3rd ed.). Cambidge England: University Press.

Fitzgerald, W. J. (1979). The red orange tilapia, a hybrid that could become a world favourite. *Fish Farming International*, *6*(1), 26-27.

Fitzsimmons, K. (1997). Introduction to tilapia nutrition. In *Tilapia Aquaculture: The International Symposium on Tilapia in Aquaculture. ICLARM Conferences Proceedings* (pp. 9-12). Florida, US.

Fitzsimmons, K. (2006). Tilapia: prospect and potential for global production. In Chhorn, L. & Carl, D. W. (Eds.), *Tilapia Biology, Culture and Nutrition* (pp. 51-72). US: The Harworth Press.

Fitzsimmons, K. (2016). Supply and demand in global tilapia markets 2016. Retrieved from https://www.was.org/meetings/showabstract.aspx?id=41768.

Food and Agriculture Organization of the United Nations. (2005). *Responsible use of antibiotics in aquaculture* (Fisheries Technical Paper No. 469). Rome, Italy: FAO.

Food and Agriculture Organization of the United Nations. (2011). *Global food losses and food waste-extent, causes and prevention*. Rome, Italy: FAO.

Food and Agriculture Organization of the United Nations. (2013). *The state of food insecurity in the world. The multiple dimensions of food security.* Rome, Italy: FAO.

Food and Agriculture Organization of the United Nations. (2014). The *state of world fisheries and aquaculture. World review of fisheries and aquaculture.* Rome, Italy: FAO.

Food and Agriculture Organization of the United Nations. (2016). *The state of world fisheries and aquaculture. Contributing to food security and nutrition for all.* Rome, Italy: FAO.

Franz, C., Baser, K. H. C., & Windisch, W. (2010). Essential oils and aromatic plants in animal feeding-a European perspective. A review. *Flavour and Fragrance Journal*, *25*(5), 327-340.

Fryer, G., & Iles, T. D. (1972). *The cichlid fishes of the Great Lakes of Africa: their biology and evolution.* Edinburgh: Oliver and Boyd.

Funatogawa, K., Hayashi, S., Shimomura, H., Yoshida, T., Hatano, T., Ito, H., & Hirai, Y. (2004). Antibacterial activity of hydrolyzable tannins derived from medicinal plants against *Helicobacter pylori*. *Microbiology and Immunology*, *48*(4), 251-261.

Gaber, H. S., El-Kasheif, M. A., Ibrahim, S. A., & Authman, M. N. (2013). Effect of water pollution in El-Rahawy drainage canal on hematology and organs of freshwater fish. *World Applied Sciences Journal*, *21*(3), 329-341.

Galman, O. R., & Avtalion, R. R. (1983). A preliminary investigation of the characteristics of red tilapias from the Philippines and Taiwan. *In Proceedings of the International Symposium on Tilapia in Aquaculture* (pp. 291–301). Nazareth, Israel, Tel Aviv.

Gao, Y., van Belkum, M. J., & Stiles, M. E. (1999). The outer membrane of Gram-negative bacteria inhibits antibacterial activity of brochocin-C. *Applied and Environmental Microbiology*, *65*(10), 4329-4333.

Gargiulo, A. M., Ceccarelli, P., Dall'Aglio, C., & Pedini, V. (1998). Histology and ultrastructure of the gut of the tilapia (*Tilapia* spp.), a hybrid teleost. *Anatomia, Histologia, Embryologia*, *27*(2), 89-94.

Gatlin, D. M., Barrows, F. T., Brown, P., Dabrowski, K., Gaylord, T. G., Hardy, R. W., Herman, E., Hu, G., Krogdahl, Å., Nelson, R., & Overturf, K. (2007). Expanding the utilization of sustainable plant products in aquafeeds: a review. *Aquaculture Research*, *38*(6), 551-579.

Goda, A. (2008). Effect of dietary Ginseng herb (Ginsana® G115) supplementation on growth, feed utilization, and hematological indices of Nile Tilapia, *Oreochromis niloticus* (L.), fingerlings. *Journal of the World Aquaculture Society*, *39*(2), 205-214.

Gómez, G. D., & Balcázar, J. L. (2008). A review on the interactions between gut microbiota and innate immunity of fish. *FEMS Immunology and Medical Microbiology*, *52*(2), 145-154.

Gopalan, C., Ramasastri, B. V., & Balasubramanian, S. C. (1984). *Nutritive value of Indian foods.* Hyderabad, India: National Institute of Nutrition (ICMR).

Grassmann, J. (2005). Terpenoids as plant antioxidants. *Vitamins and Hormones*, *72*, 505-535.

Green, R. J. (2004). *Antioxidant activity of peanut plant tissues.* (Master's thesis). Retrieved from https://repository.lib.ncsu.edu/handle/1840.16/371.

Gupta, P. D., & Birdi, T. J. (2017). Development of botanicals to combat antibiotic resistance. *Journal of Ayurveda and Integrative Medicine*, *8*, 266-275.

Hammer, K. A., Carson, C. F., & Riley, T. V. (1999). Antimicrobial activity of essential oils and other plant extracts. *Journal of Applied Microbiology*, *86*(6), 985-990.

Handa, S. S. (2008). An overview of extraction techniques for medicinal and aromatic plants. In Handa, S. S., Khanuja, S. P. S., Longo, G., & Rakesh, D. D. Eds.), *Extraction Technologies for Medicinal and Aromatic Plants* (pp. 21–52). Retrieved from https://www.unido.org/sites/default/files/200910/Extraction_technologies_for_medicinal_and_aromatic_plants_0.pdf.

Haniffa, M. A., & Abdul Kader Mydeen K. P. (2011). Evaluation of antibacterial activity of medicinal plants on fish pathogen *Aeromonas hydrophila*. *Journal of Research in Biology*, *1*, 1-5.

Han, B., Long, W. Q., He, J. Y., Liu, Y. J., Si, Y. Q., & Tian, L. X. (2015). Effects of dietary *Bacillus licheniformis* on growth performance, immunological parameters, intestinal morphology and resistance of juvenile Nile tilapia (*Oreochromis niloticus*) to challenge infections. *Fish and Shellfish Immunology*, *46*(2), 225-231.

Harada, K. (1990). Attraction activities of spices for oriental weatherfish and yellowtail. *Nippon Suisan Gakkaishi*, *56*(12), 2029-2033.

Harikrishnan, R., Balasundaram, C., Kim, M. C., Kim, J. S., Han, Y. J., & Heo, M. S. (2009). Innate immune response and disease resistance in *Carassius auratus* by triherbal solvent extracts. *Fish and Shellfish Immunology*, *27*(3), 508-515.

Harikrishnan, R., Balasundaram, C., & Heo, M. S. (2011a). Impact of plant products on innate and adaptive immune system of cultured finfish and shellfish. *Aquaculture*, *317*(1), 1-15.

Harikrishnan, R., Kim, M. C., Kim, J. S., Balasundaram, C., & Heo, M. S. (2011b). Protective effect of herbal and probiotics enriched diet on haematological and immunity status of *Oplegnathus fasciatus* (Temminck & Schlegel) against *Edwardsiella tarda. Fish and Shellfish Immunology*, *30*(3), 886-893.

Harikrishnan, R., Kim, D. H., Hong, S. H., Mariappan, P., Balasundaram, C., & Heo, M. S. (2012a). Non-specific immune response and disease resistance induced by *Siegesbeckia glabrescens* against *Vibrio parahaemolyticus* in *Epinephelus bruneus*. *Fish and Shellfish Immunology*, *33*(2), 359-364.

Harikrishnan, R., Kim, J. S., Kim, M. C., Dharaneedharan, S., Kim, D. H., Hong, S. H., Song, C.Y., Balasundaram, C. & Heo, M. S. (2012b). Effect of dietary supplementation with *Suaeda maritima* on blood physiology, innate immune response, and disease resistance in olive flounder against *Miamiensis avidus*. *Experimental Parasitology*, *131*(2), 195-203.

Harikrishnan, R., Kim, J. S., Balasundaram, C., & Heo, M. S. (2012c). Protection of *Vibrio harveyi* infection through dietary administration of *Pueraria thunbergiana* in kelp grouper, *Epinephelus bruneus*. *Aquaculture*, *324*, 27-32.

Hashimoto, G. S. O., Neto, F. M., Ruiz, M. L., Acchile, M., Chagas, E. C., Chaves, F. C. M., & Martins, M. L. (2016). Essential oils of *Lippia sidoides* and *Mentha piperita* against monogenean parasites and their influence on the hematology of Nile tilapia. *Aquaculture*, *450*, 182-186.

Hassan, W. E. (2006). *Healing herbs of Malaysia*. Kuala Lumpur, Malaysia: Federal Land Development Agency.

Hawk, P. B., Oser, B. L., & Summerson, W. H. (1954). *Practical Physiological Chemistry*. Philadelphia: The Blakiston Company.

He, X. G. (2000). Online identification of phytochemical constituents in botanical extracts by combined high-performance liquid chromatographic–diode array detection–mass spectrometric techniques. *Journal of Chromatography A*, *880*(1), 203-232.

Hernández, D. R., Pérez Gianeselli, M., & Domitrovic, H. A. (2009). Morphology, histology and histochemistry of the digestive system of South American catfish (*Rhamdia quelen*). *International Journal of Morphology*, *27*(1), 105-11.

Hibiya, T. (1982). *Atlas of fish histology*. Kodansha.

Hinkelman, K., Kempthorne, O., & Kshivsagar, A. M. (1996). *Design and analysis of experiments. Volume I: Introduction to experimental design.* Statistical Methods in Medical Research. Hoboken, New Jersey: John Wiley & Sons, Inc.

Holopainen, J. K., & Gershenzon, J. (2010). Multiple stress factors and the emission of plant VOCs. *Trends in Plant Science*, *15*(3), 176-184.

Holst, B., & Williamson, G. (2008). Nutrients and phytochemicals: from bioavailability to bioefficacy beyond antioxidants. *Current Opinion in Biotechnology*, *19*(2), 73-82.

Hrubec, T. C., Cardinale, J. L., & Smith, S. A. (2000). Hematology and plasma chemistry reference intervals for cultured tilapia (*Oreochromis* hybrid). *Veterinary Clinical Pathology*, *29*(1), 7-12.

Hu, S., Concha, C., Lin, F., & Waller, K. P. (2003). Adjuvant effect of ginseng extracts on the immune responses to immunisation against *Staphylococcus aureus* in dairy cattle. *Veterinary Immunology and Immunopathology*, *91*(1), 29-37.

Huie, C. W. (2002). A review of modern sample-preparation techniques for the extraction and analysis of medicinal plants. *Analytical and Bioanalytical Chemistry*, *373*(1), 23-30.

Hurwitz, S. H., & Meyer, K. F. (1916). Studies on the blood proteins: I. The serum globulins in bacterial infection and immunity. *Journal of Experimental Medicine*, *24*(5), 515-546.

Hutami, S., & Purnamaningsih, R. (2003). Augment clonal appointment mangga (*Curcuma mango*) through cultured in vitro. *Bull. Plasma Nutfah*, *9*(1), 39-44.

Hwang, J. H., Lee, S. W., Rha, S. J., Yoon, H. S., Park, E. S., Han, K. H., & Kim, S. J. (2013). Dietary green tea extract improves growth performance, body composition, and stress recovery in the juvenile black rockfish, *Sebastes schlegeli*. *Aquaculture International*, *21*(3), 525-538.

Ibrahim, H., Khalid, N., & Hussin, K. (2007). Cultivated gingers of peninsular Malaysia: ultilization profiles and micropropagation. *The Gardens' Bulletin Singapore*, *59*(1&2), 71-88.

Ibrahim, M. Y., Abdul, A. B., Ibrahim, T. A. T., Abdelwahab, S. I., Elhassan, M. M., & Syam, M. M. (2010). Evaluation of acute toxicity and the effect of single injected doses of zerumbone on the kidney and liver functions in Sprague Dawley rats. *African Journal of Biotechnology*, *9*(28), 4442-4450.

Ifeoma, O., & Oluwakanyinsola, S. (2013). Screening of herbal medicines for potential toxicities. In *New Insights into Toxicity and Drug Testing*. InTech.

Jamal, P., Barkat, A. A., & Amid, A. (2010). Distribution of phenolics in various Malaysian medicinal plants. *Journal of Agricultural Sciences 10*(21), 2658-2662.

Jantan, I. B., Ahmad, A. R., Ahmad, A. S., & Ali, N. A. M. (1994). A comparative study of the essential oils of five Piper species from peninsular Malaysia. *Flavour and Fragrance Journal*, *9*(6), 339-342.

Jauncey, K. (2000). Nutritional requirement. In: Beveridge, M. C. M., & McAndrew, B. J. (Eds.), *Tilapias: Biology and Exploitation* (pp. 327–375). London, UK: Kluwer Academic, Publishers.

Jeney, G. (2017). *Fish diseases: prevention and control strategies*. London, UK: Academic Press.

Jesonbabu, J., Spandana, N., & Lakshmi K, A. (2011). The potential activity of hydroxychavicol against pathogenic bacteria. *Journal of Bacteriology and Parasitology*, *2*(6), 2-5.

Jha, A. K., Pal, A. K., Sahu, N. P., Kumar, S., & Mukherjee, S. C. (2007). Haemato-immunological responses to dietary yeast RNA, ω-3 fatty acid and β-carotene in *Catla catla* juveniles. *Fish and Shellfish Immunology*, *23*(5), 917-927.

Jiang, N., Tan, N. S., Ho, B., & Ding, J. L. (2007). Respiratory protein–generated reactive oxygen species as an antimicrobial strategy. *Nature Immunology*, *8*(10), 1114.

Ji, S. C., Jeong, G. S., Im, G. S., Lee, S. W., Yoo, J. H., & Takii, K. (2007). Dietary medicinal herbs improve growth performance, fatty acid utilization, and stress recovery of Japanese flounder. *Fisheries Science*, *73*(1), 70-76.

Ji, S. C., Takaoka, O., Lee, S. W., Hwang, J. H., Kim, Y. S., Ishimaru, K., Seoka, M., Jeong, G. S., & Takii, K. (2009). Effect of dietary medicinal herbs on lipid metabolism and stress recovery in red sea bream *Pagrus major*. *Fisheries Science*, *75*(3), 665-672.

Jin, Y. B., Seo, W. D., Lee, Y. J., Lee, Y. S., & Lee, H. J. (2013). Toxicological evaluation of zerumbone on antitumor effects in mice. *African Journal of Pharmacy and Pharmacology*, *7*(8), 466-473.

Jobling M. (1994). *Fish bioenergetics*. London, UK: Chapman & Hall.

Kamel, C. (2001). Tracing modes of action and the roles of plants extracts in non-ruminants. In Garnsworthy, P. C. & Wiseman, J. (Eds.), *Recent Advances in Animal Nutrition* (pp. 135-150). Nottingham, UK: Nottingham University Press.

Karaman, I., Şahin, F., Güllüce, M., Öğütçü, H., Şengül, M., & Adıgüzel, A. (2003). Antimicrobial activity of aqueous and methanol extracts of *Juniperus oxycedrus* L. *Journal of Ethnopharmacology*, *85*(2), 231-235.

Kavitha K., Sridevi Sangeetha, K. S., Sujatha, K., & Umamaheswari, S. (2014). Phytochemical and pharmacological profile of *Justicia gendarussa* Burm f.–review. *Journal of Pharmacy Research*, *8*(7), 990-997.

Kavitha, P., & Subramanian, P. (2011). Influence of *Tribulus terrestris* on testicular enzyme in fresh water ornamental fish *Poecilia latipinna*. *Fish Physiology and Biochemistry*, *37*(4), 801-807.

Kim, S. S., & Lee, K. J. (2008). Effects of dietary kelp (*Ecklonia cava*) on growth and innate immunity in juvenile olive flounder *Paralichthys olivaceus* (Temminck et Schlegel). *Aquaculture Research*, *39*(15), 1687-1690.

Kinghorn, A. D., Pan, L., Fletcher, J. N., & Chai, H. (2011). The relevance of higher plants in lead compound discovery programs. *Journal of Natural Products*, *74*(6), 1539-1555.

Koga, A. Y., Beltrame, F. L., & Pereira, A. V. (2016). Several aspects of *Zingiber zerumbet*: a review. *Revista Brasileira de Farmacognosia*, *26*(3), 385-391.

Krishna, K. L., Mruthunjaya, K., & Patel, J. A. (2010). Antioxidant and hepatoprotective potential of stem methanolic extract of *Justicia gendarussa* Burm. *International Journal of Pharmacology*, *6*(2), 72-80.

Kumar, V., & Roy, S. (2017). Aquaculture drugs: sources, active ingredients, pharmaceutic preparations and methods of administration. *Journal of Aquaculture Research and Development*, *8*(9), 1-13.

Kurniawan, D. R., Arief, M., & Lamid, M. (2018, April). Effect of maggot (*Hermetia illucens*) flour in commercial feed on protein retention, energy retention, protein content, and fat content in tilapia (*Oreochromis niloticus*). In *IOP Conference Series: Earth and Environmental Science* (Vol. 137, No. 1, p. 012030).

Lachumy, S. J. T., Sasidharan, S., Sumathy, V., & Zuraini, Z. (2010). Pharmacological activity, phytochemical analysis and toxicity of methanol extract of *Etlingera elatior* (torch ginger) flowers. *Asian Pacific Journal of Tropical Medicine*, *3*(10), 769-774.

Laith, A. A., Ambak, M. A., Hassan, M., Sheriff, S. M., Nadirah, M., Draman, A. S., Wahab, W., Wan Ibrahim, W. N., Aznan, A. S., Jabar, A. & Najiah, M. (2017). Molecular identification and histopathological study of natural *Streptococcus agalactiae* infection in hybrid tilapia (*Oreochromis niloticus*). *Veterinary World*, *10*(1), 101-111.

Laparra, J. M., & Sanz, Y. (2010). Interactions of gut microbiota with functional food components and nutraceuticals. *Pharmacological Research*, *61*(3), 219-225.

Latiff, A. (2007). *Forestry, forest resources and forest biodiversity in Peninsular Malaysia*. Kuala Lumpur, Malaysia: Forestry Department Peninsular Malaysia.

Lavanya, G., & Brahmaprakash, G. P. (2011). Phytochemical screening and antimicrobial activity of compounds from selected medicinal and aromatic plants. *International Jornal of Science and Nature*, *2* (2): 287-291.

Lazado, C. C., & Caipang, C. M. A. (2014). Mucosal immunity and probiotics in fish. *Fish and Shellfish Immunology*, *39*(1), 78-89.

Lee, J. Y., & Gao, Y. (2012). Review of the application of garlic, *Allium sativum*, in aquaculture. *Journal of the World Aquaculture Society*, *43*(4), 447-458.

Lee, T. K., & Vairappan, C. S. (2011). Antioxidant, antibacterial and cytotoxic activities of essential oils and ethanol extracts of selected South East Asian herbs. *Journal of Medicinal Plants Research*, *5*(21), 5284-5290.

Lenfant, C., & Johansen, K. (1972). Gas exchange in gill, skin, and lung breathing. *Respiration physiology*, *14*(1-2), 211-218.

Ley, R. E., Lozupone, C. A., Hamady, M., Knight, R., & Gordon, J. I. (2008). Worlds within worlds: evolution of the vertebrate gut microbiota. *Nature Reviews Microbiology*, *6*(10), 776-788.

Li, H. B., Jiang, Y., & Chen, F. (2004). Separation methods used for *Scutellaria baicalensis* active components. *Journal of Chromatography B*, *812*(1), 277-290.

Litchfield, J. A., & Wilcoxon, F. (1949). A simplified method of evaluating dose-effect experiments. *Journal of Pharmacology and Experimental Therapeutics*, *96*(2), 99-113.

Loh, S. P., & Hadira, O. (2011). In vitro inhibitory potential of selected Malaysian plants against key enzymes involved in hyperglycemia and hypertension. *Malaysian Journal of Nutrition*, *17*(1), 77-86.

Lovell, T. (1989). *Nutrition and feeding of fish.* New York, USA: Van Nostrand Reinhold.

Lowe-McConnell, R. H. (2000). The roles of tilapias in ecosystems. In *Tilapias: Biology and Exploitation* (pp. 129-162). Netherlands: Springer.

Magid, A., & Babiker, M. M. (1975). Oxygen consumption and respiratory behaviour of three Nile fishes. *Hydrobiologia*, *46*(4), 359–367.

Magnadóttir, B. (2006). Innate immunity of fish (overview). *Fish and Shellfish Immunology*, *20*(2), 137-151.

Mahdavi, M., Hajimoradloo, A., & Ghorbani, R. (2013). Effect of *Aloe vera* extract on growth parameters of common carp (*Cyprinus carpio*).*World Journal of Medical Sciences*, *9*(1), 55-60.

Maheswaran, M. L., Padmavathy, S., & Gunalan, B. (2013). Screening and characterization of marine seaweeds and its antimicrobial potential against fish pathogens. *International Journal of Fisheries and Aquatic Studies*, *1*(1), 1-13.

Majumdar, B., Chaudhuri, S. R., Ray, A., & Bandyopadhyay, S. K. (2002). Potent antiulcerogenic activity of ethanol extract of leaf of *Piper betle* Linn by antioxidative mechanism. *Indian Journal of Clinical Biochemistry*, *17*(1), 49-57.

Malek, S. N. A., Lee, G. S., Hong, S. L., Yaacob, H., Wahab, N. A., Faizal Weber, J. F., & Shah, S. A. A. (2011). Phytochemical and cytotoxic investigations of *Curcuma mangga* rhizomes. *Molecules*, *16*(6), 4539-4548.

Manaf, S. R., & Daud, H. M. (2016). Screening of phytochemical properties and antimicrobial activity of Malaysian medicinal plants against aquatic bacteria. *Malaysian Journal of Microbiology*, *12*(4), 284-290.

Mauel, M. J., Miller, D. L., & Merrill, A. L. (2007). Hematologic and plasma biochemical values of healthy hybrid tilapia (*Oreochromis aureus*× *Oreochromis nilotica*) maintained in a recirculating system. *Journal of Zoo and Wildlife Medicine*, *38*(3), 420-424.

Mayer, F. L., & Hamelink, J. L. (1977, October). Aquatic toxicology and hazard evaluation. In *Proceedings of The First Annual Symposium on Aquatic Toxicology*. American Society for Testing & Materials. Memphis, Tennessee.

Mediani, A., Abas, F., Khatib, A., & Tan, C. P. (2013). *Cosmos caudatus* as a potential source of polyphenolic compounds: Optimisation of oven drying conditions and characterisation of its functional properties. *Molecules*, *18*(9), 10452-10464.

Metwally, M. A. A. (2009). Effects of garlic (*Allium sativum*) on some antioxidant activities in tilapia nilotica (*Oreochromis niloticus*). *World Journal of Fish and Marine Sciences*, *1*(1), 56-64.

Meyer, B. N., Ferrigni, N. R., Putnam, J. E., Jacobsen, L. B., Nichols, D. J., & McLaughlin, J. L. (1982). Brine shrimp: a convenient general bioassay for active plant constituents. *Planta Medica*, *45*(05), 31-34.

Meyer, F. P. (1991). Aquaculture disease and health management. *Journal of Animal Science*, *69*(10), 4201-4208.

Mian, G. F., Godoy, D. T., Leal, C. A. G., Yuhara, T. Y., Costa, G. M., & Figueiredo, H. C. P. (2009). Aspects of the natural history and virulence of *S. agalactiae* infection in Nile tilapia. *Veterinary Microbiology*, *136*(1), 180-183.

Micol, V., Caturla, N., Pérez-Fons, L., Más, V., Pérez, L., & Estepa, A. (2005). The olive leaf extract exhibits antiviral activity against viral haemorrhagic septicaemia rhabdovirus (VHSV). *Antiviral Research*, *66*(2), 129-136.

Miller, L. C., & Tainter, M. (1944). Estimation of the ED50 and its error by means of logarithmic-probit graph paper. *Proceedings of the Society for Experimental Biology and Medicine*, *57*(2), 261-264.

Min, B. R., Pinchak, W. E., Merkel, R., Walker, S., Tomita, G., & Anderson, R. C. (2008). Comparative antimicrobial activity of tannin extracts from perennial plants on mastitis pathogens. *Scientific Research and Essay*, *3*(2), 66-73.

Mires, D. (1995). The tilapias. In Nash, C. E., & Novotony, A. J. (Eds.), *Production of Aquatic Animals: Fishes* (pp. 133-152). New York: Elsevier.

Mohamed, F. A. (2009). Histopathological studies on *Tilapia zillii* and *Solea vulgaris* from Lake Qarun, Egypt. *World Journal of Fish and Marine Sciences*, *1*(1), 29-39.

Mohammedi, Z., & Atik, F. (2011). Impact of solvent extraction type on total polyphenols content and biological activity from *Tamarix Aphylla* (L.) Karst. *International Journal of Pharmacology and Biological Sciences*, *2*(1), 609-615.

Mona, S. N., Hossain, M. M. M., Rahman, M. Z., Alam, M. E., Rahman, M. H., Yeasmin, S. M., & Khatun, A. (2015). Protection of bacterial infection through dietary administration of *Azadirachta indica* (neem) leaf in Chinese carp after parasitic infestation. *International Journal of Fisheries and Aquatic Studies*, *2*(4), 31-37.

Monroe, S., & Polk, R. (2000). Antimicrobial use and bacterial resistance. *Current Opinion in Microbiology*, *3*(5), 496-501.

Moshawih, S., Cheema, M. S., Ahmad, Z., Zakaria, Z. A., & Hakim, M. N. (2017). A comprehensive review on *Cosmos caudatus* (ulam Raja): pharmacology, ethnopharmacology, and phytochemistry. *International Research Journal of Education and Sciences*, *1*(1), 14-31.

Mothana, R. A., & Lindequist, U. (2005). Antimicrobial activity of some medicinal plants of the Island Soqotra. *Journal of Ethnopharmacology*, *96*(1), 177-181.

Moustafa, A. M., Aly, Abd-El-Menem. A., Mesalhy, S., Elghobashy, H. A., Hasanin, S. I., Ibrahim, A. E. (2007). Influence of phenol pollution on Nile tilapia (*Oreochromis niloticus*). *Egyptian Journal of Aquatic Biology and Fisheries*, *11*(3):709-720.

Mukinda, J. T. (2005). *Acute and chronic toxicity of the flavonoid-containing plant, Artemisia afra in rodents* (Doctoral dissertation). Retrieved from http://etd.uwc.ac.za/xmlui/handle/11394/1604

Murray, A. G. (2009). Using simple models to review the application and implications of different approaches used to simulate transmission of pathogens among aquatic animals. *Preventive Veterinary Medicine*, *88*(3), 167-177.

Murray, A. L., Ponald, J. P., Alcorn, S. W., Fairgrieve, W. T., Shearer, K. D., & Roley, D. (2003). Effects of various feed supplements containing fish protein hydrolysate or fish processing by products on the innate immune functions of juvenile coho salmon (*Oncorhynchus kisutch*). *Aquaculture*, *220*(1-4), 643–653.

Musa, N., Wei, L. S., Musa, N., Hamdan, R. H., Leong, L. K., Wee, W., Amal, M. N., Kutty, B. M., & Abdullah, S. Z. (2009). Streptococcosis in red hybrid tilapia (*Oreochromis niloticus*) commercial farms in Malaysia. *Aquaculture Research*, *40*(5), 630-632.

Mustafa, R. A., Hamid, A. A., Mohamed, S., & Bakar, F. A. (2010). Total phenolic compounds, flavonoids, and radical scavenging activity of 21 selected tropical plants. *Journal of Food Science*, *75*(1), 28-35.

Najiah, M., Lee, S. W., Nadirah, M., Ruhil, H., Lee, K. L., Wendy, W., Amal, M. N. A., Basiriah, M. K., & Siti Zahrah, A. (2009). Streptococcosis in red hybrid tilapia (*Oreochromis niloticus*) commercial farms in Malaysia. *Aquaculture Research*, *40*(5), 630-632.

Najiah, M., Nadirah, M., Arief, Z., Zahrol, S., Tee, L. W., Ranzi, A. D., Amar, A. S., Laith, A. A., Mariam, M., Suzana, S., & Aida, R. J. (2011). Antibacterial activity of Malaysian edible herbs extracts on fish pathogenic bacteria. *Research Journal of Medical Plant*, *5*(6), 772-778.

Najiah, M., Wei, L. S., Seng, C. T., Wee, W., & Leong, L. K. (2008). Potential of edible plants as remedies of systemic bacterial disease infection in cultured fish. *Global Journal of Pharmacology*, *2*(2), 31-36.

Nakagawa, H., Sato, M., & Gatlin, D. M. (Eds.). (2007). *Dietary supplements for the health and quality of cultured fish*. UK: Cromwell Press.

Nalina, T., & Rahim, Z. H. A. (2007). The crude aqueous extract of *Piper betle* L. and its antibacterial effect towards *Streptococcus mutans. American Journal and Biotechnology and Biochemistry*, *3*(1), 10-15.

Nantitanon, W., Yotsawimonwat, S., & Okonogi, S. (2010). Factors influencing antioxidant activities and total phenolic content of guava leaf extract. *Food Science and Technology*, *43*(7), 1095-1103.

Nasir, B., Fatima, H., Ahmed, M., & Haq, I. U. (2015). Recent trends and methods in antimicrobial drug discovery from plant sources. *Austin Journal of Microbiology*, *1*(1), 1-12.

Naz, R., & Bano, A. (2013). Phytochemical screening, antioxidants and antimicrobial potential of *Lantana camara* in different solvents. *Asian Pacific Journal of Tropical Disease*, *3*(6), 480-486.

Nazzaro, F., Fratianni, F., De Martino, L., Coppola, R., & De Feo, V. (2013). Effect of essential oils on pathogenic bacteria. *Pharmaceuticals*, *6*(12), 1451-1474.

Ncube, N. S., Afolayan, A. J., & Okoh, A. I. (2008). Assessment techniques of antimicrobial properties of natural compounds of plant origin: current methods and future trends. *African Journal of Biotechnology*, *7*(12), 1797-1806.

Newman, D. J., & Cragg, G. M. (2012). Natural products as sources of new drugs over the 30 years from 1981 to 2010. *Journal of Natural Products*, *75*(3), 311-335.

Ng, W. K., & Hanim, R. (2007). Performance of genetically improved Nile tilapia compared with red hybrid tilapia fed diets containing two protein levels. *Aquaculture Research*, *38*(9), 965-972.

Ng, W. K., & Romano, N. (2013). A review of the nutrition and feeding management of farmed tilapia throughout the culture cycle. *Reviews in Aquaculture*, *5*(4), 220-254.

Noor Faradilla, A., & Roslinah, M. H. (2014). Isolation of allylpyrocatechol from *Piper betle* L. leaves by using high performance liquid chromatography. *Journal of Liquid Chromatography and Related Technologies*, *38*(2), 289-293.

Norazlina, M., Ehsan, S. Z., Noor Adilah, K., Lee, C. P., Farhana, E., Derick, P., Nirwana, S. I., & Norliza, M. (2013). Acute toxicity study of *Cosmos caudatus* on biochemical parameters in male rats. *Sains Malaysiana*, *42*(9), 1247-1251.

Nur-Nazifah, M. (2013). *Development and evaluation of recombinant vector cells carrying cell wall surface anchor family proteins as a vaccine against streptococcosis in red hybrid tilapia* (*Oreochromis* sp.) (Unpublished Doctoral thesis). Universiti Putra Malaysia.

Nweze, E. I., Okafor, J. I., & Njoku, O. (2004). Antimicrobial activities of methanolic extracts of *Trema guineensis* (Schumm and Thorn) and *Morinda lucida* benth used in Nigerian. *Bio-research*, *2*(1), 39-46.

Nya, E. J., & Austin, B. (2009a). Use of garlic, *Allium sativum*, to control *Aeromonas hydrophila* infection in rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Journal of Fish Diseases*, *32*(11), 963-970.

Nya, E. J., & Austin, B. (2009b). Use of dietary ginger, *Zingiber officinale* Roscoe, as an immunostimulant to control *Aeromonas hydrophila* infections in rainbow trout, *Oncorhynchus mykiss* (Walbaum). *Journal of Fish Diseases*, *32*(11), 971-977.

Nya, E. J., & Austin, B. (2011). Development of immunity in rainbow trout (*Oncorhynchus mykiss*, Walbaum) to *Aeromonas hydrophila* after the dietary application of garlic. *Fish and Shellfish Immunology*, *30*(3), 845-850.

Ogbulie, J. N., & Okpokwasili, G. C. (1999). Haematological and histological responses of *Clarias gariepinus* and *Heterobranchus bidon salis* to some bacterial disease in Rivers State, Nigeria. *Journal of National Science Foundation of Sri Lanka*, *27*(1), 1-16.

Ogunji, J., Schulz, C., & Kloas, W. (2008). Growth performance, nutrient utilization of Nile tilapia *Oreochromis niloticus* fed housefly maggot meal (magmeal) diets. *Turkish Journal of Fisheries and Aquatic Sciences*, *8*(1), 141-147.

Ojala, T., Remes, S., Haansuu, P., Vuorela, H., Hiltunen, R., Haahtela, K., & Vuorela, P. (2000). Antimicrobial activity of some coumarin containing herbal plants growing in Finland. *Journal of Ethnopharmacology*, *73*(1), 299-305.

Okechukwu, E. O. Ansa, J., & Balogun, J. K. (2007). Effects of acute nominal doses of chlorpyrifos-ethyl on some haematological indices of African catfish *Clarias gariepinus. Journal of Fisheries International*, *2*(2), 190-194.

Olmedo Sanchez J. A., Curiel Flores, A., & Orozco Hernandez, J. R. (2009). The effect of herbal growth promoter feed additive on shrimp performance. *Research Journal of Biological Sciences*, *4*(9), 1022-1024.

Oshode, O. A., Bakare, A. A., Adeogun, A. O., Efuntoye, M. O., & Sowunmi, A. A. (2008). Ecotoxicological assessment using *Clarias Gariepinus* and microbial

characterization of Leachate from Municipal solid waste landfill. *International Journal of Environmental Research*, *2*(4), 391-400.

Osuigwe, D. I., Obiekezie, A. I., & Onuoha, G. C. (2005). Some haematological changes in hybrid catfish (*Heterobranchus longifilis* x *Clarias gariepinus*) fed different dietary levels of raw and boiled jackbean (*Canavalia ensiformis*) seed meal. *African Journal of Biotechnology*, *4*(9), 1017-1021.

Padma, P. R., Lalitha, V. S., Amonkar, A. J., & Bhide, S. V. (1989). Anticarcinogenic effect of betel leaf extract against tobacco carcinogens. *Cancer Letters*, *45*(3), 195-202.

PadmaPriya, N., & Poonguzhali, T. V. (2015). Phytochemical screening and antibacterial property against human pathogenic bacteria from the leaf acetone extract of *Piper betle.* L. *Asian Journal of Biochemical and Pharmaceutical Research*, *1*(5), 251-259.

Pakravan, S., Hajimoradloo, A., & Ghorbani, R. (2012). Effect of dietary willow herb, *Epilobium hirsutum* extract on growth performance, body composition, haematological parameters and *Aeromonas hydrophila* challenge on common carp, *Cyprinus carpio*. *Aquaculture Research*, *43*(6), 861-869.

Pal, S., Ray, S. D., & Homechaudhuri, S. (2015). Evaluation of in vivo non-specific immunity and oxidative stress in *Labeo rohita* (Hamilton, 1822) infected with *Aeromonas hydrophila* as biomarker for early diagnosis. *International Journal of Fisheries and Aquatic Studies*, *3*(1), 116-124.

Pan, T. S., Yan, M. C., Chen, S. B., & Wang, X. P. (2013). Effects of ten traditional Chinese herbs on immune response and disease resistance of *Sciaenops ocellatus* (Actinopterygii: Perciformes: Sciaenidae). *Acta Ichthyologica et Piscatoria*, *43*(1) 41–49.

Pandey, A., & Tripathi, S. (2014). Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug. *Journal of Pharmacognosy and Phytochemistry*, *2*(5), 115-119.

Parekh, J., Jadeja, D., & Chanda, S. (2005). Efficacy of aqueous and methanol extracts of some medicinal plants for potential antibacterial activity. *Turkish Journal of Biology*, *29*(4), 203-210.

Park, J. H., Park, G. M., & Kim, J. K. (2015). Zerumbone, sesquiterpene photochemical from ginger, inhibits angiogenesis. *The Korean Journal of Physiology and Pharmacology*, *19*(4), 335-340.

Park, K. H., & Choi, S. H. (2012). The effect of mistletoe, *Viscum album coloratum*, extract on innate immune response of Nile tilapia (*Oreochromis niloticus*). *Fish and Shellfish Immunology*, *32*(6), 1016-1021.

Parmar, V. S., Jain, S. C., Bisht, K. S., Jain, R., Taneja, P., Jha, A. Tyagi O. D., Prasad, A. K., Wengel, J., Olsen, C. E., & Boll, P. M. (1997). Phytochemistry of the genus Piper. *Phytochemistry*, *46*(4), 597-673.

Pathiratne, A., & Rajapakshe, W., (1998). Hematological changes associated with epizootic ulcerative syndrome in the Asian cichlid fish *Etroplus suratensis*. *Asian Fisheries Science*, *11*, 203–211.

Patwardhan, B. (2005). Ethnopharmacology and drug discovery. *Journal of Ethnopharmacology*, *100*(1), 50-52.

Paval, J., Kaitheri, S. K., Potu, B. K., Govindan, S., Kumar, R. S., Narayanan, S. N., & Moorkoth, S. (2009). Anti-arthritic potential of the plant *Justicia gendarussa* Burm F. *Clinics*, *64*(4), 357-362.

Pereira, U. P., Mian, G. F., Oliveira, I. C. M., Benchetrit, L. C., Costa, G. M., & Figueiredo, H. C. P. (2010). Genotyping of *Streptococcus agalactiae* strains isolated from fish, human and cattle and their virulence potential in Nile tilapia. *Veterinary Microbiology*, *140*(1), 186-192.

Perez, C., Paul, M., & Bazerque, P. (1990). An antibiotic assay by the agar-well diffusion method. *Acta Biologiae et Medicine Experimentalis*, *15*, 113-115.

Pérez, T., Balcázar, J. L., Ruiz-Zarzuela, I., Halaihel, N., Vendrell, D., De Blas, I., & Múzquiz, J. L. (2010). Host–microbiota interactions within the fish intestinal ecosystem. *Mucosal Immunology*, *3*, 355–360.

Philip, H. E. (1984). Identification of fungicidal and nematocidal components in leaves of *Piper betle* (Piperaceae). *Journal of Agricultural and Food Chemistry*, *32*(6),1254-1256.

Philippart, J. C. L., & Ruwet, J. C. L. (1982). Ecology and distribution of tilapias. In: R. H., Lowe-Mc Connell (Eds.). *The Biology and Culture of Tilapia* (pp. 15-59). Manila, Philippines.

Pieterse, G. M., Van, Dyk, J. C. & Marchand, M. J. (2010). A histology-based fish health assessment (HBFHA) in a DDT sprayed area. In: Bornman, M. S., Barnhoorn, I. E. J., Genthe, B. DDT for malaria control: Effects in indicators and health risk. WRC Report No. *1674*(1)09.

Plumb, J. A., Schachte, J. H., Gaines, J. L., Peltier, W., & Carroll, B. (1974). *Streptococcus* sp. from marine fishes along the Alabama and northwest Florida coast of the Gulf of Mexico. *Transactions of the American Fisheries Society*, *103*(2), 358-361.

Poleksić, V., & Mitrović-Tutundžić, V. (1994). Fish gills as a monitor of sublethal and chronic effects of pollution. In: Müller, R., & R., Lloyd (Eds.). *Sublethal and chronic effects of pollutants on freshwater fish. Oxford: Fishing News Books*.

Popma, T. J., & lovshin, L. L. (1996). *Worldwide prospects for commercial production of tilapia*. Research and Development Series (No. 41). Auburn University, Auburn, Alabama: Department of Fisheries and Allied Aquaculture.

Pradhan, D., Suri, K. A., Pradhan, D. K., & Biswasroy, P. (2013). Golden Heart of the Nature: *Piper betle* L. *Journal of Pharmacognosy and Phytochemistry*, *1*(6), 147-167.

Pratheepa, V., & Sukumaran, N. (2011). Specific and nonspecific immunostimulation study of *Euphorbia hirta* on *Pseudomonas fluorescens*-infected *Cyprinus carpio*. *Pharmaceutical Biology*, *49*(5), 484-491.

Pretto-Giordano, L. G., Müller, E. E., Freitas, J. C. D., & Silva, V. G. D. (2010). Evaluation on the pathogenesis of *Streptococcus agalactiae* in Nile tilapia (*Oreochromis niloticus*). *Brazilian Archives of Biology and Technology*, *53*(1), 87-92.

Pujimulyani, D., Wazyka, A., Anggrahini, S., & Santoso, U. (2014). Antioxidative properties of white saffron extract (*Curcuma mangga* Val) in The B-Carotene Bleaching and DPPH-Radical Scavenging Methods. *Indonesian Food and Nutrition Progress*, *11*(2), 35-40.

Putra, A., Santoso, U., Lee, M. C., & Nan, F. H. (2013). Effects of dietary katuk leaf extract on growth performance, feeding behavior and water quality of grouper *Epinephelus coioides. Aceh International Journal of Science and Technology*, *2*(1), 17-25.

Raaman, N. (2006). *Phytochemical techniques* (2nd ed.) New Delhi: New Indian Publishing Agency.

Raghu M. G., & Agrawal P. (2016). Review on *Justicia gendaruss* and its applications. *International Journal of Advanced Scientific Research and Publications*, *2*(1), 5-8.

Rahalison, L. L., Hamburger, M., Hostettmann, K., Monod, M., & Frenk, E. (1991). A bioautographic agar overlay method for the detection of antifungal compounds from higher plants. *Phytochemical Analysis*, *2*(5), 199-203.

Rahman, T., Akanda, M. M. R., Rahman, M. M., & Chowdhury, M. B. R. (2009). Evaluation of the efficacies of selected antibiotics and medicinal plants on common bacterial fish pathogens. *Journal of the Bangladesh Agricultural University*, *7*(1), 163-168.

Rajeh, M. A. B., Zuraini, Z., Sasidharan, S., Latha, L. Y., & Amutha, S. (2010). Assessment of *Euphorbia hirta* L. leaf, flower, stem and root extracts for their antibacterial and antifungal activity and brine shrimp lethality. *Molecules*, *15*(9), 6008-6018.

Raman, R. P. (2017). Applicability, feasibility and efficacy of phytotherapy in aquatic animal health management. *American Journal of Plant Sciences*, *8*, 257-287.

Rambhaskar, B., & Srinivasa Rao, K. (1987). Comparative haematology of ten species of marine fish from Visakhapatnam Coast. *Journal of Fish Biology*, *30*(1), 59-66.

Ramji, N., Ramji, N., Iyer, R., & Chandrasekaran, S. (2002). Phenolic antibacterials from *Piper betle* in the prevention of halitosis. *Journal of Ethnopharmacology*, *83*(1), 149-152.

Rani, P., & Khullar, N. (2004). Antimicrobial evaluation of some medicinal plants for their anti-enteric potential against multi-drug resistant *Salmonella typhi*. *Phytotherapy Research*, *18*(8), 670-673.

Rao, Y. V., Das, B. K., Jyotyrmayee, P., & Chakrabarti, R. (2006). Effect of *Achyranthes aspera* on the immunity and survival of *Labeo rohita* infected with *Aeromonas hydrophila*. *Fish and Shellfish Immunology*, *20*(3), 263-273.

Rasdi, N. H. M., Samah, O. A., Sule, A., & Ahmed, Q. U. (2010). Antimicrobial studies of *Cosmos caudatus* kunth. (compositae). *Journal of Medicinal Plants Research*, *4*(8), 669-673.

Rathee, J. S., Patro, B. S., Mula, S., Gamre, S., & Chattopadhyay, S. (2006). Antioxidant activity of *Piper betel* leaf extract and its constituents. *Journal of Agricultural and Food Chemistry*, *54*(24), 9046-9054.

Rattanachaikunsopon, P., & Phumkhachorn, P. (2009). Protective effect of clove oil-supplemented fish diets on experimental *Lactococcus garvieae* infection in tilapia. *Bioscience, Biotechnology, and Biochemistry*, *73*(9), 2085-2089.

Rawat, A. K. S., Tripathi, R. D., Khan, A. J., & Balasubrahmanyam, V. R. (1989). Essential oil components as markers for identification of *Piper betle* L. cultivars. *Biochemical Systematics and Ecology*, *17*(1), 35-38.

Rawling, M. D., Merrifield, D. L., & Davies, S. J. (2009). Preliminary assessment of dietary supplementation of Sangrovit® on red tilapia (*Oreochromis niloticus*) growth performance and health. *Aquaculture*, *294*(1), 118-122.

Reed, L. J., & Muench, H. (1938). A simple method of estimating fifty per cent endpoints. *American Journal of Epidemiology*, *27*(3), 493-497.

Řehulka, J. (1989). Determining the optimum doses of *Kurasan* (ethoxiquinolin) and butylhydroxytoluol (BHT) in dry pellets: effect of both anti-oxidants on some haematological and condition parameters of rainbow trout, *Salmo gairdneri* Richardson. *Aquaculture Research*, *20*(3), 295-310.

Řehulka, J. (2000). Influence of astaxanthin on growth rate, condition and some blood indices of rainbow trout, *Oncorhynchus mykiss*. *Aquaculture*, *190*(1-2), 27-47.

Řehulka, J. (2002). Aeromonas causes severe skin lesions in rainbow trout (*Oncorhynchus mykiss*): clinical pathology, haematology, and biochemistry. *Acta Veterinaria Brno*, *71*(3), 351-360.

Rekha, V. P. B., Kollipara, M., Gupta, B. S., Bharath, Y., & Pulicherla, K. K. (2014). A Review on *Piper betle* L.: natureâ s promising medicinal reservoir. *American Journal of Ethnomedicine*, *1*(5), 276-289.

Rey Vázquez, G. R., & Guerrero, G. A. (2007). Characterization of blood cells and hematological parameters in *Cichlasoma dimerus* (Teleostei, Perciformes). *Tissue and cell*, *39*(3), 151-160.

Reverter, M., Bontemps, N., Lecchini, D., Banaigs, B., & Sasal, P. (2014). Use of plant extracts in fish aquaculture as an alternative to chemotherapy: current status and future perspectives. *Aquaculture*, *433*, 50-61.

Rimando, A. M., Han, B. H., Park, J. H., & Cantoria, M. C. (1986). Studies on the constituents of Philippine *Piper betle* leaves. *Archives of Pharmacal Research*, *9*(2), 93-97.

Ringø, E., Olsen, R. E., Gifstad, T. Ø., Dalmo, R. A., Amlund, H., Hemre, G. I., & Bakke, A. M. (2010). Prebiotics in aquaculture: a review. *Aquaculture Nutrition*, *16*(2), 117-136.

Roberts, R. J. (2012). *Fish Pathology*. Oxford, UK: Wiley-Blackwell.

Robinson, J. A., & Meyer, F. P. (1966). Streptococcal fish pathogen. *Journal of Bacteriology*, *92*(2), 512.

Rodkhum, C., Kayansamruaj, P., Pirarat, N., Zhou, W., Liu, Y., & Chen, G. H. (2011). Effect of water temperature on susceptibility to *Streptococcus agalactiae* serotype Ia infection in Nile tilapia (*Oreochromis niloticus*). *The Thai Journal of Veterinary Medicin*e, *41*(3), 309.

Romão, S., Donatti, L., Freitas, M. O., Teixeira, J., & Kusma, J. (2006). Blood parameter analysis and morphological alterations as biomarkers on the health of *Hoplias malabaricus* and *Geophagus brasiliensis*. *Brazilian Archives of Biology and Technology*, *49*(3), 441-448.

Romero, J., Feijoó, C. G., & Navarrete, P. (2012). Antibiotics in aquaculture–use, abuse and alternatives. In *Health and Environment in Aquaculture*. InTech.

Roomiani, L., Soltani, M., Basti, A. A., Mahmoodi, A., Mirghaed, A. T., & Yadolahi, F. (2013). Evaluation of the chemical composition and in vitro antimicrobial activity of *Rosmarinus officinalis*, *Zataria multiflora*, *Anethum graveolens* and *Eucalyptus globulus* against *Streptococcus iniae*; the cause of zoonotic disease in farmed fish. *Iranian Journal of Fisheries Sciences*, *12*(3), 702-716.

Ross, L. G. (2000). *Environmental physiology and energetics.* Fish and Fisheries (Series 25, pp. 89–128). Netherlands: Kluwer Academic Publishers.

Rukayadi, Y., Shim, J. S., & Hwang, J. K. (2008). Screening of Thai medicinal plants for anticandidal activity. *Mycoses*, *51*(4), 308-312.

Rumiyati, S., Jenie, U. A., Mubarika, S., & Kardono, L. B. (2007). Selective cytotoxicity of essential oil of *C. mangga* Val. on cell lines and its effect on expressions of p53 and Bcl-2. *In Proceeding of the International Symposium on Recent Progress in Curcumin Research.* Gadjah Mada University, Indonesia.

Ruslay, S., Abas, F., Shaari, K., Zainal, Z., Sirat, H., Israf, D. A., & Lajis, N. H. (2007). Characterization of the components present in the active fractions of health gingers (*Curcuma xanthorrhiza* and *Zingiber zerumbet*) by HPLC–DAD–ESIMS. *Food Chemistry*, *104*(3), 1183-1191.

Sahu, S., Das, B. K., Mishra, B. K., Pradhan, J., & Sarangi, N. (2007). Effect of *Allium sativum* on the immunity and survival of *Labeo rohita* infected with *Aeromonas hydrophila*. *Journal of Applied Ichthyology*, *23*(1), 80-86.

Sahu, S., Das, B. K., Mishra, B. K., Pradhan, J., Samal, S. K., & Sarangi, N. (2008). Effect of dietary *Curcuma longa* on enzymatic and immunological profiles of rohu, *Labeo rohita* (Ham.), infected with *Aeromonas hydrophila*. *Aquaculture Research*, *39*(16), 1720-1730.

Sakr, S. E. (2003). *Studies on the feeding attractants for fish* (Master's thesis). Suez-Canal University, El-Arish, Egypt.

Salvador, R., Muller, E. E., Freitas, J. C. D., Leonhadt, J. H., Pretto-Giordano, L. G., & Dias, J. A. (2005). Isolation and characterization of *Streptococcus* spp. group B in Nile tilapias (*Oreochromis niloticus*) reared in hapas nets and earth nurseries in the northern region of Parana State, Brazil. *Ciência Rural*, *35*(6), 1374-1378.

Samir, F., Amal, D., Hafedh, H., Kais, M., Néji, G., & Adel., K. (2017). Solvent extraction effects on phytochemical constituents profiles, antioxidant and antimicrobial activities and functional group analysis of *Ecballium elaterium* seeds and peels fruits. *Food Science and Technology*, *37*(3), 483-492.

Sanubol, A., Chaveerach, A., Sudmoon, R., Tanee, T., Noikotr, K., & Chuachan, C. (2014). Betel-like-scented Piper plants as diverse sources of industrial and medicinal aromatic chemicals. *Chiang Mai Journal of Science*, *41*(5), 1171–1181.

Sarkar, D., Saha, P., Gamre, S., Bhattacharjee, S., Hariharan, C., Ganguly, S., Sen, R., Mandal., G., Chattopadhyay, S., Majumdard, S., & Chatterjee, M. (2008). Anti-inflammatory effect of allylpyrocatechol in LPS-induced macrophages is mediated by suppression of iNOS and COX-2 via the NF-κB pathway. *International Immunopharmacology*, *8*(9), 1264-1271.

Sasidharan, S., Chen, Y., Saravanan, D., Sundram, K. M., & Latha, L. Y. (2011). Extraction, isolation and characterization of bioactive compounds from plants' extracts. *African Journal of Traditional, Complementary and Alternative Medicines*, *8*(1), 1-10.

Sasikumar, B. (2005). Genetic resources of Curcuma: diversity, characterization and utilization. *Plant Genetic Resources*, *3*(2), 230-251.

Satheeshkumar, P., Ananthan, G., Senthilkumar, D., Khan, A. B., & Jeevanantham, K. (2012). Comparative investigation on haematological and biochemical studies on wild marine teleost fishes from Vellar estuary, southeast coast of India. *Comparative Clinical Pathology*, *21*(3), 275-281.

Sathya Bama, S., Jayasurya Kingsley, S., Sankaranarayanan, S., & Bama, P. (2012). Antibacterial activity of different phytochemical extracts from the leaves of T. procumbens Linn.: identification and mode of action of the terpenoid compound as antibacterial. *International Journal of Pharmacy and Pharmaceutical Science*, *4*(1), 557-564.

Scherrer, R., & Gerhardt, P. (1971). Molecular sieving by the *Bacillus megaterium* cell wall and protoplast. *Journal of Bacteriology*, *107*(3), 718-735.

Schaperclaus, W., Kulow, H., & Schreckenback K., (1992). *Fish Disease. Vol. (1).* Rotterdam: CRC Press.

Schlenk, D., & Benson, W. H. (2003). *Target organ toxicity in marine and freshwater teleosts: organs*. New York: Taylor & Francis.

Sen, S., Chakraborty, R., & De, B. (2011). Challenges and opportunities in the advancement of herbal medicine: India's position and role in a global context. *Journal of Herbal Medicine*, *1*(3), 67-75.

Service, R. F. (1995). Antibiotics that resist resistance. *Science*, *270*(5237), 724-727.

Shafiei, S. N. S. (2012). *In-vitro antibacterial activity and phytochemical screening of bioactive compounds from guava (Psidium guajava L.) crude leaf extracts* (Master's thesis). Universiti Putra Malaysia.

Sharifah Raina, M. (2016). *Assessment of therapeutic and antimicrobial effects of local plants on bacterial diseases in Red hybrid tilapia* (*Oreochromis sp.*) (PhD thesis). Universiti Putra Malaysia.

Shah, P. M. (2005). The need for new therapeutic agents: what is in the pipeline?. *Clinical Microbiology and Infection*, *11*(3), 36-42.

Shahkar, E., Park, G., Lee, D. M., Kwak, S., & Bai, S. C. (2015). Effects of dietary Macsumsuk® supplementation on growth performance, haematological parameters, disease resistance and body composition of juvenile Nile tilapia, *Oreochromis niloticus* L. *Journal of Applied Animal Researc*h, *43*(2), 125-130.

Shahverdi, A. R., Abdolpour, F., Monsef-Esfahani, H. R., & Farsam, H. (2007). A TLC bioautographic assay for the detection of nitrofurantoin resistance reversal compound. *Journal of Chromatography B*, *850*(1), 528-530.

Shalaby, A. M., Khattab, Y. A., & Abdel Rahman, A. M. (2006). Effects of Garlic (*Allium sativum*) and chloramphenicol on growth performance, physiological parameters and survival of Nile tilapia (*Oreochromis niloticus*). *Journal of Venomous Animals and Toxins Including Tropical Diseases*, *12*(2), 172-201.

Shalaby, S. M. (2004). Response of Nile tilapia, *Oreochromis niloticus*, fingerlings to diets supplemented with differennt levels of fenugreek seeds (Hulba). *Mansoura University Journal of Agricultural Sciences*, *29* (5): 2231-2242.

Sheikhlar, A. (2012). *Medicinal herbs as growth and health promoters in African catfish (Clarias gariepinus, burchell)* (PhD thesis). Universiti Putra Malaysia.

Shelton, W. L. (2002). Tilapia culture in the 21st century. In Guerrero, R. D., & Guerrero-del Castillo, Los Banos, M. R., *Tilapia Farming in the 21st Century* (pp. 1-20).

Shibata, H., Kondo, K., Katsuyama, R., Kawazoe, K., Sato, Y., Murakami, K., Takaishi, Y., Arakaki, N., & Higuti, T. (2005). Alkyl gallates, intensifiers of β-lactam susceptibility in methicillin-resistant *Staphylococcus aureus*. *Antimicrobial Agents and Chemotherapy*, *49*(2), 549-555.

Shitut, S., Pandit, V., & Mehta, B. K. (1999). The antimicrobial efficiency of *Piper betle* Linn leaf (stalk) against human pathogenic bacteria and phytopathogenic fungi. *Central European Journal of Public Health*, *7*(3), 137-139.

Shui, G., Leong, L. P., & Wong, S. P. (2005). Rapid screening and characterisation of antioxidants of *Cosmos caudatus* using liquid chromatography coupled with mass spectrometry. *Journal of Chromatography B*, *827*(1), 127-138.

Sidahmed, H. M. A., Hashim, N. M., Abdulla, M. A., Ali, H. M., Mohan, S., Abdelwahab, S. I., Taha, M. M. E., Fai, L. M., & Vadivelu, J. (2015). Antisecretory, gastroprotective, antioxidant and anti-helicobcter pylori activity of zerumbone from *Zingiber Zerumbet* (L.) Smith. *Plos One*, *10*(3), 1-21.

Silva, N. C. C., & Fernandes Júnior, A. (2010). Biological properties of medicinal plants: a review of their antimicrobial activity. *Journal of Venomous Animals and Toxins Including Tropical Diseases*, *16*(3), 402-413.

Singgih, M., Damayanti, S., & Pandjaitan, N. (2014). Antimicrobial activity of standardized *Piper betel* extract and its mouthwash preparation. *International Journal of Pharmacy and Pharmaceutical Science*, *6*(7), 243-246.

Singh, G. D., Ganjoo, M., Youssouf, M. S., Koul, A., Sharma, R., Singh, S., Sangwan, P. L., Koul, D. B., Ahamad, D. B., & Johri, R. K. (2009). Sub-acute toxicity evaluation of an aqueous extract of *Labisia pumila*, a Malaysian herb. *Food and Chemical Toxicology*, *47*(10), 2661-2665.

Singtongratana, N., Vadhanasin, S., & Singkhonrat, J. (2013). Hydroxychavicol and eugenol profiling of betel leaves from *Piper betle* L. obtained by liquid-liquid extraction and supercritical fluid extraction. *Kasetsart Journal-Natural Science*, *47*, 614-623.

Siti-Zahrah, A., Padilah, B., Azila, A., Rimatulhana, R. and Shahidan, H. (2005). Multiple streptococcal species infection in cage-cultured red tilapia, but showing similar clinical signs. In Bondad-Reantaso, M. G., Mohan, C. V., Crumlish, M. & Subasinghe, R. P. (Eds.), *Proceedings of the Sixth Symposium on Diseases in Asian Aquaculture*. (pp. 332-339). Colombo, Sri Lanka.

Siti-Zahrah, A., Padilah, B., Azila, A., Rimatulhana, R. and Shahidan, H. (2008). Multiple streptococcal species infection in cage-cultured red tilapia, but showing similar clinical signs. In Bondad-Reantaso, M. G., Mohan, C. V., Crumlish, M. & Subasinghe, R. P. (Eds.), *Diseases in Asian Aquaculture VI*. (pp. 313-320). Manila, Philippines.

Smith, P., Hiney, M. P., & Samuelsen, O. B. (1994). Bacterial resistance to antimicrobial agents used in fish farming: a critical evaluation of method and meaning. *Annual Review of Fish Diseases*, *4*, 273-313.

Sofowora, A. (1993). *Medicinal plants and traditional medicine in Africa* (2nd ed.). New York: John Wiley and Sons.

Soivio, A., & Oikari, A. (1976). Haematological effects of stress on a teleost, *Esox lucius* L. *Journal of Fish Biology*, *8*(5), 397-411.

Soliman, W. S., Marzouk, M. S., Abdelaziz, M., Nermeen M., Abu-Elala, Abbas, H. H., Mona S. Z., & Sahr B.A. (2017). Trial on using some of herbal extracts as promising immunoprophylaxis feed additives in cultured *Oreochromis niloticus*. *Egypt Journal of Veterinary Science*, *48*(2), 53- 60.

Somchit, M. N., Reezal, I., Nur, I. E., & Mutalib, A. R. (2003). In vitro antimicrobial activity of ethanol and water extracts of *Cassia alata*. *Journal of Ethnopharmacology*, *84*(1), 1-4.

Soosean, C., Marimuthu, K., Sudhakaran, S., & Xavier, R. (2010). Effects of mangosteen (*Garcinia mangostana* L.) extracts as a feed additive on growth and haematological parameters of African catfish (*Clarias gariepinus*) fingerlings. *European Review for Medical and Pharmacological Sciences*, *14*, 605-611.

Sovlo, A., & Nikinmaa, M. (1981). The swelling of erythrocytes in relation to the oxygen affinity of the blood of the rainbow trout *Salmo gairdnerl* Richardson. In Picketing, A.D. (Ed.), *Stress and fish* (pp. 103-119). London, UK: Academic Press.

Srivastava, A. K. (1968). Studies on the hematology of certain freshwater teleosts. V. Thrombocytes and clotting of blood. *Anatomischer Anzeiger*, *124*(4), 368-374.

Steckert, L. D., Cardoso, L., Jerônimo, G. T., de Pádua, S. B., & Martins, M. L. (2018). Investigation of farmed Nile tilapia health through histopathology. *Aquaculture*, *486*, 161-169.

Suanyuk, N., Kanghear, H., Khongpradit, R., & Supamattaya, K. (2005). *Streptococcus agalactiae* infection in tilapia (*Oreochromis niloticus*). *Songklanakarin Journal of Science and Technology*, *27*, 307-319.

Suanyuk, N., Kong, F., Ko, D., Gilbert, G. L., & Supamattaya, K. (2008). Occurrence of rare genotypes of *Streptococcus agalactiae* in cultured red tilapia *Oreochromis* sp. and Nile tilapia *O. niloticus* in Thailand-Relationship to human isolates? *Aquaculture*, *284*(1), 35-40.

Subramanian, N., Jothimanivannan, C., & Moorthy, K. (2012). Antimicrobial activity and preliminary phytochemical screening of *Justicia gendarussa* (Burm. f.) against human pathogens. *Asian Journal of Pharmaceutical and Clinical Research*, *5*(3), 229-233.

Sugumaran, M., Gandhi, S., Sankarnatayanen, M., Yokesh, M., Poornime, M., & Rajasekhar, S. M. (2011). Chemical composition and antimicrobial activity of vellaikodi variety of *Piper betle* Linn leaf oil against dental pathogens. *International Journal of PharmTech Research*, *3*, 2135-2139.

Sukrasno, S., Fidriany, I., Anggadiredja, K., Handayani, W. A., & Anam, K. (2011). Influence of drying method on flavonoid content of *Cosmos caudatus* (Kunth) leaves. *Research Journal of Medicinal Plant*, *5*(2),189-195.

Sulaiman, M. R., Mohamad, T. A. S. T., Mossadeq, W. M. S., Moin, S., Yusof, M., Mokhtar, A. F., Zakaria, Z. A., Israf, D. A., & Lajis, N. (2010). Antinociceptive activity of the essential oil of *Zingiber zerumbet*. *Planta Medica*, *76*(02), 107-112.

Sumazian, Y., Syahida, A., Hakiman, M., & Maziah, M. (2010). Antioxidant activities, flavonoids, ascorbic acid and phenolic content of Malaysian vegetables. *Journal of Medicinal Plants Research*, *4*(10), 881-890.

Sutili, F. J., Gatlin, D. M., Heinzmann, B. M., & Baldisserotto, B. (2017). Plant essential oils as fish diet additives: benefits on fish health and stability in feed. *Reviews in Aquaculture*, *2017*. doi:10.1111/raq.12197.

Svobodova, Z., Fravda, D. and Palakova, J. (1991). *Unified methods of haematological examination of fish.* Vodnany: Research Institute of Fish Culture and Hydrobiology.

Taha, M. M. E., Abdul, A. B., Abdullah, R., Ibrahim, T. A. T., Abdelwahab, S. I., & Mohan, S. (2010). Potential chemoprevention of diethylnitrosamine-initiated and 2-acetylaminofluorene-promoted hepatocarcinogenesis by zerumbone from the rhizomes of the subtropical ginger (*Zingiber zerumbet*). *Chemico-Biological Interactions*, *186*(3), 295-305.

Takada, Y., Murakami, A., & Aggarwal, B. B. (2005). Zerumbone abolishes NF-κB and IκBα kinase activation leading to suppression of antiapoptotic and metastatic gene expression, upregulation of apoptosis, and downregulation of invasion. *Oncogene*, *24*(46), 6957-6969.

Takashima, F., & Hibiya, T. (1995). *An atlas of fish histology: normal and pathological features* (2nd ed.).

Takei, M. (1967). Studies on fishes' favorite foods: I. Feeding tests of carp, yellowtail and rainbow trout. *Bulletin of the Tokai Regional Fisheries Research Laboratory , 49*, 119- 129.

Takeuchi, T., Lu, J. U. N., Yoshizaki, G., & Satoh, S. (2002). Effect on the growth and body composition of juvenile tilapia *Oreochromis niloticus* fed raw Spirulina. *Fisheries Science*, *68*(1), 34-40.

Talpur, A. D. (2014). *Mentha piperita* (Peppermint) as feed additive enhanced growth performance, survival, immune response and disease resistance of Asian

seabass, *Lates calcarifer* (Bloch) against *Vibrio harveyi* infection. *Aquaculture*, 420, 71-78.

Talpur, A. D., & Ikhwanuddin, M. (2012). Dietary effects of garlic (*Allium sativum*) on haemato-immunological parameters, survival, growth, and disease resistance against *Vibrio harveyi* infection in Asian sea bass, *Lates calcarifer* (Bloch). *Aquaculture*, *364*, 6-12.

Talpur, A. D., & Ikhwanuddin, M. (2013). *Azadirachta indica* (neem) leaf dietary effects on the immunity response and disease resistance of Asian seabass, *Lates calcarifer* challenged with *Vibrio harveyi*. *Fish and Shellfish Immunology*, *34*(1), 254-264.

Talpur, A. D., Ikhwanuddin, M., & Bolong, A. M. A. (2013). Nutritional effects of ginger (*Zingiber officinale* Roscoe) on immune response of Asian sea bass, *Lates calcarifer* (Bloch) and disease resistance against *Vibrio harveyi*. *Aquaculture*, *400*, 46-52.

Tan, Y. J., & Tong, H. Y. (1989). The status of the exotic aquatic organisms in China. In De Silva, S. S. (Ed.), *Exotic Aquatic Organisms in Asia*. *Proceedings of a Workshop on Introduction of Exotic Aquatic Organisms in Asia* (Special Publication No. 3, pp. 35-43). Asian Fisheries Society, Manila, Philippines.

Temesgen, G. G. M. (2004). *Utilization of Gliricidia sepium leaf meal as protein source in diets of Mozambique tilapia, Oreochromis Mossambicus* (*Piscs: Cichlidae*) (unpublished Doctoral thesis). Universiti Putra Malaysia.

Tewari, D., & Kumar, M. (2014) Formulation and comparative evaluation of different Sitopaladi herbal syrups. *Pharmacia Lettre*, *6*(2):178-183.

Thabile, P. N. (2008). *Antimicrobial properties of selected Asian herbs.* (Master's thesis) University of Florida, USA.

Thilsted, S. H., James, D., Toppe, J., Subasinghe, R., & Karunasagar, I. (2014). Maximizing the contribution of fish to human nutrition. *In ICN2 Second International Conference on Nutrition.* FAO and World Health Organisation.

Timbrell, J. (2002). *Introduction to Toxicology* (3rd ed.). London: Taylor & Francis.

Tomlinson, T. R., & Akerele, O. (2015). *Medicinal plants: their role in health and biodiversity*. Philadelphia: University of Pennsylvania Press.

Tortora, G. J., Funke, B. R., & Case, C. L. (2001). *Microbiology: an introduction*. San Francisco, USA: Benjamin Cummings Publishing.

Trease, G. E., & Evans, W. C. (2002). *Pharmacognosy*. (15th ed.). London, UK: Saunders Publisher.

Trewavas, E. (1982). Tilapia: taxonomy and speciation. In: Pullin, R. S. V. & Lowe-McConnel, R. H. (Eds.), *The Biology and Culture of Tilapias*. ICLARM Conference Proceedings (No.7, pp. 3-13). Manila, Philippines.

Turan, F. (2006). Improvement of growth performance in tilapia (*Oreochromis aureus* Linnaeus) by supplementation of red clover *Trifolium pratense* in diets. *The Israeli Journal of Aquaculture*, *58*(1), 34-38.

Turan, F., Yıldırım, Y. B., & Gezer, A. (2012). Use of dietary Pelargonium sidoides extract to improve growth and body composition of narrow-clawed crayfish

Astacus leptodactylus Eschscholtz, 1823 juveniles. *Turkish Journal of Fisheries and Aquatic Sciences*, *12*(2), 233-238.

Valgas, C., Souza, S. M. D., Smânia, E. F., & Smânia Jr, A. (2007). Screening methods to determine antibacterial activity of natural products. *Brazilian Journal of Microbiology*, *38*(2), 369-380.

Van Hai, N. (2015). The use of medicinal plants as immunostimulants in aquaculture: a review. *Aquaculture*, *446*, 88-96.

Virgili, F., & Marino, M. (2008). Regulation of cellular signals from nutritional molecules: a specific role for phytochemicals, beyond antioxidant activity. *Free Radical Biology and Medicine*, *45*(9), 1205-1216.

Wahab, I. R. A., Blagojević, P. D., Radulović, N. S., & Boylan, F. (2011). Volatiles of *Curcuma mangga* Val. & Zijp (Zingiberaceae) from Malaysia. *Chemistry and Biodiversity*, *8*(11), 2005-2014.

Wang, M., & Lu, M. (2016). Tilapia polyculture: a global review. *Aquaculture Research*, *47*(8), 2363-2374.

Wassom, D. L., & Kelly, E. A. B. (1990). The role of the major histocompatibility complex in resistance to parasite infections. *Critical Reviews in Immunology*, *10*(1), 31-52.

Watanabe, T., Ohno, I., Wakiyama, N., Kusai, A., & Senna, M. (2002). Stabilization of amorphous indomethacin by co-grinding in a ternary mixture. *International Journal of Pharmaceutics*, *241*(1), 103-111.

Welcomme, R. L. (1988). *International introductions of Inland aquatic species*. FAO Fish. Technical Paper. Rome, Italy: FAO.

Welker, T. L., & Lim, C. (2011). Use of probiotics in diets of tilapia. *Journal of Aquaculture Research and Development*, *2011*. doi: 10.4172/2155-9546.S1-014

Wiegertjes, G. F., Stet, R. M., Parmentier, H. K., & van Muiswinkel, W. B. (1996). Immunogenetics of disease resistance in fish: a comparative approach. *Developmental and Comparative Immunology*, *20*(6), 365-381.

Wijayati, A. (2004). *Efficacy of some herbs on controlling vibrio spp. and their toxicity to Black Tiger Shrimp* (*Penaeus monodon Fabricius*) *postlarva* (Master's thesis). Kasetsart University.

Wink, M. (2015). Modes of action of herbal medicines and plant secondary metabolites. *Medicines*, *2*(3), 251-286.

Wirotesangthong, M., Inagaki, N., Tanaka, H., Thanakijcharoenpath, W., & Nagai, H. (2008). Inhibitory effects of Piper betle on production of allergic mediators by bone marrow-derived mast cells and lung epithelial cells. *International Immunopharmacology*, *8*(3), 453-457.

Witkowska, A. M., Hickey, D. K., Alonso-Gomez, M., & Wilkinson, M. (2013). Evaluation of antimicrobial activities of commercial herb and spice extracts against selected food-borne bacteria. *Journal of Food Research*, *2*(4), 37-54.

Wong, S., & Rawls, J. F. (2012). Intestinal microbiota composition in fishes is influenced by host ecology and environment. *Molecular Ecology*, *21*(13), 3100-3102.

Wongsathein, D. (2012). Factors affecting experimental *Streptococcus agalactiae* infection in tilapia, *Oreochromis niloticus* (Doctoral dissertation, University of Sterling). Retrieved from http://dspace.stir.ac.uk/handle/1893/10375

Wu, Y. R., Gong, Q. F., Fang, H., Liang, W. W., Chen, M., & He, R. J. (2013). Effect of *Sophora flavescens* on non-specific immune response of tilapia (GIFT *Oreochromis niloticus*) and disease resistance against *Streptococcus agalactiae*. *Fish and Shellfish Immunology*, *34*(1), 220-227.

Yang, C., Chowdhury, M. A., Huo, Y., & Gong, J. (2015). Phytogenic compounds as alternatives to in-feed antibiotics: potentials and challenges in application. *Pathogens*, *4*(1), 137-156.

Yanong, R. P., & Francis-Floyd, R. (2002). Streptococcal infections of fish. Report from University of Florida. Series from the Department of Fisheries and Aquatic Sciences, Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida.

Ye, X., Li, J., Lu, M., Deng, G., Jiang, X., Tian, Y., Quan, Y., & Jian, Q. (2011). Identification and molecular typing of Streptococcus agalactiae isolated from pond-cultured tilapia in China. *Fisheries Science*, *77*(4), 623-632.

Yob, N. J., Jofrry, S. M., Affandi, M. M. R., Teh, L. K., Salleh, M. Z., & Zakaria, Z. A. (2011). *Zingiber zerumbet* (L.) Smith: a review of its ethnomedicinal, chemical, and pharmacological uses. *Evidence-Based Complementary and Alternative Medicine, 2011*, 543216. doi:10.1155/2011/543216

Yu, J. H., Han, J. J., & Park, S. W. (2010). Haematological and biochemical alterations in Korean catfish, *Silurus asotus*, experimentally infected with *Edwardsiella tarda*. *Aquaculture Research*, *41*(2), 295-302.

Zahran, E., Risha, E., AbdelHamid, F., Mahgoub, H. A., & Ibrahim, T. (2014). Effects of dietary *Astragalus polysaccharides* (APS) on growth performance, immunological parameters, digestive enzymes, and intestinal morphology of Nile tilapia (*Oreochromis niloticus*). *Fish and Shellfish Immunology*, *38*(1), 149-157.

Zainin, N. S., Lau, K. Y., Zakaria, M., Son, R., Abdull, R., & Rukayadi, Y. (2013). Antibacterial activity of *Boesenbergia rotunda* (L.) Mansf. A. extract against *Escherichia coli*. *International Food Research Journal*, *20*(6), 3319-3323.

Zakaria, Z. A., Mohamad, A. S., Chear, C. T., Wong, Y. Y., Israf, D. A., & Sulaiman, M. R. (2010). Antiinflammatory and antinociceptive activities of *Zingiber zerumbet* methanol extract in experimental model systems. *Medical Principles and Practice*, *19*(4), 287-294.

Zamri-Saad, M., Amal, M. N. A., & Siti-Zahrah, A. (2010). Pathological changes in red tilapias (*Oreochromis* spp.) naturally infected by *Streptococcus agalactiae*. *Journal of Comparative Pathology*, *143*(2), 227-229.

Zamri-Saad, M., Amal, M. N. A., Siti-Zahrah, A., & Zulkafli, A. R. (2014). Control and prevention of streptococcosis in cultured tilapia in Malaysia: a review. *Pertanika Journal of Tropical Agricultural Science*, *37*(4), 389-410.

Zana Hama, G. K. (2015). *Effects of selected herbal extracts on sexual maturity, growth performance and immune reaponse of Nile tilapia* (*Oreochromis niloticus Linnaeus*) (Master's thesis). Universiti Putra Malaysia.

Zar, J. H. (1984). *Biostatistical analysis* (2nd ed.). New Jersey, USA: Prentice Hall Inc.

Zeng, H. W., Jiang, Y. Y., Cai, D. G., Bian, J., Long, K., & Chen, Z. L. (1997). Piperbetol, methylpiperbetol, piperol a and piperol b: a new series of highly specific PAF receptor agonists from *Piper betle*. *Planta medica*, *63*(04), 296-298.

Zeng, H., Ren, Z. L., & Guo, Q. (1996). Application of allicin in tilapia feed. *China Feed*, *21*, 29-30.