

Chapter 9

Sustainable Tilapia Farming, the Role of Culture Systems



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Abstract Considering environmental sustainability and vulnerability to the effects of climate change on fish production, sustainable and adaptable aquaculture systems are necessary”. Biofloc technology (BFT) and recirculating aquaculture systems (RAS) are eco-friendly, water efficient, highly productive intensive farming systems, which are not associated with adverse environmental impacts, such as habitat destruction, water pollution and eutrophication, biotic depletion, ecological effects on biodiversity due to captive fish and exotic species escape, disease outbreaks, and parasite transmission. Moreover, BFT and RAS operate in an indoor controlled environment, and thus, are only minimally affected by climatic factors, including rainfall variation, flood, drought, global warming, cyclone, salinity fluctuation, ocean acidification, and sea level rise. This chapter provides into insight the application of these techniques for sustainable tilapia production, which focuses on their effects on growth performance, immune response, and disease resistance.

Keywords Tilapia · Sustainable · Biofloc technology · RAS

9.1 Introduction

Aquaculture, is one of the fastest food-producing sectors, with an average annual growth rate of 5.3% during the period 2001–2018, and production has increased by over 600% since 1990 (FAO 2020b). Global aquaculture production achieved 82.1 million tons in 2018, of which inland aquaculture produced 51.3 million tons (62%), while coastal and marine aquaculture¹ reached 30.8 million tons (38%) (FAO 2020b). Aquaculture is performed in various environments and regions, employing various technologies, and cultured systems, and raising many species (Ahmad et al. 2022). Asia accounts for around 90% of global aquaculture production (FAO 2020a, b), and aquaculture, with its expansion outpacing global population growth,

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is playing an important role in boosting food production for contribution to food security and human nutrition (Pradeepkiran 2019). Although the expansion of aquaculture, the challenge of feeding a growing global population, which is expected to reach 9.7 billion by 2050 (Bahar et al. 2020), is a vivid reality, which is central in global political and scientific debates (Berners-Lee et al. 2018). Because of population growth and stagnation of capture fisheries, global aquaculture production is challenged to achieve remarkable targets, estimated to possibly reach 109 million tons in 2030 (FAO 2020a, b), and 140 million tons in 2050 (Waite et al. 2014).

To achieve the required further increase in global seafood production, aquaculture is envisaged as the only available solution, but it could bring additional adverse environmental effects if its expansion is not based on sustainable farming systems (Ahmed and Turchini 2021). Accordingly, the rapid growth of aquaculture has been linked to raising concerns about its environmental sustainability (Ahmad et al. 2022; Tom et al. 2021). Broadly, aquaculture has already been increasingly associated with a great variety of negative environmental impacts, including habitat destruction, water pollution and eutrophication, biotic depletion, disease and parasite transmission, and greenhouse gas (GHG) emissions (Ahmed et al. 2019; (Carballeira Braña et al. 2021; Adegbeye et al. 2019; Kosten et al. 2020). For example, some intensive aquaculture practices have been reported to cause antibiotic pollution, eutrophication, land occupation, and other environmental hazards (Dauda et al. 2019). The invasion of exotic fish species in some aquaculture systems has been reported to have potentially negative effects on biodiversity and the ecosystem; (Banha et al. 2022; Kiruba-Sankar et al. 2018). Freshwater aquaculture, particularly tilapia, has been reported to cause adverse effects on water resources with a broad range of impacts on the biodiversity, ecosystems, and societies (Bashir et al. 2020; Kaleem and Bio Singou Sabi 2021; Moyo and Rapatsa 2021). Tilapia aquaculture has also been censured for a variety of environmental issues, including water pollution and pathogen transmission to wild fish (Shaaban et al. 2021). The environment, on the other hand, has a series of impacts and imposes certain limitations on aquaculture, with climate change posing a significant threat to increasing global fish production (Baag and Mandal 2022; Maulu et al. 2021). A wide range of climatic factors, such as rainfall, flood, drought, cyclone, global warming, sea surface temperature change, salinity fluctuation, ocean acidification, and sea level rise have a significant impact on aquaculture practices (Ahmed et al. 2019). In essence, future climate change will certainly have adverse impacts on sustainable increasing aquaculture production (Ahmed et al. 2019; Boyd et al. 2020). It is therefore necessary to develop and implement adaptation strategies to cope with these challenges.

Considering the environmental concerns and impacts, as well as the vulnerability to the effects of climate change and other environmental variables of fish production in aquaculture, one of the potential and increasingly proposed adaptation strategies is the implementation of sustainable aquaculture systems, such as BFT and RAS. These systems allow for rising fish in a land-based, indoor, and controlled environment to minimize the direct interactions between the production processes and the environment (Ahmed and Turchini 2021; Khanjani et al. 2022). They offer benefits

in improving aquaculture production that could contribute to the achievement of sustainable development goals (Bossier and Ekasari 2017; Nguyen et al. 2021). This technology could result in higher productivity with less impact on the environment. Furthermore, biofloc systems may be developed and performed in integration with other food production, thus promoting productive integrated systems, aiming at producing more food and feed from the same area of land with less input (Nisar et al. 2022). The chapter aims to address different aspects of BFT and RAS as alternatives for sustainable tilapia production.

9.2 Biofloc Technology

9.2.1 History of Biofloc Technology (BFT)

The biofloc system (BFT) evolved as an alternative to the conventional aquaculture system that is used in shrimp and tilapia productions (Ulloa Walker et al. 2020). It originated in the 1970s at the French Research Institute for Exploitation of the Sea (IFREMER) with Gerard Cuzon as the pioneer (Emerenciano et al. 2012; Devi and Kurup 2015). The BFT was then widely applied in commercial shrimp farming (Samocha et al. 2019). In the 1990s, different studies at pilot and commercial scales were conducted in the USA with penaeid shrimp led by J. Stephen Hopkins and with finfish at the Technion-Israel Institute of Technology led by Yoram Avnimelech (Emerenciano et al. 2013). In the mid-2000s, several studies on penaeid shrimp were conducted at the Federal University of Rio Grande-FURG (Brazil) led by Wilson Wasielesky and the Texas A&M University (Corpus Christi Campus, USA) led by Tzachi Samocha. After that, there was a significant increase in the number of scientific publications about biofloc technology worldwide. The number has increased from less than 10 in 2009 to more than 100 publications in 2018, with studies conducted mainly in Brazil, China, the United States of America, Mexico, and India (Ulloa Walker et al. 2020), helping to strengthen the technology and boost the industry. Another important factor for such progress was the wide range of courses and lectures offered in both scientific and commercial events for the scientific community, academia, and aquaculturists. However, despite the progress and benefits of BFT as reported by the scientific community and academia, there is still room for its commercial expansion. For example, in Indonesia, it is estimated that only 20–25% of shrimp production has occurred using biofloc technology (Thong 2014). Among the reasons behind such a scenario are the higher implementation and production costs compared to traditional land-based systems, and the complexities in the management and implementation of the technology, which requires greater technical knowledge and permanent monitoring of water quality (Avnimelech 2015).

9.2.2 Principles of Biofloc Technology

The BFT operates on the principle of nutrient recycling by maintaining a higher carbon: nitrogen (C: N) ratio above 15 to stimulate the mass growth of heterotrophic bacteria (Guo et al. 2020). Higher C: N ratio is maintained when more carbon sources, such as molasses, cassava, hay, sugarcane, starch, wheat bran, cellulose, etc., are sprayed on the surface of pond water with continuous aeration (Ogello et al. 2021). Under favorable BFT conditions, up to 0.5 g of heterotrophic bacterial biomass g^{-1} substrate of carbon can be produced (Eding et al. 2006). With the information that 1 g of carbon produces 0.5 g of bacteria, farmers can estimate quantities of floc in the culture systems (Nisar et al. 2022). The biofloc process stimulates the natural growth of macro-aggregates of organisms that enhance self-nitrification in the culture water (Jamal et al. 2020).

In outdoor BFT systems, the photosynthetic pathway that produces algae normally precedes the bio-floccing process (Ogello et al. 2021). The algae provide a substrate to which the bioflocs attach and are usually referred to as green bioflocs (Ahmad et al. 2017; Khanjani and Sharifinia 2020). Under indoor conditions, bioflocs are mainly bacteria and are referred to as brown bioflocs (El-Sayed 2021; Emerenciano et al. 2021). With the addition of an adequate carbon source bacterial floc stimulates a second production line that involves the degradation of organic wastes by bacteria to produce billions of bacterial cells under optimum aeration conditions (Khanjani et al. 2022). During this process, autotrophic and heterotrophic bacteria proliferate and attract billions of other cells including diatoms, fungi, algae, protozoans, and various types of plankton (Avnimelech 2007; Bossier and Ekasari 2017). The traditional aquaculture ponds lack injection of carbon source, and aeration mechanisms and thus harbor fewer and less diverse bacterial communities, as opposed to BFT (Felix and Menaga 2021). Small quantities of bacteria cannot form substantial flocs in the culture system. The sediment of traditional ponds accumulates higher quantities (49%) of nitrogenous waste while the BFT pond sediments have less (5%) nitrogenous waste (Ogello et al. 2021).

9.2.3 Biofloc in Tilapia

Tilapia has played an important role in global aquaculture in recent decades (FAO 2020a). It ranks the second most cultured fish species worldwide due to its fast growth, resistance to various environmental conditions, and capability of being produced in dense and ultra-dense forms (Menaga et al. 2019; Avnimelech 2007; Avnimelech 2015; Khanjani et al. 2021). Moreover, tilapia is an omnivore species that filter food particles and easily feeds on a rich natural food source and biofloc-dependent microorganisms (Durigon et al. 2020; Prabu et al. 2019). Considering the above considerations, tilapia is a suitable candidate for the biofloc system (Khanjani et al. 2022). The biofloc technique has been accepted in many countries over the past

decade (Vyas 2020). Currently, the biofloc technique has been successfully developed in large-scale farms in Asia (China, South Korea), Latin America, and Central America, as well as on a small scale in the United States, Italy, and other countries. In addition, many research centers and academic institutions are expanding BFT, mainly in key areas such as growth, nutrition, reproduction, microbial ecology, biotechnology, and economics (Khanjani and Sharifinia 2020).

9.2.3.1 Biofloc as Growth Promoters

In situ utilization of microbial flocs generated in biofloc systems by some aquaculture organisms as well as the utilization of processed biofloc as a feed ingredient has been well documented (Kuhn et al. 2009; Kuhn et al. 2010; Anand et al. 2014). It has been demonstrated that the concentrations of free amino acids such as alanine, glutamate, arginine, and glycine, which are known attractants in the shrimp diet, are present in biofloc (Vyas 2020; Ahmad et al. 2017). Levels in biofloc were found to be comparable to that of the shrimp commercial diet suggesting that biofloc are likely to be recognized as food particles by some aquaculture organisms. Furthermore, biofloc technology application in larviculture may provide an easily accessible food source for the larvae outside the regular feeding moments, thus minimizing possible negative social interaction during feeding (Ekasari et al. 2015b).

Studies by various researchers have reported that the biofloc system improves the growth performance of the Nile tilapia (Azim and Little 2008; Luo et al. 2013; Mirzakhani et al. 2019; Khanjani et al. 2021; Table 9.1). The improved growth performance was attributed to optimum water quality and continuous production of biofloc. Biofloc contains poly beta-hydroxybutyrate (De Schryver and Verstraete 2009) and bioactive compounds, such as carotenoids, chlorophylls, and phytosteroids that promote the growth of cultured aquatic organisms. The adaptability of tilapia to biofloc consumption and digestion of microbial protein has been demonstrated in several studies (Azim and Little 2008). Avnimelech (2007) reported that the production of biofloc in fish ponds can meet 50% of tilapia's protein requirements. Khanjani et al. (2021) found that tilapia feed better on the biofloc impacted by molasses daily, resulting in the highest biomass increase and the lowest feed conversion ratio. Based on their results, the highest feed conversion ratio and the lowest protein efficiency were observed in the control treatment without biofloc. Researchers have found that a combination of biofloc and artificial foods improves feed conversion ratio and feed efficiency (Khanjani et al. 2021; Mirzakhani et al. 2019).

Biofloc includes bacterial proteins, polyhydroxy butyrate, and bacteria containing peptidoglycan or lipopolysaccharides, which can promote growth performance (Khanjani and Sharifinia 2020). In addition, biofloc have probiotic properties that help fish in digestion and absorption of artificial diets. In the study using orange peel-derived pectin (OPDP) added to the biofloc system, Doan et al. (2018) indicated that the inclusion of 10 g kg⁻¹ OPDP significantly improved the growth performance and feed utilization of Nile tilapia. Similarly, a significant increase in growth

Table 9.1 Weight gain (WG), Specific growth rate (SGR), Food conversion efficiency (FCE), Food conversion ratio (FCR), Protein efficiency ratio (PER), Survival rate (SR), and digestive enzyme of Nile tilapia in biofloc and RAS systems

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Orange peels derived pectin (OPDP)	Nile tilapia (<i>Oreochromis niloticus</i>) (9.09 ± 0.05 g)	0 (control in clear water), 0 (control in biofloc system), 5, 10, and 20 g kg ⁻¹ OPDP for 8 weeks	SGR, WG, and FW ↑ FCR ↓	Doan et al. (2018)
BFT	Stocking density	Tilapias (0.51 ± 0.05 g)	166 orgs m ⁻³ (LD, low density), 333 orgs m ⁻³ (MD, middle density) and 600 orgs m ⁻³ (HD, high density) 120 days	Growth performance ↓ FCR ↑	Liu et al. (2018)
BFT	Carbon sources	Nile tilapia (<i>O. niloticus</i>) 2.7 ± 0.4 g	100% of MO, 75% of MO+ 25% of WF, 50% of MO+ 50% of WF, 25% of MO+ 75% of WF, and 100% of WF	Growth performance ↑	Mirzakhani et al. (2019)
BFT	Pizzeria by-product	Nile tilapia (<i>O. niloticus</i>) (2.90 ± 0.02 g)	0, 20, 40, 60, 80 and 100% 38 days	Growth performance ↑ Economic benefits ↑	de Sousa et al. (2019)
BFT	Dietary digestible protein and digestible energy	Nile tilapia (<i>O. niloticus</i>) (1.25 ± 0.15 g)	Digestible protein (22, 26 and 30% DP) and digestible energy (3000, 3150 and 3300 kcal kg ⁻¹) 42 days	Pepsine activity ↑ Trypsine activity ↑	Durigon et al. (2019)
BFT	Carbon sources	Fingerlings	Corn flour (CF), wheat flour (WF), sugar (SU) and a control (C)	FCR and water ↓ Survival rate ↑	García-Ríos et al. (2019)
BFT	OPDP and <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (1.25 ± 0.15 g) 5.92 ± 0.08 g	Diet 1 (0 g kg ⁻¹ OPDP and 0 CFU g ⁻¹ <i>L. plantarum</i>), diet 2 (10 g kg ⁻¹ OPDP), diet 3 (108 CFU g ⁻¹ <i>L. plantarum</i>), and diet 4 (10 g kg ⁻¹ OPDP +108 CFU g ⁻¹ <i>L. plantarum</i>)	Growth performance ↑ FCR ↓	Van Doan et al. (2019)

BFT	<i>in-situ</i> and <i>ex-situ</i> biofloc	Nile tilapia (<i>O. niloticus</i>) (5.1 ± 0.05 g/fish)	T1-biofloc developed within the culture systems (<i>in-situ</i>), T2-biofloc supplementation in fish feed (<i>ex-situ</i>) and C- Control without biofloc	Growth performance ↑	Menaga et al. (2019)
BFT	Biochar	Nile tilapia (<i>O. niloticus</i>) (36.2 g/fish)	B; only biochar, GB; biochar + glucose, while for the control (G) only glucose	Growth performance →	Abakari et al. (2020)
BFT	Jaggery-based BFT	Nile tilapia (<i>O. niloticus</i>) (32.2 ± 10.1 g/fish)	22.5%, 27.7%, and 32.3% digestible protein (DP) and 6% lipid	Growth performance ↑	Elayaraja et al. (2020)
BFT	Chestnut polyphenols (CSP)	Nile tilapia (<i>O. niloticus</i>) (12.77 ± 0.17 g)	0, 1, 2, 4, and 8 g kg ⁻¹ of CSP	Growth performance ↑	H. Van Doan et al. (2020a)
BFT	Watermelon rind powder (WMRP)	Nile tilapia (<i>Oreochromis niloticus</i>) (17.14 ± 0.12 g)	0 (Diet 1- control), 20 g kg ⁻¹ WMRP (Diet 2), 40 g kg ⁻¹ WMRP (Diet 3), 80 g kg ⁻¹ WMRP (Diet 4), and 160 g kg ⁻¹ WMRP (Diet 5)	Growth performance ↑ FCR ↓	Hien Van Doan et al. (2020b)
BFT	Phosphatidylcholine	Nile tilapia (<i>O. niloticus</i>) (8.03 ± 0.03 g)	0, 400, 800 and 1200 mg/kg of feed 40 days	Growth performance → Energy metabolism ↑	Sousa et al. (2020)
BFT	<i>Tenebrio molitor</i> meal (TM)	Nile tilapia (<i>O. niloticus</i>) (2.08 ± 0.19 g)	0% or control, 5, 10, 15 and 20% of TM	Productivity and survival rate ↑	Tubin et al. (2020)
BFT	Density and dietary carbon sources	Nile tilapia (<i>O. niloticus</i>) (50.47 ± 0.05 g)	Stocking densities [20, 40 and 60 fish per m ³] and fed the basal diet without carbon sources or with broken rice flour (BRF) or broken wheat grain flour (BWGF) 84 days	Growth performance ↑ Feed utilization ↑	Zaki et al. (2020)

(continued)

Table 9.1 (continued)

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Pineapple peel powder (PAPP)	Nile tilapia (<i>O. niloticus</i>) (20.91 ± 0.11 g)	0, 10, 20, 30 and 40 g kg ⁻¹ PAPP 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2021a)
BFT	Watermelon rind powder (WMRP) + <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (16.57 ± 0.14 g)	Diet 1 (0 g kg ⁻¹ WMRP and 0 CFU g ⁻¹ L. <i>plantarum</i>) (control), Diet 2 (40 g kg ⁻¹ WMRP), Diet 3 (10 ⁸ CFU g ⁻¹ LP), and Diet 4 (40 g kg ⁻¹ WMRP + 10 ⁸ CFU g ⁻¹ LP) 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2021b)
BFT	Pineapple peel powder (PAPP) + <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (20.91 ± 0.11 g)	Diet 1 (0 g kg ⁻¹ PAPP and 0 CFU g ⁻¹ L. <i>plantarum</i>) (control), Diet 2 (10 g kg ⁻¹ PAPP), Diet 3 (10 ⁸ CFU g ⁻¹ LP), and Diet 4 (10 g kg ⁻¹ PAPP + 10 ⁸ CFU g ⁻¹ LP) 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2021d)
BFT	Amla (<i>Phyllanthus emblica</i>) fruit extract	Nile tilapia (<i>O. niloticus</i>) (10.48 ± 0.56 g)	0, 5, 10, 20, and 40 mg kg ⁻¹ AFE 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2022b)
BFT	Light levels	Nile tilapia (<i>O. niloticus</i>) (1.73 ± 0.16 g)	24 h of light (24hL), 12 h of light/12 h of darkness (12hL/12hD) and 24 h of darkness (24hD)	Carcass quality ↑ Digestive and hepatic enzymes ↑	Kharjani and Sharifinia (2021)
BFT	Symbiotics	Nile tilapia (<i>O. niloticus</i>) (30–35 g)	BFT with and without symbiotics 40 days	Growth performance ↑	Laice et al. (2021)
BFT	Stocking densities	Nile tilapia (<i>O. niloticus</i>) (133.91 g)	18.75, 37.50, 56.25, and 75.00 fish·m ⁻³ 260 days	Growth performance ↑	Manduca et al. (2021)

	Beneficial bacteria	Nile tilapia (<i>O. niloticus</i>) (8.63 ± 3.35g)	T1, SR control; T2, SR + SSP; T3, SR + MSP; T4, BF + MSP; T5, BF + SSP; T6, BF control 112 days	Growth performance ↑	Mohammadi et al. (2021)
BFT	Coffee silverskin (CSS)	Nile tilapia (<i>O. niloticus</i>) (15.54 ± 0.21 g)	CSS1 (Control), CSS2 (10 g kg ⁻¹), CSS3 (20 g kg ⁻¹), CSS4 (40 g kg ⁻¹), and CSS5 (80 g kg ⁻¹) 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2021c)
BFT	Carbon sources and stocking densities	Nile tilapia (<i>O. niloticus</i>) (5.15 ± 1.12 g)	Low stocking density (LSD), 140 fish /m ³ and high stocking density, (HSD), 280/m ³ 98 days	Growth performance ↑	El-Hawarry et al. (2021)
BFT	Salinities	Nile tilapia (<i>O. niloticus</i>) (13.78 ± 0.62 g)	T1 (5 ppt), T2 (10 ppt), T3 (15 ppt), T4 (20 ppt) and control (0 ppt) 90 days	Growth performance ↑ Survival rate ↑ Carcass quality ↑	Kumari et al. (2021)
BFT	Dietary phytase	Nile tilapia (<i>O. niloticus</i>) (29.8 g)	6-phytase (3000 FTU/kg; Quantum Blue™, P-F + Phy), and positive (C) and negative (P-F) control 154–156 days	Growth performance →	Green et al. (2021)
BFT	Spent coffee grounds (SCG)	Nile tilapia (<i>O. niloticus</i>) (15.25 ± 0.07 g)	SCG1 (control), SCG2 (10 g kg ⁻¹), SCG3 (20 g kg ⁻¹), SCG4 (40 g kg ⁻¹), and SCG5 (80 g kg ⁻¹) 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2022a)
BFT	Host-associated probiotic <i>Bacillus altitudinis</i> B61-34b	Nile tilapia (<i>O. niloticus</i>) (25.50 ± 0.52 g)	0 (BAA1—Control), 10 ⁶ (BAA2), 10 ⁷ (BAA3), 10 ⁸ (BAA4) and 10 ⁹ (BAA5) CFU ml ⁻¹ 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2021e)
BFT	Host-associated probiotic <i>Lactobacillus paracasei</i> l61-27b	Nile tilapia (<i>O. niloticus</i>) (25.40 ± 0.52 g)	LP1 = 0 (Control), LP2 = 10 ⁶ CFU mL ⁻¹ , LP3 = 10 ⁷ CFU mL ⁻¹ , LP4 = 10 ⁸ CFU mL ⁻¹ , and LP5 = 10 ⁹ CFU mL ⁻¹) 8 weeks	Growth performance ↑ FCR ↓	Van Doan et al. (2021c)
BFT	Grade feeding rates	Nile tilapia (<i>O. niloticus</i>) (3.1 ± 0.1 g)	0%, 2.5%, 5.0%, 7.5%, and 10% of body weight per day 70 days	Growth performance ↑	Oliveira et al. (2021)

(continued)

Table 9.1 (continued)

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	17- α -methyltestosterone	Nile tilapia (<i>O. niloticus</i>) (Fry)	60, 90, 120, 150 and 180 mg kg ⁻¹	Masculinization rates \uparrow	Costa e Silva et al. (2022)
BFT	Feeding levels and stocking densities	Nile tilapia (<i>O. niloticus</i>) (3.2 \pm 0.05 g)	Feeding levels (0, 15, 30, 45 and 100) and 2 stocking densities (500 fish/m ³ and 1000fish/m ³)	Growth performance \uparrow	Sarsangi Aliabadi et al. (2022)
BFT	Longan seed powder (LS)	Nile tilapia (<i>O. niloticus</i>) (13.82 \pm 0.06 g)	Control (LS0), 10 (LS10), 20 (LS20), 40 (LS40), and 80 (LS80) g kg ⁻¹ LS 8 weeks	Growth performance \uparrow FCR \downarrow	Wannavijit et al. (2022)
BFT	Rambutan seed (RS)	Nile tilapia (<i>O. niloticus</i>) (14.77 \pm 0.80 g)	0, 5, 10, 20, and 40 g kg ⁻¹ of RS 8 weeks	Growth performance \uparrow FCR \downarrow	Xuan et al. (2022)
BFT	Rambutan peel (RP)	Nile tilapia (<i>O. niloticus</i>) (17.14 \pm 0.12 g)	0 g kg ⁻¹ (control – RP0); 10 g kg ⁻¹ (RP10); 20 g kg ⁻¹ (RP20); 40 g kg ⁻¹ (RP40), and 80 g kg ⁻¹ (RP80) 8 weeks	Growth performance \uparrow FCR \downarrow	Le Xuan et al. (2022)
BFT	Chitosan	Nile tilapia (<i>O. niloticus</i>) (1.70 \pm 0.36 g)	30, 60 and 90 ppm and 10, 20 and 30 ppm of chitosan	Growth performance \uparrow Digestive enzymes \uparrow FCR \downarrow	Chutia et al. (2022)
RAS	Light intensity and photoperiod	Nile tilapia (<i>O. niloticus</i>) (5 \pm 0.9 g)	(1000, 2000, and 3000 lx) and photoperiods (12L:12D, 18L:6D, 24L:0D) 160 days	Growth performance \uparrow FCR \downarrow Stress \rightarrow	Wang et al. (2020)

RAS	Magnetic field	Nile tilapia (<i>O. niloticus</i>) (7.16 ± 0.05 g)	0.00, 0.10, 0.15 and 0.20T 70 days	Growth performance ↑ FCR ↓	Hassan et al. (2018)
RAS	Rearing systems and dietary probiotic	Nile tilapia (<i>O. niloticus</i>) (embryos)	FTS + control diet, RAS + <i>B. subtilis</i> coated diet (RASB) 33 days	Survival rate ↑ Beneficial bacteria in gut ↑	Deng et al. (2022)
BFT-RAS	Culture systems	Nile tilapia (<i>O. niloticus</i>) (0.17 ± 0.00 g)	Clear-water (CW), biofloc (BF), or hybrid (HY) 9 weeks	Growth performance ↑ FCR ↓	Fleckenstein et al. (2018)
RAS + BFT	Nitrogen and phosphorus budgets	Nile tilapia (<i>O. niloticus</i>) (3.54 ± 2.82 g)	(BFT) aquaculture system and a recirculation aquaculture system (RAS) during over-wintering of tilapia 64 days	Recovery rate of N in BFTs ↑ P recovery rate →	Cao et al. (2020)
BFT-RAS	Protein levels	Nile tilapia (<i>O. niloticus</i>) (39.1 ± 2.5 g)	23, 27, 31 or 35% crude protein 9 weeks	Growth performance ↑ FCR ↓	Nguyen et al. (2021)

performance and feed utilization were observed in Nile tilapia fed dietary inclusion of OPDP and *Lactobacillus plantarum*, chestnut polyphenols (CSP), watermelon rind powder (WMRP), pineapple peel powder (PAPP), watermelon rind powder (WMRP) + *L. plantarum*, pineapple peel powder (PAPP) + *L. plantarum*, amla (*Phyllanthus emblica*) fruit extract, coffee silverskin (CSS), spent coffee grounds (SCG) (Van Doan et al. 2019; 2020a, b; 2021a, b, c, d; 2022a, b). Significant improvement in growth performance and FCR may be attributable to the bioactive compounds of these supplementations, which act not only as a nutrient source for fish but also as carbon sources for microbial protein production in biofloc systems. In studies using host-associated probiotics (*Bacillus altitudinis* B61-34b and *Lactobacillus paracasei* 161-27b), the authors indicated that supplementation of host-associated probiotics in indoor biofloc system resulted in better growth performance and feed utilization compared to the control group. This may be due to the complementary roles of biofloc and *B. altitudinis*. Numerous investigations have demonstrated that biofloc offers an essential nutrient source for tilapia (Ekasari et al. 2014a; Green et al. 2019). The addition of *Bacillus* spp. in cultured water or diets lowers ammonium levels in fish culture systems (Dash et al. 2018; Elsabagh et al. 2018). Furthermore, the presence of favorable microbial flocs and external probiotics will likely boost the number of valuable microbiota in the tilapia's digestive system (Rohani et al. 2022; de Sousa et al. 2019). Increased secretion of digestive enzymes through the colonization of bacteria facilitates the absorption of nutrients by the intestinal epithelial cells (Liu et al. 2017). *Bacillus* produces many biological substances, including cellulase, phytase, tannase, chitinase, xylanase, protease, amylase, and lipase (Ringø 2020). Favorable bacteria also release several nutrients, in particular vitamins, amino acids, and fatty acids, and diminish lethal feedstuffs and infectious bacteria (Zaineldin et al. 2021). Recently, the dietary inclusion of rambutan and long seed powder or rambutan peel in the Nile tilapia diet led to an increase in growth rate and feed utilization (Xuan et al. 2022; Wannavijit et al. 2022). It has been reported that these seeds are known as carbon sources (Yang et al. 2015; Lawtae and Tangsathikulchai 2021) and has hence been used in biofloc system (Liu et al. 2019). It has been observed that adding carbon to a biofloc system causes heterotrophic bacteria to utilize the inorganic nitrogen by changing the water C: N ratio, resulting in a higher microbial protein source for the host and improved water quality (Guo et al. 2020). In addition, incorporating a carbon source leads to the formation of biofloc, a new protein source for fish (Krummenauer et al. 2020; Tinh et al. 2021). Additionally, these products also act as potential prebiotics or carbohydrate (Estrada-Gil et al. 2022; Jahurul et al. 2020). Similarly, supplementation of pizzeria by-product de Sousa et al. (2019); dietary digestible protein and digestible energy (Durigon et al. 2019); phosphatidylcholine (Sousa et al. 2020); *Tenebrio molitor* meal (TM) (Tubin et al. 2020); symbiotics (Laice et al. 2021); beneficial bacteria (Mohammadi et al. 2021); dietary phytase (Green et al. 2021), and chitosan (Chutia et al. 2022) led to higher growth rate and feed utilization.

Another common study aspect using biofloc technology is the application of different carbon sources and their effects on tilapia growth and feed conversion ratio. (Mirzakhani et al. (2019) reported that fish in a biofloc system with 100% of wheat

flour at a C:N ratio of 15:1 showed the highest growth performance with improved intestine histoarchitecture. Wheat flour as a major source of starch and energy can also provide substantial amounts of other nutrients such as protein, vitamins, and phytochemicals and especially high fiber content (ca. 12%) compared to the lower fiber content in molasses (ca. 0.5%) (Shewry and Hey 2015). These nutrients might enhance the biochemical composition and bioactive compounds of biofloc. In addition, dispersed particles of wheat flour in water may provide a good substrate for the development and growth of microorganisms and bacteria because of which the nutrition value of the produced biofloc increases, ultimately influencing the fish growth and immune response (Mirzakhani et al. (2019). García-Ríos et al. (2019) indicated that the fingerlings obtained in BFT, with corn and sugar as C sources, had a similar growth rate to the control. However, the BFT promotes significant savings in feed (41.1 to 58.9%) and water (67.4 to 75.5%) compared to the traditional method. Similar results were in the study of Zaki et al. (2020), where the authors indicated that Increased growth and feed utilization were recorded in 40 fish per m³ fed with broken rice flour. El-Hawarry et al. (2021) also found that the growth rate was improved in the groups of fish under low stocking density with molasses and glycerol as carbon sources. Carbon source affects the cultured species' growth depending on the formatted biofloc characteristics, such as its "volume, chemical composition, and ability to store bioactive compounds (Wang et al. 2015; Zhao et al. 2016). Additionally, microbial flocs which are formed from different carbon sources act as a supplemental food source that constantly provides additional essential amino acids profile (microbial protein), polyunsaturated fatty acids, minerals, vitamins, and an external source of digestive enzymes (Avnimelech 2007; Azim and Little 2008; Bakhshi et al. 2018; De Schryver and Verstraete 2009). In contrast, the use of biochar as an alternative carbon source for biofloc technology did not affect the growth rate of Nile tilapia (Abakari et al. 2020). Although biochar is regarded as a recalcitrant carbon source, the utilization of biochar-derived carbon by heterotrophic bacteria has been described (Farrell et al. 2013).

Effects of stocking density on Nile tilapia growth raised in biofloc systems have been conducted by several researchers. Liu et al. (2018) showed that low stocking density (166 fish/m⁻³) improved growth performance and FCR of Nile tilapia raised in the biofloc system. Similarly, Manduca et al. (2021) reported that tilapia stocking density in BFT around 33 fish m⁻³ had higher profitability since it produces a large proportion of harvested fish that reach high body weights, and possibly high selling prices, combined with desirable biomass. Recently, Sarsangi Aliabad et al. (2022) also suggested that the stocking density of 1000/m³ for larviculture of tilapia in BFT uses water and equipment more efficiently. Biofloc acts as the natural food that contributes significantly to the nutrition of tilapia fingerlings, allowing the reduction of the feeding rations. Biofloc consumption corresponds to 50% of the daily food of tilapia (Avnimelech 2007). Another study revealed that 25% of the protein requirement of tilapia could be provided by floc consumption (Avnimelech 2015). Besides, significantly improved growth performance, FCR, and digestive enzymes were observed in Nile tilapia raised in biofloc combined with different conditions, such as in-situ and ex-situ biofloc (Menaga et al. 2019), jaggery-based BFT (Elayaraja

et al. 2020); light levels (Khanjani and Sharifinia 2021); salinities (Kumari et al. 2021); grade feeding rates (Oliveira et al. 2021), and 17- α -methyltestosterone (Costa e Silva et al. 2022).

9.2.3.2 Biofloc as Immunostimulants

Bioflocs also offer a lot of MAMPs (microbial-associated molecular patterns), which may be recognized as immunostimulants, resulting in higher resistance to diseases (Ekasari et al. 2014b, 2015a). Additionally, it consists of a wide range of organic compounds, such as carotenoids, chlorophylls, bromophenols, phytosterols, and antibacterials that have a positive effect on immune factors of cultivated aquatic species (Crab et al. 2010; Najdegerami et al. 2016; Bakhshi et al. 2018; Mirzakhani et al. 2019).

The effects of biofloc in combination with different functional feed additives on the immune response of Nile tilapia have been reported in previous studies (Doan et al. 2018; Van Doan et al. 2019, 2020a, b; 2021a, b, c, d; 2022a, b; Xuan et al. 2022; Le Xuan et al. 2022; Wannavijit et al. 2022; Table 9.2). Similar findings were observed in Nile tilapia fed in-situ and ex-situ biofloc (Menaga et al. 2019); phosphatidylcholine (Sousa et al. 2020); symbiotics (Laice et al. 2021); beneficial bacteria (Mohammadi et al. 2021), and probiotics (Bañuelos-Vargas et al. 2021). These substances act as immunostimulants and/or carbon sources for the proliferation of microbial proteins in the biofloc system.

Carbon source applications in the biofloc system could result in better immune response in Nile tilapia. Mirzakhani et al. (2019) indicated that fish reared in a biofloc system based on 100% wheat flour and a C/N ratio of 15 demonstrated the humoral immune response. Similarly, a significant increase in innate and specific immune responses was observed in Nile tilapia raised in biofloc with biochar (Abakari et al. 2020) and jaggery-based BFT (Elayaraja et al. 2020) as carbon sources. Carbon sources play a vital role in the proliferation of microbial protein in biofloc systems, which in turn act as immunostimulants for culture species (Panigrahi et al. 2019). Carbon sources and stocking densities also have a great impact on Nile tilapia's immune response raised under the biofloc system. El-Hawarry et al. (2021) indicated that the growth rate and growth-related genes were improved in the groups of fish under low stocking density (LSD) with molasses and glycerol as carbon sources. Recently, (Sarsangi Aliabad et al. 2022) also reported that the BFT system improved water quality, growth performance, and immune function of Nile tilapia fry.

9.2.3.3 Biofloc as Disease Prevention Techniques

In biofloc systems, aquaculture animals may also benefit from reduced pathogen pressure (Bossier and Ekasari 2017). Some studies demonstrated that the presence of potentially pathogenic bacteria might be reduced in biofloc systems (Gustilatov et al. 2022; de Lima Vieira et al. 2021; Khanjani et al. 2022; Table 9.3). Increase disease

Table 9.2 Immune responses of tilapia cultured biofloc system

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Stocking density	<i>Tilapia</i> (0.51 ± 0.05 g)	166 orgs m ⁻³ (LD, low density), 333 orgs m ⁻³ (MD, middle density) and 600 orgs m ⁻³ (HD, high density) 120 days	Lysozyme activity ↓ Complement 3 activity ↓ Glutathione level ↓	Liu et al. (2018)
BFT	Orange peels derived pectin	Nile tilapia (9.09 ± 0.05 g)	0 (control in clear water), 0 (control in biofloc system), 5, 10, and 20 g kg ⁻¹ OPDP for 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Doan et al. (2018)
BFT	<i>in-situ</i> and <i>ex-situ</i> biofloc	Nile tilapia (<i>O. niloticus</i>) (5.1 ± 0.05 g/fish)	T1-biofloc developed within the culture systems (<i>in-situ</i>), T2-biofloc supplementation in fish feed (<i>ex-situ</i>) and C- Control without biofloc	Immune gene expressions ↑	Menaga et al. (2019)
BFT	Carbon sources	Nile tilapia (<i>O. niloticus</i>) (2.7 ± 0.4 g)	100% of MO, 75% of MO+ 25% of WF, 50% of MO+ 50% of WF, 25% of MO+ 75% of WF, and 100% of WF	Humoral immune responses ↑	Mirzakhani et al. (2019)
BFT	OPDP and <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (1.25 ± 0.15 g) 5.92 ± 0.08 g	Diet 1 (0 g kg ⁻¹ OPDP and 0 CFU g ⁻¹ <i>L. plantarum</i>), diet 2 (10 g kg ⁻¹ OPDP), diet 3 (108 CFU g ⁻¹ <i>L. plantarum</i>), and diet 4 (10 g kg ⁻¹ OPDP + 108 CFU g ⁻¹ <i>L. plantarum</i>)	Skin mucus immunity ↑ Serum immunity ↑	Van Doan et al., (2019)
BFT	Biochar	Nile tilapia (<i>O. niloticus</i>) (36.2 g/fish)	B; only biochar, GB; biochar + glucose, while for the control (G) only glucose	Immune parameters ↑	Abakari et al. (2020)
BFT	Jaggery-based BFT	Nile tilapia (<i>O. niloticus</i>) (32.2 ± 10.1 g/fish)	22.5%, 27.7%, and 32.3% digestible protein (DP) and 6% lipid	Innate immunity ↑ Immune gene expressions ↑	Elayaraja et al. (2020)

(continued)

Table 9.2 (continued)

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Chestnut polyphenols (CSP)	Nile tilapia (<i>O. niloticus</i>) (12.77 ± 0.17 g)	0, 1, 2, 4, and 8 g kg ⁻¹ of CSP	Skin mucus immunity ↑ Serum immunity ↑	Van Doan et al. (2020a)
BFT	Watermelon rind powder (WMRP)	Nile tilapia (<i>O. niloticus</i>) (17.14 ± 0.12 g)	0 (Diet 1- control), 20 g kg ⁻¹ WMRP (Diet 2), 40 g kg ⁻¹ WMRP (Diet 3), 80 g kg ⁻¹ WMRP (Diet 4), and 160 g kg ⁻¹ WMRP (Diet 5)	Immune responses ↑	Van Doan et al. (2020b)
BFT	Phosphatidylcholine	Nile tilapia (<i>O. niloticus</i>)	0, 400, 800 and 1200 mg/kg of feed	Antioxidant enzymes Liperoxidation in liver ↓	Sousa et al. (2020)
BFT	Pineapple peel powder	Nile tilapia (<i>O. niloticus</i>) (20.91 ± 0.11 g)	0, 10, 20, 30 and 40 g kg ⁻¹ PAPP 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Van Doan et al. (2021a)
BFT	Watermelon rind powder (WMRP) + <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (16.57 ± 0.14 g)	Diet 1 (0 g kg ⁻¹ WMRP and 0 CFU g ⁻¹ L. <i>plantarum</i>) (control), Diet 2 (40 g kg ⁻¹ WMRP), Diet 3 (10 ⁸ CFU g ⁻¹ LP), and Diet 4 (40 g kg ⁻¹ WMRP + 10 ⁸ CFU g ⁻¹ LP) 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Van Doan, Seyed Hossein Hoseinifar, et al. (2021b)
BFT	Pineapple peel powder (PAPP) + <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (20.91 ± 0.11 g)	Diet 1 (0 g kg ⁻¹ PAPP and 0 CFU g ⁻¹ L. <i>plantarum</i>) (control), Diet 2 (10 g kg ⁻¹ PAPP), Diet 3 (10 ⁸ CFU g ⁻¹ LP), and Diet 4 (10 g kg ⁻¹ PAPP + 10 ⁸ CFU g ⁻¹ LP) 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Van Doan et al. (2021d)
BFT	Amla (<i>Phyllanthus emblica</i>) fruit extract	Nile tilapia (<i>O. niloticus</i>) (10.48 ± 0.56 g)	0, 5, 10, 20, and 40 mg kg ⁻¹ AFE 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Van Doan et al. (2022b)

BFT	Symbiotics	Nile tilapia (<i>O. niloticus</i>) (30–35 g)	BFT with and without symbiotics 40 days	Hematological parameters ↑	Laice et al. (2021)
BFT	Beneficial bacteria	Nile tilapia (<i>O. niloticus</i>) (8.63 ± 3.35g)	T1, SR control; T2, SR + SSP; T3, SR + MSP; T4, BF + MSP; T5, BF + SSP; T6, BF control 112 days	Innate immune response ↑	Mohammadi et al. (2021)
BFT	Coffee silverskin (CSS)	Nile tilapia (<i>O. niloticus</i>) (15.54 ± 0.21 g)	CSS1 (Control), CSS2 (10 g kg ⁻¹), CSS3 (20 g kg ⁻¹), CSS4 (40 g kg ⁻¹), and CSS5 (80 g kg ⁻¹) 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Hien Van Doan et al. (2021c)
BFT	Carbon source and stocking density	Nile tilapia (<i>O. niloticus</i>) (5.15 ± 1.12 g)	Low stocking density (LSD), 140 fish/m ³ and high stocking density, (HSD), 280/m ³ 98 days	Growth-related genes ↑	El-Hawary et al. (2021)
BFT	Probiotics	Nile tilapia (<i>O. niloticus</i>) (6.7 ± 0.2 g)	Stocking densities (D1, 120 fish/m ³ ; D2, 240 fish/m ³) with biofloc plus probiotics	Immune response ↑	Bañuelos-Vargas et al. (2021)
BFT	Spent coffee grounds (SCG)	Nile tilapia (<i>O. niloticus</i>) (15.25 ± 0.07 g)	SCG1 (control), SCG2 (10 g kg ⁻¹), SCG3 (20 g kg ⁻¹), SCG4 (40 g kg ⁻¹), and SCG5 (80 g kg ⁻¹) 8 weeks	Skin mucus immunity ↑ Serum immunity ↑	Van Doan et al. (2022a)
BFT	Feeding levels and stocking densities	Nile tilapia (<i>O. niloticus</i>) (3.2 ± 0.05 g)	Feeding levels (0, 15, 30, 45 and 100) and 2 stocking densities (500 fish/m ³ and 1000fish/m ³)	Innate immune response ↑	Sarsangi Aliabad et al. (2022)
BFT	Rambutan seed (RS)	Nile tilapia (<i>O. niloticus</i>) (14.77 ± 0.80 g)	0, 5, 10, 20, and 40 g kg ⁻¹ of RS 8 weeks	Skin mucus immunity ↑ Serum immunity ↑ Gene expressions ↑	Xuan et al. (2022)

(continued)

Table 9.2 (continued)

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Longan seed powder (LS)	Nile tilapia (<i>O. niloticus</i>) (13.82 ± 0.06 g)	Control (LS0), 10 (LS10), 20 (LS20), 40 (LS40), and 80 (LS80) g kg ⁻¹ LS 8 weeks	Skin mucus immunity ↑ Serum immunity ↑ Gene expressions ↑	Wannavijit et al. (2022)
BFT	Rambutan peel (RP)	Nile tilapia (<i>O. niloticus</i>) (17.14 ± 0.12 g)	0 g kg ⁻¹ (control - RP0); 10 g kg ⁻¹ (RP10); 20 g kg ⁻¹ (RP20); 40 g kg ⁻¹ (RP40), and 80 g kg ⁻¹ (RP80) 8 weeks	Skin mucus immunity ↑ Serum immunity ↑ Gene expressions ↑	Le Xuan et al. (2022)

Table 9.3 Increase disease resistance of tilapia cultured under biofloc system

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Orange peels derived pectin	Nile tilapia (9.09 ± 0.05 g)	0 (control in clear water), 0 (control in biofloc system), 5, 10, and 20 g kg ⁻¹ OPDP for 8 weeks	Resistance to <i>S. agalactiae</i> ↑	Doan et al. (2018)
BFT	Dietary digestible protein and digestible energy	Nile tilapia (<i>Oreochromis niloticus</i>) (1.25 ± 0.15 g)	Digestible protein (22, 26 and 30% DP) and digestible energy (3000, 3150 and 3300 kcal kg ⁻¹) 42 days	Ectoparasite spread ↓	Durigon et al. (2019)
BFT	<i>in-situ</i> and <i>ex-situ</i> biofloc	Nile tilapia (<i>O. niloticus</i>) (5.1 ± 0.05 g/fish)	T1-biofloc developed within the culture systems (<i>insitu</i>), T2-biofloc supplementation in fish feed (<i>exsitu</i>) and C- Control without biofloc	Resistance to <i>A. hydrophila</i> ↑	Menaga et al. (2019)
BFT	OPDP and <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (1.25 ± 0.15 g) 5.92 ± 0.08 g	Diet 1 (0 g kg ⁻¹ OPDP and 0 CFU g ⁻¹ <i>L. plantarum</i>), diet 2 (10 g kg ⁻¹ OPDP), diet 3 (108 CFU g ⁻¹ <i>L. plantarum</i>), and diet 4 (10 g kg ⁻¹ OPDP +108 CFU g ⁻¹ <i>L. plantarum</i>)	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2019)
BFT	Jaggery-based BFT	Nile tilapia (<i>O. niloticus</i>) (32.2 ± 10.1 g/fish)	22.5%, 27.7%, and 32.3% digestible protein (DP) and 6% lipid	Resistance to <i>A. hydrophila</i> ↑	Elayaraja et al. (2020)
BFT	Chestnut polyphenols (CSP)	Nile tilapia (<i>O. niloticus</i>) (12.77 ± 0.17 g)	0, 1, 4, and 8 g kg ⁻¹ of CSP	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2020a)
BFT	Watermelon rind powder (WMRP)	Nile tilapia (<i>Oreochromis niloticus</i>) (17.14 ± 0.12 g)	0 (Diet 1- control), 20 g kg ⁻¹ WMRP (Diet 2), 40 g kg ⁻¹ WMRP (Diet 3), 80 g kg ⁻¹ WMRP (Diet 4), and 160 g kg ⁻¹ WMRP (Diet 5)	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2020b)
BFT	Pineapple peel powder (PAPP)	Nile tilapia (<i>O. niloticus</i>) (20.91 ± 0.11 g)	0, 10, 20, 30 and 40 g kg ⁻¹ PAPP 8 weeks	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2021a)

(continued)

Table 9.3 (continued)

Studied systems	Studies parameters	Fish species	Doses and supplementations duration	Results	References
BFT	Pineapple peel powder (PAPP) + <i>L. plantarum</i>	Nile tilapia (<i>O. niloticus</i>) (20,91 ± 0.11 g)	Diet 1 (0 g kg ⁻¹ PAPP and 0 CFU g ⁻¹ <i>L. plantarum</i>) (control), Diet 2 (10 g kg ⁻¹ PAPP), Diet 3 (10 ⁸ CFU g ⁻¹ LP), and Diet 4 (10 g kg ⁻¹ PAPP + 10 ⁸ CFU g ⁻¹ LP) 8 weeks	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2021d)
BFT	Coffee silverskin (CSS)	Nile tilapia (<i>O. niloticus</i>) (15.54 ± 0.21 g)	CSS1 (Control), CSS2 (10 g kg ⁻¹), CSS3 (20 g kg ⁻¹), CSS4 (40 g kg ⁻¹), and CSS5 (80 g kg ⁻¹) 8 weeks	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2021c)
BFT	Amla (<i>Phyllanthus emblica</i>) fruit extract	Nile tilapia (<i>O. niloticus</i>) (10,48 ± 0.56 g)	0, 5, 10, 20, and 40 mg kg ⁻¹ AFE 8 weeks	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2022b)
BFT	Beneficial bacteria	Nile tilapia (<i>O. niloticus</i>) (8,63 ± 3.35g)	T1, SR control; T2, SR + SSP; T3, SR + MSP; T4, BF + MSP; T5, BF + SSP; T6, BF control 112 days	Resistance to <i>A. hydrophila</i> ↑	Mohammadi et al. (2021)
BFT	Spent coffee grounds (SCG)	Nile tilapia (<i>O. niloticus</i>) (15.25 ± 0.07 g)	SCG1 (control), SCG2 (10 g kg ⁻¹), SCG3 (20 g kg ⁻¹), SCG4 (40 g kg ⁻¹), and SCG5 (80 g kg ⁻¹) 8 weeks	Resistance to <i>S. agalactiae</i> ↑	Van Doan et al. (2022a)

resistance against *Streptococcus agalactiae* and *Aeromonas hydrophila* have been reported in Nile tilapia-fed orange peels derived pectin (OPDP) (Doan et al. 2018); OPDP and *L. plantarum* (Van Doan et al. 2019); in-situ and ex-situ biofloc (Menaga et al. 2019); jaggery-based BFT (Elayaraja et al. 2020); chestnut polyphenols (CSP) (Van Doan et al. 2020a); watermelon rind powder (WMRP) (Van Doan et al. 2020b); pineapple peel powder (PAPP) (Van Doan et al. 2021a); pineapple peel powder (PAPP) + *L. plantarum* (Van Doan et al. 2021d); coffee silverskin (CSS) (Van Doan et al. 2021c); amla (*Phyllanthus emblica*) fruit extract (Van Doan et al. 2022b); beneficial bacteria (Mohammadi et al. 2021); spent coffee grounds (SCG) (Van Doan et al. 2022a). It has been also reported that a biofloc system could reduce ectoparasite spread Durigon et al. (2019). A significant increase in disease resistance may be attributable to the presence of MAMPs in the biofloc system, which may be recognized as immunostimulants, resulting in higher resistance to diseases (Ekasari et al. 2014b). In addition, it may be due to the prebiotic and probiotic properties of feed additives, which are known to enhance the immune response and disease resistance of Nile tilapia (Cavalcante et al. 2020; Cano-Lozano et al. 2022).

9.3 Recirculating Aquaculture Systems (RAS)

9.3.1 Brief History of Development

Though in its infancy and still considered to be a recent innovation, the basic technology of RAS has existed for over 65 years, with the first, pioneering RAS research activity being conducted in Japan in the 1950s (Murray et al. 2014; Saeki 1958). According to Espinal and Matulić (2019), the technology of RAS including aquaponics has been developed over the past 40 years. In the 1970s, a German program demonstrated the feasibility of intensive carp production in RAS, and subsequently, the Danish Aquaculture Institute undertook an innovative effort to develop further technical aspects of RAS (Goldman 2016). The idea for commercial fish production in RAS was first fostered in Denmark in the mid-1970s, and the first commercial RAS was then built in 1980 (Warrer-Hansen 2015). The Danish efforts supported the development of one of the initial commercial RAS industries, specifically for the production of European eel (*Anguilla Anguilla*) (Goldman 2016). This work inspired the subsequent further development and uptake of RAS in other European countries in the late 1980s and 1990s (Martins et al. 2010). Over the last 25–35 years, a significant and growing experience in designing, building, and operating RAS, particularly in Nordic countries, has been reported (Dalsgaard et al. 2013). The initial success of the RAS-based European eel industry also inspired to development of RAS in North America (Goldman 2016). In China, marine RAS was initiated in the 1980s, and since then China has made considerable progress in RAS (Ying et al. 2015). Since the 2000s, further development of RAS has occurred in Europe, North America, Australia, and other aquaculture-producing countries (Espinal and Matulić 2019; Martins et al. 2010).

A significant acceleration in the development of RAS technology has been observed over the last two decades (Espinal and Matulić 2019), and RAS have become popular in recent years. RAS has been developed to grow fish where inadequate biophysical conditions, water scarcity, poor water quality, and unfavorable environment exist (Murray et al. 2014). According to Malone (2013), RAS provides an alternative production method when environmental regulations, disease, land availability, salinity, temperature, and water supply prevent more cost-effective alternatives. However, other factors stimulated the development and implementation of RAS. For example, RAS are increasingly being used for Mediterranean marine fish and salmonid production cycle, particularly for juvenile stages, before being transferred into outdoor grow-out systems, such as cages or flow-through raceways (Bostock et al. 2016; Clarke and Bostock 2017; Terjesen et al. 2013). In fact, RAS can be used for broodstock and seedstock production, which can support cage and net-pen aquaculture (Malone 2013). In Europe and North America, RAS was developed as an alternative to the cage culture of salmon (Murray et al. 2014). RAS has also been developed to culture exotic fish species, to avoid adverse effects on native species and biodiversity (Malone 2013; Murray et al. 2014).

9.3.2 Basic Principles of RAS Operation and Production

RAS are land-based, indoor fish-rearing facilities, where fish are stocked in tanks within a controlled environment, and where filtration is applied to purify water by removing metabolic wastes of stock, before being recirculated into the system itself. Water purification is achieved through mechanical and/or biological filtration, sterilization, and oxygenation. Different levels of sophistication and efficiency can be achieved, but generally, all RAS have a high degree (>90%) of water reuse (Murray et al. 2014; Badiola et al. 2012). RAS provides opportunities to enhance waste management, reduce water usage, and nutrient recycling (Martins et al. 2010; Murray et al. 2014; van Rijn 2013).

Although RAS have been initially developed and are ideally suited to produce freshwater as well as warm water fish species, RAS is flexible and can be modified and adapted to be operated with brackish and marine water as well as cold water species (Helfrich and Libey 1991). Therefore, by decoupling fish production from the marine environment, RAS may offer an alternative to traditional and net pen aquaculture (O'Shea et al. 2019). RAS can also provide suitable environmental conditions for fish species that are sensitive to water quality (Zhang et al. 2011). Despite RAS the potential to produce diverse seafood products, RAS is generally utilized to culture high-value fish, with high stocking densities and year-round production to offset high operational costs (Dalsgaard et al. 2013; Martins et al. 2010; Murray et al. 2014). Nevertheless, other fish species, including arctic char, clarias, halibut, pangasius, tilapia, and turbot are also commonly produced in RAS (Badiola et al. 2018; Ngoc et al. 2016a; Ngoc et al. 2016b; Summerfelt and Vinci 2008). The selection of fish species can be “market-driven” due to a high return on

investment to keep the RAS profitable (Badiola et al. 2017). The choice of fish also depends on the fast-growing and hardy fish in RAS (Badiola et al. 2018).

RAS can be categorized into five types: (1) hatchery and grow-out, (2) breeding, (3) long-term holding, (4) short-term holding, and (5) display (Yanong 2012). Moreover, RAS can be incorporated into an “integrated agriculture-aquaculture” system, which is known as aquaponics (Martins et al. 2010). Aquaponics is considered a particular type of RAS, where vegetable plants are included with fish to provide water filtration and crop diversification (Goddek et al. 2019). RAS have greater control over production outcomes, and the productivity of RAS depends on culture species, stocking densities, feeding rate, duration of the production cycle, and other management aspects. According to available scientific literature, the stocking densities of RAS range from 70 to 120 kg/m³ with feed conversion ratio (FCR) values from 0.8 to 1.1. RAS can be of various sizes including small, medium, and large (Helfrich and Libey 1991), with a large-scale RAS typically being able to produce 400e500 tons of fish per annum (Murray et al. 2014). However, even higher stocking densities and total production values are currently reported by some commercial producers. According to (Bregnballe 2015), RAS are highly productive intensive farming, which generates vast quantities of fish (500 tons/ha/year) in a comparatively small volume of water. Because of higher production, RAS is often referred to as “hyper” or “super” intensive farming (O’Shea et al. 2019).

9.3.3 RAS in Tilapia Culture

There is limited information regarding the application of RAS in Nile tilapia farming. It has been reported that RAS is a costly engineering approach, with a high initial investment in installation and operation (Murray et al. 2014). The reported annual production cost of RAS (US\$2250e8800 per ton) is considerably higher than conventional pond aquaculture (US\$2000 per ton) (Waite et al. 2014). Additionally, the Economic viability of RAS requires a long payback period, on average 8 years (Badiola et al. 2012). Wang et al. (2020) indicated that light intensity and photoperiod manipulation did not cause a significant chronic stress response in tilapia. This study demonstrated that light intensity, especially at 2000 lx, and photoperiod manipulation could stimulate the growth of tilapia in the RAS and significantly affect economic profitability. Another study using a different magnetic field (Hassan et al. 2018) showed that based on the growth, water properties, and serum biochemistry, it was concluded that magnetized water at 0.15 T intensity may improve tilapia growth in recirculating aquaculture systems. Recently, Deng et al. (2022) indicated that rearing fish larvae in RAS supports better survival compared to the flow-through system, while dietary probiotic supplementation further modulates the gut bacterial composition and stimulates the presence of beneficial bacteria during early life. It has been reported that RAS has a more stable and diverse microbial community composition, which could result in better growth performance compared to other culture systems (Deng et al. 2022). In addition, RAS could reduce

the ammonium and toxic gas in the culture system (Villar-Navarro et al. 2021; Nguyen et al. 2021).

A comparison between RAS, clear water, and biofloc system has been conducted in Nile tilapia. (Fleckenstein et al. 2018) indicated that clear water or hybrid systems may be a better choice for tilapia nurseries than chemoautotrophic biofloc systems due to the short-term periods in which nurseries operate and the volatility of nitrification in biofloc systems. In another study, Cao et al. (2020) indicated that There was no significant difference between the RASs and BFT aquaculture systems in terms of P recovery rate. The regular backwashing of the drum filter and biological filter in RAS accounted for $41 \pm 2\%$ of input N and $39 \pm 2\%$ of input P. Approximately 54% of unassimilated nitrogen N was removed by nitrification in the BFT aquaculture systems. The results from the present study suggest that nitrification may be the dominant pathway for ammonia removal in a BFT aquaculture system rather than by heterotrophic bacterial assimilation. RAS is characterized as a closed aquaculture and water reuse system; however, without an efficient and effective system for the treatment of discharged water and solids, this characterization only seems to indicate potential. Significantly, the treatment of the solids and water discharged from RASs has been suggested by (Luo et al. 2013). In BFT systems, most unused N and P are retained in biofloc and nitrate in tanks. The biofloc can be used for shrimp feed (Ray et al. 2017) or for feeding *Artemia* (Luo et al. 2017). Nitrate may be reduced by denitrification and dissimilatory nitrate reduction to ammonium (DNRA) activities in the BFT systems (Chutivisut et al. 2014). In these respects, the production activity in BFT systems may be more closed than that of RAS (Cao et al. 2020).

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