Review of Attached and Suspended Biomass Applications Integrated to Recirculating Aquaculture Systems

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Abstract

Intensification of aquaculture production must meet global protein requirements without placing excessive demand on earth’s resource capacity. Recirculating Aquaculture Systems (RAS) incorporate intensive culture protocols controlling the aquaculture production environment, promoting sustainable resource consumption. Both periphyton (fixed film) and biofloc (suspended growth) microorganism cultures are being integrated to aquaculture production in RAS, cages and, ponds, with varied climatic conditions and culture species, both independently and in combination. Periphyton and biofloc form the basis of aquaculture ecosystems wherein the highest trophic level is the commercial species under production. Production advantages include feed substitute grazing on produced biomass with improved Food Conversion Ratio (FCR) and growth rate, destressing, probiotic and pathogen control and, water quality management. Review of deployment of fixed film and suspended growth to RAS across materials, processes, conditions and species increases understanding of both commonality and distinctions in utilization of the options and, supports protocols for their use in intensive aquaculture production. New frontiers are predicted where periphyton and biofloc are commercially invested for live feed production, hatchery,
nursery and, intensive grow out aquaculture production across a widening range of species.

**Keywords**: periphyton, biofloc, biomass, consortia, RAS

1. Introduction

The significant increases anticipated in aquaculture production place resource demands on earth's capacity which will vary depending on the means of aquaculture production. It is important to consider the Life Cycle Assessment (LCA) of new production. Design basis for aquaculture production systems needs to address the next 37% increase in aquaculture production, as estimated by FAO to be in place by 2030 (FAO, 2018).

An increase of aquaculture production of 37%, in perspective, effectively requires duplication of over a third of every piece of existing aquaculture production from entirely new resource allocations. Alternatively, with aquaculture having reached the production capacity of earth's oceans, duplication of a third of the oceans' production capacity is envisioned, by 2030. The projected increase in aquaculture production will unlikely be able to rely solely on past successes in sustainable production (Martinez-Porchas & Martinez-Cordova, 2012). Meeting such a production increase requires innovative approaches to satisfy the critical need for input resources to production.
1.1 Sustainability of RAS

Design basis to achieve such expansion should include Key Performance Indicators (KPIs) to measure improvement over the status quo in meeting the critical need for resources as in Table 1, below.

**Table 1 Proposed KPIs for commercial RAS applications.**

| Number | Key Performance Indicator                                           | Priority |
|--------|---------------------------------------------------------------------|----------|
| 1.     | Food Security.                                                      | Medium   |
| 2.     | Carbon Footprint and GHG reduction                                  | Medium   |
| 3.     | Biosecurity for both farm and external environments.               | High     |
| 4.     | Land use minimization.                                             | High     |
| 5.     | Complete utilization of input nutrition and energy resources.      | High     |
| 6.     | Waste and effluent discharge management and reuse.                 | High     |
| 7.     | Water demand minimization.                                         | High     |

Aquaculture is receiving global attention along with cross-border attention to quality control, i.e., certification programs. Responsible sourcing of aquatic products, initiated by the global demand side market, increasingly underscore this view. In local contexts, permitting of new facilities is educated in the impact of resource constraints and their potential impacts on sustainable production, and may incorporate measures to reduce impacts.

Various forms of ocean farming system will produce measurable improvement, in line with the above KPIs, as new technology and systems are coming on line.
Extensive production systems based on large scale rice-fish systems are additional examples of traditional systems, which may be sustainably expanded, i.e., through more comprehensive utilization. Recirculating Aquaculture Systems (RAS) are being considered as an aquaculture system class with unique potential to meet critical aquatic production needs. RAS have the unique capability to be situated close to market, require less land and water supply than extensive systems, generate less effluent per unit production and, are able to utilize peri-urban land designated commercial, industrial or agricultural.

RAS exerts sufficient control over all aspects of the aquaculture production environment to permit high to ultra-high intensity culture, embodying less and potentially minimum resource intensity. The aquaculture production sector is responding with commercial RAS farms including Asia’s biggest farmer, Charoen Pokphand Foods CPL (CPF) planning full conversion to indoor operations (Gibson, 2018) and, with the advent of large-scale land based salmon farm RAS (Undercurrent News, 2018; Poppick, 2018). New RAS facilities frequently reference anticipated continuous improvement in terms of resource efficiency. Resource efficiency is referred to in terms not only of meeting but of eventually exceeding environmental standards, setting the stage for envisioned expansion of facilities.

1.2 Integrated Biomass RAS

Implementation of commercial RAS has been historically impeded by
uncertainty of assured revenue. Reasons for the lack of confidence in RAS commercial investment include a lack of prototyping of specific systems and species and, slow adoption of RAS concepts by the aquaculture industry. The economics of RAS have also been compared unfavorably with less intensive methods of aquaculture production, typically due to perceived lower capital investment, lower technical level of operation, reduced operation and maintenance cost. The overriding fact is that extensive to intensive pond culture is the status quo in global terrestrial production. The decisions currently being taken to invest in large scale RAS reflect on the ability of RAS to reduce footprint, making sites available and, to control environmental factors, which is effective in overcoming permitting hurdles. From the perspective of sustainability, permitting itself is an important issue. The ability to permit biosecure facilities near urban centers allows the reduction of distance to market, important in reducing carbon footprint due to product transport. RAS facilities implemented close to or, within urban center markets for aquaculture product also move aquaculture employment closer to urban centers, with further corresponding reductions in carbon footprint possible, along with potential social advantages.

Beyond permitting advantages, other commercial advantages include improving food conversion ratio (FCR), growth rate and, survival rate. Application of production-integrated biomass technologies can support intensification in RAS through the maintenance of water quality and, the recycling of wastes to positive probiotic and nutrition inputs. For example, only about one third of feed is beneficially utilized by production species in conventional aquaculture production systems (Avnimelech, 2006).
The capability to achieve production from a larger proportion of input feed results in lower carbon footprint for feed transport.

Browdy, Ray, Leffler and Avinimelech (2012) estimated that a conventional pond rearing system might be considered to produce only around 2,000kg of fish/hectare and, to lose about 45,000m³ of water to evaporation, seepage and drainage per year. This results in water demand of 45m³/kg fish at a production rate of 0.2kg fish/m². Water conserving RAS production, on the other hand was estimated to consume 1m³ water/kg fish at a production rate of 10 to 100kg fish/m².

Improved sustainability of aquaculture production needs to be considered against the backdrop of the huge projected increase in aquaculture production (FAO. 2018). While the status quo of aquaculture production is largely based on extensive systems, increase in production should be implemented using the most sustainable production systems, which requires certainty of RAS economics in order to attract the appropriate capital investment.

Both periphyton and biofloc microorganism cultures are utilized as integrated components of aquaculture production to produce positive impact on economics of aquaculture production of commercial species. This review looks at the indications within existing research that integrated biomass can contribute effectively to commercial RAS. In addition, it investigates whether commercial RAS utilizing integrated biomass will see improvement in sustainability performance and, ease of permitting. Water consumption, feed sourcing and, effluent discharge are key areas influencing sustainability and, are increasingly pivotal in permitting of production.
facilities and capacity expansions hence, investors will increasingly seek improvements in these areas.

1.3 The Role of Microbes

Production-integrated periphyton and biofloc applications involve captive ecosystems of microorganisms, consisting of multiple genera of bacteria, algae, plankton and simple multi-cellular organisms. Periphyton is a term used for the matrix of biota which form on surfaces, while biofloc and biofloc technology (BFT) refer to the matrix of biota in suspension as “flocs”. Periphyton biomass develops similarly to that developed in external biofilters with low-light environments highly dominated by bacteria (Avnimelech, 2006). Use of periphyton in production volumes is steadily developing (Martınez-Cordova, Emerenciano, Miranda-Baeza and Martinez-Porchas, 2015) although more research appears to be conducted in commercial demonstration than in academic settings. BFT is a well-developed field with prolific examination in both research and commercial settings (Crab, Delfoirdt, Bossier and Verstaete, 2012). It is important to maintain the distinction between biofloc and periphyton in investigations to permit optimization of the different opportunities presented by their different constituents and operational mechanisms.

Inside production tanks, the presence of biota, being supported with waste products and un-consumed feed, forms the basis of a more expansive ecosystem wherein the highest trophic level is the commercial species under production. The biota typically act as an in-situ biofilter and are able to improve or, manage water quality. The biota biomass is directly exposed to the commercial species during rearing and utilized as
feed by the cultured species, which may provide both nutrition and health benefits
(Nevejan et al., 2016). Significant research evidences that the presence of biota biomass
within production volumes supports quorum sensing based resistance to disease
outbreak for a large number of fresh water and sea water species (Turan, Chormey,
Buyukpinar, Engin and Bakirdere, 2017; Ruwandeepika, Karunasagar, Bossier and
Defoirdt, 2015; Defoirdt, 2014; Pande, Scheie, Benneche, Wille, Sorgeloos, Bossier
and Defoirdt, 2013; Crab, Lambert, Defoirdt and Verstraet, 2010; Nhan, Cam, Wille,
Defoirdt, Bossier and Sorgeloos, 2010; Defoirdt, Boon, Sorgeloos, Verstraete and
Bossier, 2007).

In addition, biomass may prove to contain or, to be manipulated to contain
compounds which have similar effects to those provided by added probiotics in
aquaculture production systems, i.e., through accumulation of poly-beta-hydroxybutyrate
(PHB) present in biomass which is consumed and thereafter protects production species
from infection by pathogenic bacteria (Crab et al, 2012). Further, bacteria utilize
digestive enzymes which may be doubly employed after consumption by production
species (Martinez-Cordova et al., 2015). Bacterial biomass may also encourage the
development and presence of zooplankton providing trophic upgrading of waste feed
and byproducts to multi-cellular, live feed organisms (Nie, Liu, Liu, Yu, Liu, and Zhou,
2017).

1.4 Periphyton (Fixed Film)

Fixed film microorganisms or, periphyton consist of bacteria, algae and plankton
typically in a matrix bonded with extracellular polymeric substance (EPS) produced by
the (bacteria and algae) and attached to a surface (Pandey, Bharti and Kumar, 2014). In an aquaculture production volume, surface area is needed upon which biofilm may form and reside where, the added material provided to increase surface area is sometimes referred to as substrate. Substrate can be provided by addition of media, either living and structural (e.g., vegetative roots or, macroalgae) or purely structural (i.e., mesh). Periphyton microbiology will vary depending on the complexity of the media, which has also been studied in detail in wastewater treatment engineering applications (Eding, Kamstra, Verreth, Huisman and Klapwijk, 2006). Incorporation of media used in biological treatment of wastewater, to maintain advantageous biota capable of processing aquaculture waste in-situ may receive more attention in future research.

On a 2D fixed film surface the microbiology is simplified with inner, intermediate and outer layer defined by layers of aerobic, facultative and, anaerobic bacteria. These layers work cooperatively utilizing microbiological consortia interaction and molecular diffusion to process and cycle nutrients in the water through different pathways to the respective bacteria (Eding et al., 2006; Paerl and Pinckney, 1996). Typically, the anaerobic layer at the inner level will generate gases which can force biofilm to "slough" from the surface. However, where the full biomass is exposed to the production species it is subject to grazing by the culture species which may replace sloughing action. In the case of living 2D media, e.g., seaweed surface or suspended vegetation root surface, there are beneficial exchanges between the living media and, the attached growth microorganisms and, water quality is influenced by the living media as well as by the attached biomass growth on it (Morgan, Martin and Bouchard, 2008).
The media may alternatively be provided with three dimensional (3D) depth in the form of structural crevasses in the material surface, cavities or, layers of porous media. 3D effects include creation of zones with lower agitation and/or oxygen supply. In addition to supporting facultative and anaerobic bacteria, the 3D effects may provide refuge for development of additional trophic levels containing zooplankton which feed on bacteria and, may adhere to periphyton. Although several studies looking at feed organisms association with media have not found correlation (Ahsan, Sharker, Alam, Siddik and Nahar, 2014), studied substrate, such as bamboo does not constitute 3D media. The more complex the media, the more complex the ecosystem that may be supported by it. Thus, 3D media may prove to have the potential to upgrade bacterial biomass to a more varied source of live feed, produced in-situ from waste products.

Research into the microbiology of 3D media exists as studies of biofiltration (or biological treatment) media in wastewater treatment systems and, in biofilters external to aquaculture production tank volumes of commercial species. There are limited studies which evaluate the advantages of 3D media within production volumes hence, this is an identified area of further potential research.

The periphyton approach in aquaculture production can be considered as a biofilter element that is integrated to the production tank of a commercial species. Biofilters are operated as multistage water treatment units and, may emit high levels of bacterial biomass to be recirculated into the production tank in RAS. Different types of RAS biofilter have been ranked by way of comparison (Malone and Pfeiffer, 2006). The microbiological population of external biofilters is employed to improve water quality.
and is not typically designed to convert waste feed to useful biomass, to subsequently become available as a probiotic or nutritional source to the commercial species, as has been discussed by Avnimelech, (2006). However, some evidence of biofilters improving FCR and nutrition over non-biological treatment types constituted early recognition that integrated production of microorganisms would have a positive impact on aquaculture production. Study has been extended for comparison of biofiltration with periphyton media and biofloc, integrated to aquaculture production tanks, on the basis of their performance in management of water quality (Crab, Avnimelech, Defoirdt, Bossier and Verstaete, 2007).

Reviews either dedicated to or, containing in-depth review of periphyton media applications have described the use of synthetic (e.g., meshes or plastic surfaces) and natural material (e.g., bamboo or tree branches) periphyton media (Pandey et al., 2014; Azim and Little, 2006; Verdegem and Ekram-Ul-Azim, 2001; Dutta, Kalita and Phukan, 2018; Nevejan, De Shryver, Will, Dierckens, Baruah and Von Stappen, 2016; Martinez-Cordova et al., 2015).

Macroalgae has been investigated as a living media in aquaculture application, where the seaweed action on water contamination is synergistic with biofilm resident on the seaweed (Brito, Arantes, Magnotti, Derner, Pchara et al., 2014; Chopin, Buschmann, Halling, Troell, Kautsky et al., 2001). Seaweed is a living and potentially edible periphyton substrate, which carries a microbiome of microorganisms which assist in growth and protect the host macroalgae (Egan, Harder, Burke, Steinberg, Kjelleberg and Thomas, 2012; Singh and Reddy, 2016). Coral reefs are a form of living fixed film
media whose biome is also inclusive of a range of bacteria in symbiosis with algae (Ainsworth, Krause, Bridge, Torda, Raina et al., 2015). Understanding of coral bacterial biota may benefit coral management as well as improved biota integration to aquaculture production.

1.5 BFT (Suspended Growth)

Biofloc technology (BFT) employs a suspended complex of microbes and detritus and may be considered similar to a periphyton application, where attachment is to the dynamic flocs which have similarities to periphyton biomass. BFT is the utilization of suspended growth biomass integrated into aquaculture production. The integrated suspended growth approach provides biodegradation of water borne nutrients. Genomic research carried out by Vargas-Albores, Martinez-Cordova, Gollas and Martinez-Porchas (2019) suggested that both nitrogenous and carbonaceous compounds could be processed by bacteria within a unit biofloc.

Study of BFT initially focused on performance in management of water quality (Crab et al., 2007). Reviews of BFT applications, including those reviews which consider periphyton systems against the same criteria of performance, demonstrate the appreciation for microbial systems to benefit health, nutrition, economics and, sustainability in aquaculture production (Azim and Little, 2006; Nevejan et al., 2016; Martinez-Cordova et al., 2015). One key factor in the adoption of BFT is the ability and preference of the production species to consume feed suspended in the water column (Kent, Browdy and Leffler, 2011). The ability of Pacific Whiteleg Shrimp Litopenaeus vannamei to consume non-flocculated microalgae was examined by Kent et al. (2011).
Biofloc can be manipulated to produce a larger floc to facilitate uptake by a greater variety of species but, planktivorous species are the most natural match for BFT.

Extensive reviews of the development of BFT technology have described a range of applications within the past decade. Significant success has been noted at a commercial level due to the probiotic and nutritional benefits of BFT but, the technology is far from having become the status quo or, business-as-usual in aquaculture production (Crab et al., 2012; Kavitha et al., 2018; Emerenciano, Gaxiola and Cuzon, 2013).

BFT has demonstrated that integrated biomass systems are effective from an economic perspective but, more analysis is required to define whether BFT is suitable to meet the criteria of super intensification of RAS and, for which production species it will be commercially advantageous to employ at nursery and/or grow out stages.

1.6 Periphyton and BFT Concurrent Integrated Biomass

Both BFT and periphyton aquaculture production systems employ microbial matrices within the production volume. Microbial matrices are resilient and, both types are able to support both nitrification and carbon conversion simultaneously. However, the addition of periphyton to BFT production increases the complexity of the production system and may lead to inconsistencies within the production volume or, local or system wide inefficiencies due to environmental factors compounded by the presence of both.

Mixing, diffusion or current flow may be altered by inserted material barriers or changes in the viscosity and/or other properties of water may occur due to suspended
solids. Such changes may amplify impact of tank geometry and equipment selection in experimental and full-scale application. Biofloc is somewhat more energy intensive than periphyton due to the requirements of biofloc suspension and high dissolved oxygen. Barriers and lower light environment resulting from periphyton addition may positively influence feeding behavior (lower perceived competition for food), reduce levels of hierarchical stress and, harmonize sexual development but, may complicate suspension and aeration to suit biofloc. Concurrent application needs to keep in view the different characteristics of each technology, for example, where a particular effect such as visibility reduction may be cumulative in application of both technologies.

The advantages of including periphyton in an intensive production BFT system for *L. vannamei* (300 shrimp/m²) were revealed where periphyton acted as an additional food source, in study conducted by Ferreira, Lara, Wasielsky and Abreu (2016). Schweitzer, Arantes, Baloi, Cosodio, Vinatea, et al (2013) performed a similar experiment finding that even with low biomass development on periphyton media that weight gain, final mass and survival rate increased with periphyton addition and, concluded that periphyton media most likely reduced stress to achieve these results. Investigation into the understanding of genomic functions is establishing many potential pathways for analyzing and improving complex production systems.

**1.7 Genomics in Integrated Biomass Systems**

Increasing use of genomic microbiological techniques increases definition of cause and effect related to physical inputs and species traits. However, whether approaching combined systems from a micro or macro level there are potential
interactions whose influence cannot be easily quantified but, which may significantly affect results. Environmental factors produced by the application of combined technologies are able to have positive and/or negative impact on aquaculture production both directly and, through cumulative effect.

Genomic techniques are being employed to good effect toward the optimization of both BFT and periphyton systems. Both systems benefit from studies completed to analyze fixed film and suspended growth microbial consortia in wastewater treatment systems (Wagner, Loy, Nogueira, Purkhold, Lee and Daims, 2002). Martinez-Porchas and Vargas-Albores (2017) reviewed the status of microbial metagenomics in aquaculture, describing studies of microbial diversity, consortia, antibiotic resistance, probiotics amongst others and pointed out the criticality of more study into high intensity aquaculture production systems. An example of analysis of the aquatic microbial diversity in RAS, culturing tilapia with both BFT and periphyton, was investigated by da Silva, Cavalcante, de Carvalho, Vieira, e Sa and de Sousa (2016) with the conclusions that higher final weight resulted from both types of application but, only periphyton demonstrated bacterial consortia carrying lower pathogenic potential. Periphyton may be represented by study of autotrophic nitrifying biofilm as carried out by Kindaichi, Ito and Okabe (2003) who showed that the biofilm was diverse and stable and, that carbon metabolism coexisted with nitrifiers and denitrifiers and, the biofilm represented an efficient food web preventing accumulation of metabolites or wastes.
In the case of BFT, a production of 100 to 600 kg/hectare per day of protein is possible from waste and carbon addition but, the means to optimize the metabolism of the complex microbial consortia responsible, relative to cost of inputs and value of outputs, requires further genomic investigation (Martınez-Porchas and Vargas-Albores, 2017).

1.8 Manipulation of Integrated Biomass Systems

BFT experiences mounting research interest into the extent to which biofloc can be manipulated to improve characteristics of biofloc and, positively influence production performance and quality. Martinez-Cordova et al (2015) reviewed progress in the field and produced an updated table outlining manipulation of control parameters (C:N ratio, energy, DO, temperature, pH and ionic levels and, light exposure) to produce influence in biofloc composition, size, structure and stability.

While considering extensive, pond rearing of fish, Azim, Wahab, van Dam, Beveridge, Milstein and Verdegem (2001) found that such systems with added periphyton media could yield up to about 5 metric tonnes per hectare, without supplementary feed. In the case of freshwater prawn, Macrobrachium rosenbergii rearing at 20,000 juveniles per hectare, still an extensive system, Haque, Akter and Pervin (2014) concluded that periphyton-supported farming, could yield over 36% improvement in net yield through using additives such as corn flour to maintain a high C:N ratio. Periphyton have been shown to maintain higher metabolism, versatility, functional redundancy and more consistent carbon-source utilization than biofloc (Lyons
and Dobbs, 2012) which, should support more exploration of the potential for manipulation of periphyton with a focus on carbon supplementation.

1.9 Species Performance in Integrated Biomass Systems

There are abundant examples of application of both periphyton and BFT to aquaculture of a variety of species of fish and shrimp and, strong evidence for commercial potential which has been assessed in the many available reviews. Shrimp appear to favor periphyton as grazing is more efficient for their feeding approach. Some fish are planktivorous and can consume biofloc readily but, herbivorous, detritivorous and omnivorous fish are claimed to prefer periphyton (Martínez-Cordova et al., 2015).

Periphyton supported culture has been examined at full-scale most often with extensive pond systems. Pandey et al (2014) have summarized research on the effects of a variety of substrates. Organic materials (e.g., bamboo, bagasse, straw, etc.) are generally favored by extensive pond farmers. Significant positive indications for commercial production have been demonstrated for rohu (L. rohita), common carp (Cyprinarchus carpio), penaeid shrimp (Penaeopenaeus paulensis, Penaeusesculentus, L. vannamei, P. monodon, M. rosenbergii, Farfanteopenaeus merguiensis), tropical rock lobster (Panulirusornatus) and, bottom feeders. Azim and Little (2006) have reported increased fish production of 30-115% and 30-210% in carp monoculture and polyculture respectively and production of 9t/ha/year in carp polyculture without any supplementary feed.
However, BFT is considered viable for *M. rosenbergii*, *L. vannamei* and, tilapia such as *Oreochromis niloticus* x *Oreochromis aureus* (Crab et al., 2012). Emerenciano et al (2013) summarized the history of BFT from development in France in the 1970’s with different penaeid species, through to the 1990’s in Israel and USA with tilapia and white shrimp, *L. vannamei*. In 1988, 1000m$^2$ concrete RAS tanks demonstrated 20 to 25 ton/ha/year with two crops in USA and, Belize Aquaculture farm achieved about 11 to 26 ton/ha/cycle in ponds. Emerenciano et al (2013) further reported that Marvesta farm, USA reaches output of approximately 45 tons of shrimp per year in 570 m$^3$ indoor raceways while, BFT is employed in large-scale shrimp farming in Asia, Latin and Central America and, in smaller operations in USA, South Korea, Brazil, Italy, China and other countries.

### 1.10 Protocols for Integrated Biomass Application to RAS

The emerging technology of BFT and periphyton applications is being applied to cages, ponds and Recirculating Aquaculture Systems (RAS) with varied climatic conditions and culture species; to improve health, growth rate and utilization of pond volumes and, to decrease operational costs and environmental impacts. Successful designs must achieve commercial advantages to meet the KPI's outlined in table 1. Design requirements need to be identified along with opportunities to be addressed and, commercial potentials available, as set out in table 2, below.
Table 2 Proposed design requirements for commercial RAS applications.

| Sustainability, social and regulatory design requirement | Opportunity                                                                 | Commercial potential                                                                 |
|----------------------------------------------------------|-----------------------------------------------------------------------------|---------------------------------------------------------------------------------------|
| Localization of feed supply\(^{a1}\)                     | Produce feeds from agriculture & aquaculture waste;                         | Feed price risk reduction; Potential to reduce feed costs;                            |
|                                                          | Customization and quality control of feed;                                 | Lower cost substrates input as feed resources.                                        |
|                                                          | Lower carbon footprint of feed value chain.                                 |                                                                                       |
| Aquaculture production free of climate, land and water constraints | Aquaculture close to market, infrastructure, personnel;                     | Low product transport costs; Dedicated market; Access to personnel & R&D facilities;  |
|                                                          | Flexible land selection;                                                   | Lower land costs;                                                                      |
|                                                          | Lower water sourcing & discharge constraint;                               | Lower water related costs.                                                            |
|                                                          | Lower carbon footprint of product,                                         |                                                                                       |
| Production of live feed – in-situ integrated to production\(^{a1}\) | Survival rate & disease control;                                            | Capture value of feed production; Bio-security improved with reduced interfaces and on-farm feed and health control. |
|                                                          | Input lower impact substrates as resources;                               |                                                                                       |
|                                                          | Improved growth rates and reduction of FCR;                                |                                                                                       |
|                                                          | Lower carbon footprint of feed.                                            |                                                                                       |
| Disease management technology free of antibiotics\(^{a1}\) | Use of farm generated probiotics and feed replacements.                    | Improved market value; Improved ability to customize product to market.               |

\(^{a1}\)Aquaculture production systems need to be certified inclusive of new systems

Some important factors need to be examined from the perspective of implementation of BFT, periphyton or, combined systems to intensive RAS, which might include those presented in table 3, below.
Table 3 Factors in implementation of integrated periphyton and biofloc to RAS.

| Number | Factor                                                                 | Priority |
|--------|------------------------------------------------------------------------|----------|
| 1.     | System optimization by species and geography, including biomass        | Medium   |
|        | potential utilization rate and productivity per unit volume.            |          |
| 2.     | Water quality management efficiencies (water quality usage and management) | High     |
| 3.     | Performance in recycling waste to nutritional and health promoting biomass, | High     |
| 4.     | Energy requirements.                                                   | High     |
| 5.     | Retention viability between crops.                                     | Medium   |
| 6.     | Robustness in commercial application.                                  | High     |
| 7.     | Capital and operating costs and financial modelling                    | High     |
| 8.     | Commercial certification and permitting aspects.                       | High     |
| 9.     | Carbon footprint of feed and other resource consumption and, delivery to market. | Medium   |

2. Conclusions

The objective of achieving sustainable intensification of aquaculture production to protect earth's resources, while drastically increasing overall aquaculture production, must include commercial means to implement intensive to hyper-intensive RAS. The relative opportunities in terms of improving water quality and, generating health-promoting and nutritional biomass provided by periphyton and biofloc approaches justify further study, the basis for which has been reviewed. There is a need to process completed research on the integrated biomass approaches and, to incorporate it into
protocols of commercial implementation so that the challenges of massive expansion of
the industry can be met sustainably. Intensive, scientific approach to sustainable
commercial expansion is less well represented than it needs to be to support highly
intensive RAS, anticipated to be required in meeting massive increase in aquaculture
production under global resource constraints.

Examples of application of integrated biomass to aquaculture are abundant,
providing strong evidence for extended commercial potential although, to a lesser extent
for RAS specifically. The practical application to date tends to reside in extensive
systems while, implementation of applications of more sophisticated technology needed
to raise BFT and periphyton productivity are still limited (Van Dam, Beveridge, Azim
and Verdegem, 2002). There is still insufficient demonstration of the established
potentials of production integrated biomass in RAS systems, to enable their
incorporation in large scale commercial facilities.

Examination of effects of periphyton and BFT deployment in RAS across varied
processes, conditions and, species will support protocols to improve commercial RAS
production economics. More research is expected to reveal commercial capabilities for
periphyton systems, which can deliver feed to a wider range of species than can BFT.
New frontiers are predicted for optimized periphyton and BFT systems, designed to
support intensive RAS grow-out, with further applications live feed production and,
hatchery and nursery operations, across a widening range of species.
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