


Review

# Research Advances on Tilapia Streptococcosis

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**Abstract:** *Streptococcus agalactiae*, often referred to as group B streptococci (GBS), is a severe pathogen that can infect humans as well as other animals, including tilapia, which is extremely popular in commercial aquaculture. This pathogen causes enormous pecuniary loss, and typical symptoms of streptococcosis—the disease caused by *S. agalactiae*—include abnormal behavior, exophthalmos, and meningitis, among others. Multiple studies have examined virulence factors associated with *S. agalactiae* infection, and vaccines were explored, including studies of subunit vaccines. Known virulence factors include capsular polysaccharide (CPS), hemolysin, Christie-Atkins-Munch-Peterson (CAMP) factor, hyaluronidase (HAase), superoxide dismutase (SOD), and serine-threonine protein kinase (STPK), and effective vaccine antigens reported to date include GapA, Sip, OCT, PGK, FbsA, and EF-Tu. In this review, I summarize findings from several studies about the etiology, pathology, virulence factors, and vaccine prospects for *S. agalactiae*. I end by considering which research areas are likely to yield success in the prevention and treatment of tilapia streptococcosis.

**Keywords:** tilapia; *Streptococcus agalactiae*; streptococcosis; virulence factors; subunit vaccine



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## 1. Introduction

*Streptococcus agalactiae* (*S. agalactiae*), also commonly referred to as group B streptococcus (GBS), is a severe pathogen that can infect humans and a diversity of other animals, including reptiles, frogs, bovines, fish (including tilapia), and pigs, among others [1–3]. Streptococcosis—the disorder caused by GBS infection—is a major obstacle faced by the tilapia aquaculture industry: in 2011, streptococcosis caused the loss of \$40 million in the tilapia industry in China, owing to high morbidity and mortality, which can reach up to 80% in outbreaks [4,5]. Given this potential for loss, it is unsurprising that researchers are striving to develop effective means to control tilapia streptococcosis outbreaks. There has been some progress in research regarding serotypes and virulence mechanisms, and there are some promising prospects for effective vaccines; this review considers progress over the past several years from investigations about these topics.

GBS, like many other pathogenic species, possesses many virulence factors, biomolecular factors that promote the ability of pathogens to infect and/or damage hosts [6–8]. Among GBS virulence factors, a large number are known to affect adherence and invasion of host cells, as well as evasion of host immunity [7]. A better understanding of these virulence factors can support the development of control and therapeutic strategies. Studies of GBS isolated from humans have split virulence factors into pore-forming toxins, factors for immune evasion, resistance to antimicrobial peptides (AMPs), host-cell adherence and invasion, and other virulence factors [7–9]. Pore-forming toxins—vitaly important pathogenesis factors—can facilitate the entry of GBS into host cells. The most common kinds of pore-forming toxins are  $\beta$ -hemolysin/cytolysin ( $\beta$ -H/C) and Christie-Atkins-Munch-Peterson (CAMP) factor. GBS can encode several virulence factors that promote immune evasion. The adherent factors, such as fibrinogen-binding protein (FbsA/B/C), laminin-binding protein (Lmb), the immunogenic bacterial adhesin (BibA), Pili (PilA/B/C),

serine-rich repeat (srr),  $\alpha$  C protein, and others, are responsible for GBS binding to extracellular matrix (ECM) components [7,8]. As a  $\beta$ -hemolytic bacterium, GBS has the hemolytic property, which is essential for immune evasion and further infection [7,8]. Immune evasion factors include C5 $\alpha$  peptidase (ScpB), hemolytic pigment, superoxide dismutase (SodA), HylB, and so on. Several classes of resistance to AMPs factors have been discovered, like D-alanylation of LTA and penicillin-binding proteins (PBPs). In addition, pili possess the ability to mediate GBS resistance to AMPs [7–9]. Recently, other virulence factors, cyclic di-AMP and cell wall-anchored ectonucleotidase (CdnP) have been discovered [10].

The abuse of antibiotics to control streptococcosis can cause many problems, such as the resistance of strains [11,12]. Moreover, the application regimes for antibiotics can be inaccurate, and antibiotics can deleteriously affect food quality and safety. Vaccines—which induce adaptive immune responses—can overcome some of these challenges. Given their potent efficacy, the development of vaccines for preventing GBS outbreaks has been widely investigated, including inactivated vaccines, attenuated vaccines, subunit vaccines, and DNA vaccines.

## 2. The Etiology of Tilapia Streptococcosis

Several studies have sought to identify the pathogen responsible for tilapia streptococcosis. Although this topic has been controversial, it is now acknowledged that the major pathogen for tilapia streptococcosis affecting aquaculture is *Streptococcus agalactiae* (GBS) [13]. In addition, there is evidence that *Streptococcus iniae* can cause similar disease symptoms, and it is now clear that many aspects contribute to outbreaks of tilapia streptococcosis, including environmental conditions as well as the presence of certain viruses and fungi [14,15].

*S. agalactiae*, a Gram-positive bacterium, has a spherical shape with dimensions ranging from 0.2 to 1.0 microns in diameter [16]. When cultured, *S. agalactiae* may grow in pairs or chains. It is a facultative anaerobe and is catalase and oxidase negative; it possesses the capacity for lactic acid fermentation [17] and is classified into Lancefield group B streptococcus (based on the presence and type of its surface antigens) [18]. The serotypes of GBS strains are assessed based on a capsular polysaccharide antigen, and to date, GBS has been classified into ten kinds of distinct serotypes, including Ia, Ib, II–IX [19,20]. Among them, serotypes Ia, Ib, II, and III are the most prevalent in tilapia infections. Notably, in 2018, our group identified that among others also the IX serotype was an important agent in tilapia streptococcosis [11]. This serotype has the potential to become a major infectious strain for tilapia.

Tilapia streptococcosis is mainly observed in temperate and tropical tilapia-culturing areas, including China [13], Malaysia [21], Austria [22], Brazil [23], Columbia [24], and Thailand [25], among other areas. Typically, large-scale infections of tilapia will breakthrough in relatively warm seasons, especially summer. The mortality rate can reach 50–70% in less than a week [26].

Although there is variability among tilapia, some of the most common symptoms and pathological signs include erratic swimming and loss of orientation, unilateral or bilateral exophthalmia (also named “pop-eye”), anorexia, abdominal distention, darkening of the skin, and hemorrhaging skin around the anus or at the base of the fins, as well as abdominal dropsy, pale, but enlarged liver, inflammations around the heart and kidney, and meningitis [26,27]. There has been heated discussion about potential modes of transmission: the introduction of infected fish is the most common mode, yet there is also solid evidence supporting vertical transmission [26].

## 3. Virulence Factors

### 3.1. Capsular Polysaccharide

The capsular polysaccharide (CPS) is a pathogenic factor widely distributed among *Streptococcus* serotypes; this molecule comprises glucose, galactose, N-acetylglucosamine,

and N-acetylneuraminic acid [28]. CPS has been a traditional epidemiological tool for investigating *S. agalactiae* infections in humans [29]; it is generally used for strain typing. CPS is known to contribute to disease severity [30], and molecular serotyping techniques have an elevated discriminatory power for epidemiological studies [31].

Experiments have shown that bacterial cells lacking the capacity to produce CPS lost their virulence in a neonatal rat model of lethal group B *Streptococcus* infection [32,33]. Similarly, using tilapia challenge assays, CPS-deficient GBS showed attenuated pathogenesis. And this mutant GBS was also cleared more easily from tilapia spleen tissue compared to the wild type GBS strain examined [34]. It is now understood that CPS can suppress the aggregation of the complement factor C3b to inhibit phagocytotic killing by host cells [35,36]. Sialylated CPS mimics cell surface carbohydrate epitopes and thereby decreases host immune recognition [37]. GBS is known to regulate CPS production in response to environmental signals to adjust the capacity for adherence and host invasion [38]. Further, Barato et al. (2016) used an infection model and showed that mutant GBS cells (unencapsulated) displayed increased adhesion to the tilapia intestinal epithelium [39].

### 3.2. CAMP Factor

The CAMP factor (also called co-hemolysin) is encoded by the *cfb* gene. This is an extracellular protein of 23.5 kDa [40] that functions to promote GBS pathogenesis [41–43]. In vivo assays have shown that partially purified CAMP factors can lead to death in rabbits [44]. Briefly, currently, understanding of the pathogenic impacts of CAMP includes its oligomerization to support forming discrete pores on host membranes and its binding to glycosylphosphatidylinositol (GPI)-anchored proteins, which can promote cell lysis [45]. Recently, two research groups identified the structure of CAMP, which provided more details about its perforating function [46,47]. Podbielski et al. (1994) demonstrated that a full-sized recombinant CAMP-factor exerts co-hemolytic activity [42]. Note that *cfb* is widely used as an identification marker for *Streptococcus agalactiae* due to its exclusive expression in GBS [40]; however, more recent studies have shown that there are homologs of *cfb* in *Streptococcus pyogenes*, *Listeria monocytogenes*, *Mobiluncus curtisii*, and *Propionibacterium acnes* [48,49].

### 3.3. HAase (Hyaluronidase)

Encoded by the *hylB* gene, GBS promotes its invasion of hosts by secreting HAase to specifically hydrolyze the host-cell-wall component hyaluronic acid into unsaturated disaccharide units as the end product. Its degradative enzyme function by cleaving the glycosidic bond between N-acetyl- $\beta$ -D-glucosamine and D-glucuronic acid residues destroys the host's normal connective tissues and nervous system, which leads to expose to the host tissue cells to bacterial toxins and further facilitate deep tissue penetration during infection [50,51]. It is also now clear that GBS uses HAase to counteract host immune responses [52]. Whereas a host can normally promptly respond by generating hyaluronan (HA) polymers, from which small fragments ultimately combine with Toll-like receptors (TLRs) to elicit inflammatory responses, the secreted HAase from GBS degrades proinflammatory HA fragments down into their component disaccharides, thus blocking the host's TLR2/4 signaling responses [52].

### 3.4. Cel-EIIB

The GBS phosphotransferase system (PTS) system, which is known to regulate bacterial virulence, can phosphorylate sugar substrates, including lactose, fructose, cellobiose, mannose, and sorbose [53]. Cellobiose-PTS (cel-PTS) is ubiquitously expressed in different serotypes of GBS, and strains genetically deficient for cel-PTS have decreased colonization ability and virulence [54]. One study showed that there are different expression levels of the cel-PTS component cel-EIIB between low and high virulence GBS [55,56]. Xu et al. (2018) reported that a cel-EIIB knockout strain showed a decreased ability of cellobiose utilization, as well as significantly reduced biofilm formation ability compared with the wild-type

GBS strain [57]. It was also notable that knockout of cel-EIIB caused a 20% decrease in the accumulative mortality of tilapia due to GBS infection; the authors speculated that cel-EIIB knockout significantly decreased invasion and colonization efficiency [57].

### 3.5. *LuxS/AI-2 Quorum Sensing System*

Quorum sensing (QS) refers to a coordinated mode of gene expression regulation that supports bacterial communication and group activity [58]. The *LuxS* gene encodes S-ribosyl homocysteinase, which catalyzes the biosynthesis of the QS signaling molecule known as autoinducer 2 (AI-2), a furanosyl borate diester. *LuxS* is conserved among GBS serotypes and is ubiquitously expressed [59,60]. This 483 bp gene is composed of a conserved active center (H57, H61, C127) and a Zn<sup>2+</sup>-binding site (H-T-I-E-H) [61]. Ma et al. have reported that a mutant strain deficient for *LuxS* was defective for quorum sensing and displayed a more than 30-fold reduction in acid resistance compared to the wild-type strain [61]. The cell adherence was also decreased in the mutant strain. A study in tilapia showed a significantly decreased extent of infection and demonstrated that reintroducing *LuxS* into the *luxS* rescued hypervirulence [61].

### 3.6. *Other Virulence Factors*

Besides the aforementioned virulence factors, there are also other virulence factors, which have not been experimentally confirmed in tilapia GBS infections, but, which have been widely studied in mammalian GBS infections, including, for example, fibrinogen receptor (*FbsA*, *FbsB* and *FbsC*) [62], superoxide dismutase (SOD), serine-threonine protein kinase (STPK), C5a peptidase (*scp*) [14], *serine-rich repeat glycoproteins* (*srr1* and *srr2*) [63],  $\beta$ -hemolysin/cytolysin (*cylE*) [64], *pili* [65], proteins C $\alpha$  (*bca*) [66], *neul* [67], and  $\alpha$ -like protein (*Alp*) [68], among others.

## 4. Progress Made in GBS Vaccinology

### 4.1. *Inactivated Whole-Cell Vaccine*

Formalin-inactivated and heat-killed whole-cell vaccines have been widely used in studies of pathogenic infection. Pasnik et al. (2005) conducted experiments wherein the relative percentage of survival (RPS) upon post-vaccination challenge was 49–50% [69,70]. Evans et al. (2004) performed similar assays using an inactivated whole-cell vaccination approach and challenge methods and recorded an RPS of 80% [71]. Thus, inactivated whole-cell approaches are useful for inducing immune responses to support basic studies and can confer protection against GBS infection (Table 1).

### 4.2. *SAGs (S. Agalactiae Ghosts)*

So-called *S. agalactiae* ghosts (SAGs) are empty cell envelopes from dead cells that have been explored as vaccine candidates owing to the presence of too many innate immunostimulatory agonists and their potent ability to activate innate and adaptive immunity responses. SAGs have been shown to induce cytokine secretion, which, in turn, contributes to the recruitment of T and B cells to lymph nodes. The recruitment of lymphocytes can increase the recognition of foreign antigens to elicit strong immune responses [72]. Wang et al. (2018) reported that immunization of tilapia with SAG elicited significantly higher resistance against GBS compared to the PBS-immunized controls [73] (Table 1). Moreover, SAG-immunized tilapia has stronger innate immunity (including phagocytic activity, lysozyme and superoxide dismutase activities) and adaptive immunity, especially IgM antibody titers. Finally, they found that the SAG-immunized tilapia showed significantly higher cytokine production (IL-1 $\beta$ , TNF- $\alpha$ , and TGF- $\beta$ ) than control animals.

### 4.3. *Sip (Surface Immunogenic Protein)*

There are some issues like inter-batch variability that can make use of inactivated whole-cell approaches less attractive than using vaccination approaches, which rely on more narrowly focused antigens (especially subunit vaccines) [74,75].

Sip is encoded by a gene in the GBS chromosome; this cell surface protein has been demonstrated as highly conserved; it is present in all GBS serotypes [76,77]. Given this broad distribution, it is unsurprising that Sip has been explored as a candidate for developing a subunit vaccine against GBS infection [78,79]. A 2014 study explored the immunogenicity of a Sip DNA vaccine, reporting an RPS value of 57% [79]; this was considered to represent relative effective immunoprotection. He et al. (2014) evaluated the truncated surface immunogenic protein (tSip) subunit vaccine and reported an RPS of 90% [78] (Table 1).

#### 4.4. Surface Antigen Metabolic Enzymes

There have been attempts to assess the immunogenicity of enzymes present at the GBS surface, including phosphoglycerate kinase (PGK) [80], ornithine carbamoyl-transferase (OCT) [80], and the GapA subunit of glyceraldehyde-3-phosphate dehydrogenase of GBS [75,80]. PGK is responsible for its virulence and immunoprotective functions [81]. Using the recombinant PGK protein, Wang et al. (2014) found that PGK (as a subunit vaccine) against GBS infection of tilapia gave an RPS value of 82.4% [80]. The same study investigated OCT, which catalyzes the production of citrulline and phosphate from ornithine and carbamoyl phosphate substrates. Using recombinant OCT protein as a vaccine subunit gave an RPS value of 58.8%. Another study examining GapA as a subunit vaccine showed an RPS value of 63.3% [75] (Table 1).

#### 4.5. Fibrinogen-Binding Protein A, $\alpha$ -Enolase, and GroEL

FbsA and enolase are adhesion proteins on the surface of bacteria. Yi et al. (2014) assessed potential immune protection against GBS infection upon vaccination with FbsA and Enolase, reporting RPS values of 40.63% and 62.50%, respectively [82]. The above data stated that the FbsA and enolase are multifaceted functions, including activating the host's innate immune responses and relevant antibody responses and immunoprotection function. Thus, both of them can be efficient subunit vaccine candidates (Table 1).

GroEL is a heat shock protein (Hsp) of the chaperonin family of molecular chaperones; it is ubiquitously expressed in many bacteria [83]. This highly conserved chaperone functions in the proper folding of proteins. Li et al. (2019) reported that immunization of tilapia with GroEL (delivered with FC or FIC adjuvants) conferred protection against GBS, with an RPS value of 68.61% [74]. At the same time, which can increase antibody titers by promoting lymphocyte proliferation [74,84] (Table 1).

**Table 1.** Inactivated whole-cell vaccines and subunit vaccines explored for controlling tilapia streptococcosis.

Vaccine	RPS	Adjuvant	Vaccination	Challenge	Year	Reference
Inactivated	49–80%	Without	Intraperitoneal	Intraperitoneal	2004–2005	[69–71]
Inactivated	97%	Without/feed-based adjuvant	Oral	Intraperitoneal	2004	[71]
Sip	57%	–	Intragastrical	Intraperitoneal	2014	[79]
tSip	90%	FIA	Intraperitoneal	Intraperitoneal	2014	[78]
PGK	82.4%	Montanide ISA 763 AVG	Intraperitoneal	Intraperitoneal	2014	[80]
OCT	58.8%	Montanide ISA 763 AVG	Intraperitoneal	Intraperitoneal	2014	[80]
GapA	63.3%	FCA + FIA	Intraperitoneal	Intraperitoneal	2016	[75]
GapA	45.6%	Montanide ISA 763 AVG	Intraperitoneal	Intraperitoneal	2016	[75]
FbsA	40.63%	Adjuvant	Intraperitoneal	Intraperitoneal	2014	[82]
Enolase	62.50%	Adjuvant	Intraperitoneal	Intraperitoneal	2014	[82]
LIC	70%	Without	Feed-based	Intraperitoneally	2014	[85]
GroEL	68.61%	FCA + FIA	Subcutaneous	Intraperitoneally	2019	[74]
CWSAP 465	77.5%	FIA	Intraperitoneal	Intraperitoneal	2016	[86]
CWSAP1035	72.5%	FIA	Intraperitoneal	Intraperitoneal	2016	[86]
ISP	48.61%	FCA + FIA	Intraperitoneal	Intraperitoneal	2016	[87]
SAG	86.67%	–	Intraperitoneal	Intraperitoneal	2018	[73]

Notes: Inactivated—inactivated whole-cell vaccine, RPS—relative percentage of survival, FIA—Freund's incomplete adjuvant, FCA—Freund's complete adjuvant,—DNA vaccine without adjuvant, without—without adjuvant, LIC—cell wall surface anchor family protein, CWSAP—cell wall surface anchor family protein, ISP—immunogenic secreted protein.



#### 4.6. Alternatives to Vaccines for Controlling Tilapia Streptococcosis

There has been research into other methods for managing, preventing, and treating tilapia streptococcosis, including intestinal microbiota modifier [88,89] and bacteriophages. Probiotics investigated to date include *Bacillus subtilis*, *Bacillus licheniformis*, and *Lactobacillus rhamnosus*. Liu et al. (2017) [90] reported that *Bacillus subtilis* could enhance tilapia growth, digestive enzyme activities, innate immune responses, and GBS resistance. Abarike et al. (2018) [91] reported that *Bacillus licheniformis* could also promote tilapia growth, immune responses, and GBS resistance. Xia et al. (2018) [92] showed supplementation with *Lactobacillus rhamnosus* increased tilapia growth, intestinal microbiota, immune responses, and GBS resistance. As for bacteriophages, Luo et al. (2018) [93] showed that tilapia treated with the bacteriophage HN48 had about 60% greater survival than control animals. Notably, there have been additional proposals about how to control GBS infections and tilapia streptococcosis, including, for example, the use of Chinese medicinal herbs [94] and alteration of water temperature [65,95], among others.

#### 5. Conclusions and Prospects

This review has considered research progress into tilapia streptococcosis regarding etiology, pathology, epidemiology, and prospects for control, including immunology-based approaches. As genomics, transcriptomics, proteomics, metabonomics, and bioinformatics continue to mature, it is clear that ever-more-powerful tools can be applied to identify and better understand the biological basis and control prospects for GBS infections of many animals generally and of tilapia streptococcosis in particular [5,12,96]. At the same time, an increasing number of vaccine candidates and adjuvant (including Freund's adjuvant, Montanide adjuvant, aluminum-based adjuvants and others) are being explored, and this is a promising area for control of disease outbreaks in aquaculture. There remain virulence factors of GBS isolated from tilapia, which are still not fully understood, so there is a knowledge gap for active research teams to fill. In the near future, it seems likely that antibiotic treatment will remain the main control approach; however, in the long run, as scientific research prompts breakthroughs in understanding and developing effective technologies, there may be some innovative treatments and control measures like vaccines plus probiotic and immune enhancers.

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#### References

1. Suanyuk, N.; Kong, F.; Ko, D.; Gilbert, G.L.; Supamattaya, K. Occurrence of rare genotypes of *Streptococcus agalactiae* in cultured red tilapia *Oreochromis sp.* and Nile tilapia *O. niloticus* in Thailand—Relationship to human isolates? *Aquaculture* **2008**, *284*, 35–40. [[CrossRef](#)]
2. Wang, E.L.; Wang, K.Y.; Chen, D.F.; Geng, Y.; Huang, L.Y.; Wang, J.; He, Y. Molecular cloning and bioinformatic analysis of the *Streptococcus agalactiae* neuA gene isolated from tilapia. *Genet. Mol. Res. GMR* **2015**, *14*, 6003–6017. [[CrossRef](#)]
3. Wang, R.; Li, L.; Huang, Y.; Luo, F.; Liang, W.; Gan, X.; Huang, T.; Lei, A.; Chen, M.; Chen, L. Comparative genome analysis identifies two large deletions in the genome of highly-passaged attenuated *Streptococcus agalactiae* strain YM001 compared to the parental pathogenic strain HN016. *BMC Genom.* **2015**, *16*, 897. [[CrossRef](#)]
4. Chen, M.; Li, L.P.; Wang, R.; Liang, W.W.; Huang, Y.; Li, J.; Lei, A.Y.; Huang, W.Y.; Gan, X. PCR detection and PFGE genotype analyses of streptococcal clinical isolates from tilapia in China. *Vet. Microbiol.* **2012**, *159*, 526–530. [[CrossRef](#)] [[PubMed](#)]
5. Zhang, Z.; Yu, A.; Lan, J.; Zhang, Y.; Zhang, H.; Li, Y.; Hu, M.; Cheng, J.; Wei, S.; Lin, L. Draft Genome Sequence of an Attenuated *Streptococcus agalactiae* Strain Isolated from the Gut of a Nile Tilapia (*Oreochromis niloticus*). *Genome Announc.* **2017**, *5*, e01627-16. [[CrossRef](#)]

6. Surve, M.V.; Anil, A.; Kamath, K.G.; Bhutda, S.; Sthanam, L.K.; Pradhan, A.; Srivastava, R.; Basu, B.; Dutta, S.; Sen, S. Membrane Vesicles of Group B Streptococcus Disrupt Feto-Maternal Barrier Leading to Preterm Birth. *PLoS Pathog.* **2016**, *12*, e1005816. [[CrossRef](#)] [[PubMed](#)]
7. Armistead, B.; Oler, E.; Adams Waldorf, K.; Rajagopal, L. The Double Life of Group B Streptococcus: Asymptomatic Colonizer and Potent Pathogen. *J. Mol. Biol.* **2019**, *431*, 2914–2931. [[CrossRef](#)] [[PubMed](#)]
8. Vornhagen, J.; Adams Waldorf, K.M.; Rajagopal, L. Perinatal Group B Streptococcal Infections: Virulence Factors, Immunity, and Prevention Strategies. *Trends Microbiol.* **2017**, *25*, 919–931. [[CrossRef](#)]
9. Rajagopal, L. Understanding the regulation of Group B Streptococcal virulence factors. *Futur. Microbiol.* **2009**, *4*, 201–221. [[CrossRef](#)] [[PubMed](#)]
10. Andrade, W.A.; Firon, A.; Schmidt, T.; Hornung, V.; Fitzgerald, K.A.; Kurt-Jones, E.A.; Trieu-Cuot, P.; Golenbock, D.T.; Kaminski, P.A. Group B Streptococcus Degrades Cyclic-di-AMP to Modulate STING-Dependent Type I Interferon Production. *Cell Host Microbe* **2016**, *20*, 49–59. [[CrossRef](#)] [[PubMed](#)]
11. Zhang, Z.; Lan, J.; Li, Y.; Hu, M.; Yu, A.; Zhang, J.; Wei, S. The pathogenic and antimicrobial characteristics of an emerging Streptococcus agalactiae serotype IX in Tilapia. *Microb. Pathog.* **2018**, *122*, 39–45. [[CrossRef](#)]
12. Zhang, Z.; Li, Y.; Hu, M.; Yu, A. Comparative transcriptome profiling reveals a mechanism of Streptococcus agalactiae resistance to florfenicol. *Microb. Pathog.* **2020**, *142*, 104098. [[CrossRef](#)]
13. Wei, S.; Zhang, Z.; Li, Y.; Hu, M.; Yu, A.; Zhang, H.; Lan, J.; Zhang, Z.; Yan, Y.; Lin, L. Epidemic and antibiotic resistance of Streptococcus agalactiae isolated from tilapia (*GIFT Oreochromis niloticus*) in Guangdong Province. *J. Fish. China* **2016**, *40*, 503–511.
14. Mishra, A.; Nam, G.H.; Gim, J.A.; Lee, H.E.; Jo, A.; Kim, H.S. Current Challenges of Streptococcus Infection and Effective Molecular, Cellular, and Environmental Control Methods in Aquaculture. *Mol. Cells* **2018**, *41*, 495–505.
15. Laith, A.A.; Ambak, M.A.; Hassan, M.; Sheriff, S.M.; Nadirah, M.; Draman, A.S.; Wahab, W.; Ibrahim, W.N.; Aznan, A.S.; Jabar, A.; et al. Molecular identification and histopathological study of natural Streptococcus agalactiae infection in hybrid tilapia (*Oreochromis niloticus*). *Vet. World* **2017**, *10*, 101–111. [[PubMed](#)]
16. Phares, C.R.; Lynfield, R.; Farley, M.M.; Mohle-Boetani, J.; Harrison, L.H.; Petit, S.; Craig, A.S.; Schaffner, W.; Zansky, S.M.; Gershman, K.; et al. Active Bacterial Core surveillance/Emerging Infections Program, N. Epidemiology of invasive group B streptococcal disease in the United States, 1999–2005. *JAMA* **2008**, *299*, 2056–2065. [[CrossRef](#)] [[PubMed](#)]
17. Schuchat, A. Epidemiology of group B streptococcal disease in the United States: Shifting paradigms. *Clin. Microbiol. Rev.* **1998**, *11*, 497–513. [[CrossRef](#)] [[PubMed](#)]
18. Lancefield, R.C. A Serological Differentiation of Specific Types of Bovine Hemolytic Streptococci (Group B). *J. Exp. Med.* **1934**, *59*, 441–458. [[CrossRef](#)]
19. Slotved, H.C.; Kong, F.; Lambertsen, L.; Sauer, S.; Gilbert, G.L. Serotype IX, a Proposed New Streptococcus agalactiae Serotype. *J. Clin. Microbiol.* **2007**, *45*, 2929–2936. [[CrossRef](#)] [[PubMed](#)]
20. Gherardi, G.; Imperi, M.; Baldassarri, L.; Pataracchia, M.; Alfarone, G.; Recchia, S.; Orefici, G.; Dicuonzo, G.; Creti, R. Molecular epidemiology and distribution of serotypes, surface proteins, and antibiotic resistance among group B streptococci in Italy. *J. Clin. Microbiol.* **2007**, *45*, 2909–2916. [[CrossRef](#)] [[PubMed](#)]
21. Amal, M.N.; Zamri-Saad, M.; Iftikhar, A.R.; Siti-Zahrah, A.; Aziel, S.; Fahmi, S. An outbreak of Streptococcus agalactiae infection in cage-cultured golden pompano, *Trachinotus blochii* (Lacepede), in Malaysia. *J. Fish Dis.* **2012**, *35*, 849–852. [[CrossRef](#)] [[PubMed](#)]
22. Bowater, R.O.; Forbes-Faulkner, J.; Anderson, I.G.; Condon, K.; Robinson, B.; Kong, F.; Gilbert, G.L.; Reynolds, A.; Hyland, S.; McPherson, G.; et al. Natural outbreak of Streptococcus agalactiae (GBS) infection in wild giant Queensland grouper, *Epinephelus lanceolatus* (Bloch), and other wild fish in northern Queensland, Australia. *J. Fish Dis.* **2012**, *35*, 173–186. [[CrossRef](#)] [[PubMed](#)]
23. Pereira Ude, P.; Dos Santos, A.R.; Hassan, S.S.; Aburjaile, F.F.; Soares Sde, C.; Ramos, R.T.; Carneiro, A.R.; Guimaraes, L.C.; Silva de Almeida, S.; Diniz, C.A.; et al. Complete genome sequence of Streptococcus agalactiae strain SA20-06, a fish pathogen associated to meningoencephalitis outbreaks. *Stand. Genom. Sci.* **2013**, *8*, 188–197. [[CrossRef](#)] [[PubMed](#)]
24. Hernandez, E.; Figueroa, J.; Iregui, C. Streptococcosis on a red tilapia, *Oreochromis* sp. farm: A case study. *J. Fish Dis.* **2009**, *32*, 247–252. [[CrossRef](#)] [[PubMed](#)]
25. Suwannasang, A.; Suanyuk, N.; Tantikitti, C. Phenotypic and genotypic characterization of Streptococcus agalactiae isolates from cultured tilapia in Thailand. *Fish Shellfish Immunol.* **2013**, *34*, 243–249. [[CrossRef](#)]
26. Pradeep, P.J.; Suebsing, R.; Sirthammajak, S.; Kampeera, J.; Jitrakorn, S.; Saksmerprom, V.; Turner, W.; Palang, I.; Vanichviriyakit, R.; Senapin, S. Evidence of vertical transmission and tissue tropism of Streptococcosis from naturally infected red tilapia (*Oreochromis* spp.). *Aquac. Rep.* **2016**, *3*, 58–66. [[CrossRef](#)]
27. Amal, M.N.A.; Zamrisaad, M. Streptococcosis in tilapia (*Oreochromis niloticus*): A review. *Pertanika J. Trop. Agric. Sci.* **2011**, *2*, 195–206.
28. Wessels, M.R.; Benedi, W.J.; Jennings, H.J.; Michon, F.; DiFabio, J.L.; Kasper, D.L. Isolation and characterization of type IV group B Streptococcus capsular polysaccharide. *Infect. Immun.* **1989**, *57*, 1089–1094. [[CrossRef](#)] [[PubMed](#)]
29. Dogan, B.; Schukken, Y.H.; Santisteban, C.; Boor, K.J. Distribution of serotypes and antimicrobial resistance genes among Streptococcus agalactiae isolates from bovine and human hosts. *J. Clin. Microbiol.* **2006**, *43*, 5899–5906. [[CrossRef](#)] [[PubMed](#)]

30. Zhao, Z.; Kong, F.X.; Gidding, H.; Morgan, J.; Gilbert, G. Distribution of genotypes and antibiotic resistance genes among invasive *Streptococcus agalactiae* (group B streptococcus) isolates from Australasian patients belonging to different age groups. *Clin. Microbiol. Infect.* **2008**, *14*, 260–267. [[CrossRef](#)] [[PubMed](#)]
31. Kong, F.; Ma, L.; Gilbert, G.L. Simultaneous detection and serotype identification of *Streptococcus agalactiae* using multiplex PCR and reverse line blot hybridization. *J. Med. Microbiol.* **2005**, *54 Pt 12*, 1133–1138. [[CrossRef](#)]
32. Rubens, C.E.; Wessels, M.R.; Heggen, L.M.; Kasper, D.L. Transposon mutagenesis of type III group B *Streptococcus*: Correlation of capsule expression with virulence. *Proc. Natl. Acad. Sci. USA* **1987**, *84*, 7208–7212. [[CrossRef](#)]
33. Wessels, M.R.; Rubens, C.E.; Benedi, V.J.; Kasper, D.L. Definition of a bacterial virulence factor: Sialylation of the group B streptococcal capsule. *Proc. Natl. Acad. Sci. USA* **1989**, *86*, 8983–8987. [[CrossRef](#)] [[PubMed](#)]
34. Zhang, D.; Ke, X.; Liu, Z.; Cao, J.; Su, Y.; Lu, M.; Gao, F.; Wang, M.; Yi, M.; Qin, F. Capsular polysaccharide of *Streptococcus agalactiae* is an essential virulence factor for infection in Nile tilapia (*Oreochromis niloticus* Linn.). *J. Fish Dis.* **2019**, *42*, 293–302. [[CrossRef](#)] [[PubMed](#)]
35. Marques, M.B.; Kasper, D.L.; Pangburn, M.K.; Wessels, M.R. Prevention of C3 deposition by capsular polysaccharide is a virulence mechanism of type III group B streptococci. *Infect. Immun.* **1992**, *60*, 3986–3993. [[CrossRef](#)] [[PubMed](#)]
36. Campbell, J.R.; Baker, C.J.; Edwards, M.S. Deposition and degradation of C3 on type III group B streptococci. *Infect. Immun.* **1991**, *59*, 1978–1983. [[CrossRef](#)]
37. Lemire, P.; Houde, M.; Lecours, M.P.; Fittipaldi, N.; Segura, M. Role of capsular polysaccharide in Group B *Streptococcus* interactions with dendritic cells. *Microbes Infect.* **2012**, *14*, 1064–1076. [[CrossRef](#)] [[PubMed](#)]
38. Nizet, V.; Kim, K.S.; Stins, M.; Jonas, M.; Chi, E.Y.; Nguyen, D.; Rubens, C.E. Invasion of brain microvascular endothelial cells by group B streptococci. *Infect. Immun.* **1997**, *65*, 5074–5081. [[CrossRef](#)] [[PubMed](#)]
39. Barato, P.; Martins, E.R.; Vasquez, G.M.; Ramirez, M.; Melo-Cristino, J.; Martinez, N.; Iregui, C. Capsule impairs efficient adherence of *Streptococcus agalactiae* to intestinal epithelium in tilapias *Oreochromis* sp. *Microb. Pathog.* **2016**, *100*, 30–36. [[CrossRef](#)] [[PubMed](#)]
40. Bernheimer, A.W.; Linder, R.; Avigad, L.S. Nature and mechanism of action of the CAMP protein of group B streptococci. *Infect. Immun.* **1979**, *23*, 838–844. [[CrossRef](#)]
41. Lang, S.; Palmer, M. Characterization of *Streptococcus agalactiae* CAMP factor as a pore-forming toxin. *J. Biol. Chem.* **2003**, *278*, 38167–38173. [[CrossRef](#)]
42. Podbielski, A.; Blankenstein, O.; Luticken, R. Molecular characterization of the *cfb* gene encoding group B streptococcal CAMP-factor. *Med Microbiol. Immunol.* **1994**, *183*, 239–256. [[CrossRef](#)]
43. Jurgens, D.; Sterzik, B.; Fehrenbach, F.J. Unspecific binding of group B streptococcal cocytolysin (CAMP factor) to immunoglobulins and its possible role in pathogenicity. *J. Exp. Med.* **1987**, *165*, 720–732. [[CrossRef](#)]
44. Skalka, B.; Smola, J. Lethal effect of CAMP-factor and UBERIS-factor—A new finding about diffusible exosubstances of streptococcus *agalactiae* and *Streptococcus uberis*. *Zent. fur Bakteriol. 1 Abt. Orig. A Med. Mikrobiol. Infekt. und Parasitol.* **1981**, *249*, 190–194. [[CrossRef](#)]
45. Hensler, M.E.; Quach, D.; Hsieh, C.J.; Doran, K.S.; Nizet, V. CAMP factor is not essential for systemic virulence of Group B *Streptococcus*. *Microb. Pathog.* **2008**, *44*, 84–88. [[CrossRef](#)]
46. Jin, T.; Brefo-Mensah, E.; Fan, W.; Zeng, W.; Li, Y.; Zhang, Y.; Palmer, M. Crystal structure of the *Streptococcus agalactiae* CAMP factor provides insights into its membrane-permeabilizing activity. *J. Biol. Chem.* **2018**, *293*, 11867–11877. [[CrossRef](#)] [[PubMed](#)]
47. Li, Y.; Zeng, W.; Li, Y.; Fan, W.; Ma, H.; Fan, X.; Jiang, J.; Brefo-Mensah, E.; Zhang, Y.; Yang, M.; et al. Structure determination of the CAMP factor of *Streptococcus agalactiae* with the aid of an MBP tag and insights into membrane-surface attachment. *Acta Crystallogr. D Struct. Biol.* **2019**, *75 Pt 8*, 772–781. [[CrossRef](#)]
48. Gase, K.; Ferretti, J.J.; Primeaux, C.; McShan, W.M. Identification, cloning, and expression of the CAMP factor gene (*cfa*) of group A streptococci. *Infect. Immun.* **1999**, *67*, 4725–4731. [[CrossRef](#)] [[PubMed](#)]
49. Sorensen, M.; Mak, T.N.; Hurwitz, R.; Ogilvie, L.A.; Mollenkopf, H.J.; Meyer, T.F.; Bruggemann, H. Mutagenesis of *Propionibacterium acnes* and analysis of two CAMP factor knock-out mutants. *J. Microbiol. Methods* **2010**, *83*, 211–216. [[CrossRef](#)] [[PubMed](#)]
50. Li, S.; Kelly, S.J.; Lamani, E.; Ferraroni, M.; Jedrzejak, M.J. Structural basis of hyaluronan degradation by *Streptococcus pneumoniae* hyaluronate lyase. *EMBO J.* **2000**, *19*, 1228–1240. [[CrossRef](#)]
51. Mello, L.V.; De Groot, B.L.; Li, S.; Jedrzejak, M.J. Structure and flexibility of *Streptococcus agalactiae* hyaluronate lyase complex with its substrate. Insights into the mechanism of processive degradation of hyaluronan. *J. Biol. Chem.* **2002**, *277*, 36678–36688. [[CrossRef](#)] [[PubMed](#)]
52. Kolar, S.L.; Kyme, P.; Tseng, C.W.; Soliman, A.; Kaplan, A.; Liang, J.; Nizet, V.; Jiang, D.; Murali, R.; Ardit, M.; et al. Group B *Streptococcus* Evades Host Immunity by Degrading Hyaluronan. *Cell Host Microbe* **2015**, *18*, 694–704. [[CrossRef](#)] [[PubMed](#)]
53. Sutcliffe, I.C.; Black, G.W.; Harrington, D.J. Bioinformatic insights into the biosynthesis of the Group B carbohydrate in *Streptococcus agalactiae*. *Microbiology* **2008**, *154 Pt 5*, 1354–1363. [[CrossRef](#)]
54. McAllister, L.J.; Ogunniyi, A.D.; Stroehrer, U.H.; Paton, J.C. Contribution of a genomic accessory region encoding a putative cellobiose phosphotransferase system to virulence of *Streptococcus pneumoniae*. *PLoS ONE* **2012**, *7*, e32385.
55. Li, W.; Su, Y.L.; Mai, Y.Z.; Li, Y.W.; Mo, Z.Q.; Li, A.X. Comparative proteome analysis of two *Streptococcus agalactiae* strains from cultured tilapia with different virulence. *Vet. Microbiol.* **2014**, *170*, 135–143. [[CrossRef](#)]



56. Su, Y.L.; Feng, J.; Liu, C.; Li, W.; Xie, Y.D.; Li, A.X. Dynamic bacterial colonization and microscopic lesions in multiple organs of tilapia infected with low and high pathogenic *Streptococcus agalactiae* strains. *Aquaculture* **2017**, *471*, 190–203. [[CrossRef](#)]
57. Xu, J.; Xie, Y.D.; Liu, L.; Guo, S.; Su, Y.L.; Li, A.X. Virulence regulation of cel-EIIB protein mediated PTS system in *Streptococcus agalactiae* in Nile tilapia. *J. Fish Dis.* **2019**, *42*, 11–19. [[CrossRef](#)] [[PubMed](#)]
58. Miller, M.B.; Bassler, B.L. Quorum sensing in bacteria. *Annu. Rev. Microbiol.* **2001**, *55*, 165–199. [[CrossRef](#)]
59. Wang, Y.; Zhang, W.; Wu, Z.; Zhu, X.; Lu, C. Functional analysis of luxS in *Streptococcus suis* reveals a key role in biofilm formation and virulence. *Vet. Microbiol.* **2011**, *152*, 151–160. [[CrossRef](#)]
60. Zhao, L.; Xue, T.; Shang, F.; Sun, H.; Sun, B. *Staphylococcus aureus* AI-2 quorum sensing associates with the KdpDE two-component system to regulate capsular polysaccharide synthesis and virulence. *Infect. Immun.* **2010**, *78*, 3506–3515. [[CrossRef](#)]
61. Ma, Y.; Hao, L.; Ke, H.; Liang, Z.; Ma, J.; Liu, Z.; Li, Y. LuxS/AI-2 in *Streptococcus agalactiae* reveals a key role in acid tolerance and virulence. *Res. Vet. Sci.* **2017**, *115*, 501–507. [[CrossRef](#)] [[PubMed](#)]
62. Liu, Y.; Li, L.; Huang, T.; Wu, W.; Liang, W.; Chen, M. The Interaction between Phagocytes and *Streptococcus agalactiae* (GBS) Mediated by the Activated Complement System is the Key to GBS Inducing Acute Bacterial Meningitis of Tilapia. *Animals* **2019**, *9*, 818. [[CrossRef](#)] [[PubMed](#)]
63. Su, Y.; Liu, C.; Deng, Y.; Cheng, C.; Ma, H.; Guo, Z.; Feng, J. Molecular typing of *Streptococcus agalactiae* isolates of serotype Ia from tilapia in southern China. *FEMS Microbiol. Lett.* **2019**, *366*, 13. [[CrossRef](#)] [[PubMed](#)]
64. Kayansamruaj, P.; Pirarat, N.; Katagiri, T.; Hirono, I.; Rodkhum, C. Molecular characterization and virulence gene profiling of pathogenic *Streptococcus agalactiae* populations from tilapia (*Oreochromis sp.*) farms in Thailand. *J. Vet. Diagn. Invest.* **2014**, *26*, 488–495. [[CrossRef](#)]
65. Kayansamruaj, P.; Pirarat, N.; Hirono, I.; Rodkhum, C. Increasing of temperature induces pathogenicity of *Streptococcus agalactiae* and the up-regulation of inflammatory related genes in infected Nile tilapia (*Oreochromis niloticus*). *Vet. Microbiol.* **2014**, *172*, 265–271. [[CrossRef](#)]
66. Wu, W.; Li, L.; Liu, Y.; Huang, T.; Liang, W.; Chen, M. Multiomics analyses reveal that NOD-like signaling pathway plays an important role against *Streptococcus agalactiae* in the spleen of tilapia. *Fish Shellfish Immunol.* **2019**, *95*, 336–348. [[CrossRef](#)]
67. Yamaguchi, M.; Hirose, Y.; Nakata, M.; Uchiyama, S.; Yamaguchi, Y.; Goto, K.; Sumitomo, T.; Lewis, A.L.; Kawabata, S.; Nizet, V. Evolutionary inactivation of a sialidase in group B *Streptococcus*. *Sci. Rep.* **2016**, *6*, 28852. [[CrossRef](#)]
68. Jalalifar, S.; Havaei, S.A.; Motallebirad, T.; Moghim, S.; Fazeli, H.; Esfahani, B.N. Determination of surface proteins profile, capsular genotyping, and antibiotic susceptibility patterns of Group B *Streptococcus* isolated from urinary tract infection of Iranian patients. *BMC Res. Notes* **2019**, *12*, 437. [[CrossRef](#)]
69. Pasnik, D.J.; Evans, J.J.; Klesius, P.H. Duration of protective antibodies and correlation with survival in Nile tilapia *Oreochromis niloticus* following *Streptococcus agalactiae* vaccination. *Dis. Aquat. Org.* **2005**, *66*, 129–134. [[CrossRef](#)]
70. Pasnik, D.J.; Evans, J.J.; Panangala, V.S.; Klesius, P.H.; Shelby, R.A.; Shoemaker, C.A. Antigenicity of *Streptococcus agalactiae* extracellular products and vaccine efficacy. *J. Fish Dis.* **2005**, *28*, 205–212. [[CrossRef](#)]
71. Evans, J.J.; Klesius, P.H.; Shoemaker, C.A. Efficacy of *Streptococcus agalactiae* (group B) vaccine in tilapia (*Oreochromis niloticus*) by intraperitoneal and bath immersion administration. *Vaccine* **2004**, *22*, 3769–3773. [[CrossRef](#)] [[PubMed](#)]
72. Hajam, I.A.; Dar, P.A.; Won, G.; Lee, J.H. Bacterial ghosts as adjuvants: Mechanisms and potential. *Vet. Res.* **2017**, *48*, 37. [[CrossRef](#)] [[PubMed](#)]
73. Wang, Q.; Wang, X.; Wang, X.; Feng, R.; Luo, Q.; Huang, J. Generation of a novel *Streptococcus agalactiae* ghost vaccine and examination of its immunogenicity against virulent challenge in tilapia. *Fish Shellfish Immunol.* **2018**, *81*, 49–56. [[CrossRef](#)] [[PubMed](#)]
74. Li, W.; Li, Y.; Hu, Y.Z.; Mo, X.B.; Xu, G.H.; Xie, L.W.; Li, A.X. GroEL, a novel vaccine candidate of piscine *Streptococcus agalactiae* identified by immunoproteome. *Fish Shellfish Immunol.* **2019**, *84*, 377–383. [[CrossRef](#)] [[PubMed](#)]
75. Zhang, Z.; Yu, A.; Lan, J.; Zhang, H.; Hu, M.; Cheng, J.; Zhao, L.; Lin, L.; Wei, S. GapA, a potential vaccine candidate antigen against *Streptococcus agalactiae* in Nile tilapia (*Oreochromis niloticus*). *Fish Shellfish Immunol.* **2017**, *63*, 255–260. [[CrossRef](#)] [[PubMed](#)]
76. Rioux, S.; Martin, D.; Ackermann, H.W.; Dumont, J.; Hamel, J.; Brodeur, B.R. Localization of surface immunogenic protein on group B streptococcus. *Infect. Immun.* **2001**, *69*, 5162–5165. [[CrossRef](#)]
77. Xue, G.; Yu, L.; Li, S.; Shen, X. Intranasal immunization with GBS surface protein Sip and ScpB induces specific mucosal and systemic immune responses in mice. *FEMS Immunol. Med. Microbiol.* **2010**, *58*, 202–210. [[CrossRef](#)]
78. He, Y.; Wang, K.Y.; Xiao, D.; Chen, D.F.; Huang, L.; Liu, T.; Wang, J.; Geng, Y.; Wang, E.L.; Yang, Q. A recombinant truncated surface immunogenic protein (tSip) plus adjuvant FIA confers active protection against Group B streptococcus infection in tilapia. *Vaccine* **2014**, *32*, 7025–7032. [[CrossRef](#)] [[PubMed](#)]
79. Huang, L.Y.; Wang, K.Y.; Xiao, D.; Chen, D.F.; Geng, Y.; Wang, J.; He, Y.; Wang, E.L.; Huang, J.L.; Xiao, G.Y. Safety and immunogenicity of an oral DNA vaccine encoding Sip of *Streptococcus agalactiae* from Nile tilapia *Oreochromis niloticus* delivered by live attenuated *Salmonella typhimurium*. *Fish Shellfish Immunol.* **2014**, *38*, 34–41. [[CrossRef](#)] [[PubMed](#)]
80. Wang, Y.T.; Huang, H.Y.; Tsai, M.A.; Wang, P.C.; Jiang, B.H.; Chen, S.C. Phosphoglycerate kinase enhanced immunity of the whole cell of *Streptococcus agalactiae* in tilapia, *Oreochromis niloticus*. *Fish Shellfish Immunol.* **2014**, *41*, 250–259. [[CrossRef](#)]
81. Lee, K.W.; Shalaby, K.A.; Thakur, A.; Medhat, A.M.; Karim, A.M.; LoVerde, P.T. Cloning of the gene for phosphoglycerate kinase from *Schistosoma mansoni* and characterization of its gene product. *Mol. Biochem. Parasitol.* **1995**, *71*, 221–231. [[CrossRef](#)]

82. Yi, T.; Li, Y.W.; Liu, L.; Xiao, X.X.; Li, A.X. Protection of Nile tilapia (*Oreochromis niloticus* L.) against *Streptococcus agalactiae* following immunization with recombinant FbsA and  $\alpha$ -enolase. *Aquaculture* **2014**, *428–429*, 35–40. [[CrossRef](#)]
83. Zeilstra-Ryalls, J.; Fayet, O.; Georgopoulos, C. The universally conserved GroE (Hsp60) chaperonins. *Annu. Rev. Microbiol.* **1991**, *45*, 301–325. [[CrossRef](#)] [[PubMed](#)]
84. Yi, L.; Wang, Y.; Ma, Z.; Lin, H.X.; Xu, B.; Grenier, D.; Fan, H.J.; Lu, C.P. Identification and characterization of a *Streptococcus equi* ssp. *zooepidemicus* immunogenic GroEL protein involved in biofilm formation. *Vet. Res.* **2016**, *47*, 50. [[CrossRef](#)]
85. Nur-Nazifah, M.; Sabri, M.Y.; Siti-Zahrah, A. Development and efficacy of feed-based recombinant vaccine encoding the cell wall surface anchor family protein of *Streptococcus agalactiae* against streptococcosis in *Oreochromis* sp. *Fish Shellfish Immunol.* **2014**, *37*, 193–200. [[CrossRef](#)] [[PubMed](#)]
86. Liu, H.; Zhang, S.; Shen, Z.; Ren, G.; Liu, L.; Ma, Y.; Zhang, Y.; Wang, W. Development of a vaccine against *Streptococcus agalactiae* in fish based on truncated cell wall surface anchor proteins. *Vet. Rec.* **2016**, *179*, 359. [[CrossRef](#)] [[PubMed](#)]
87. Li, W.; Wang, H.Q.; He, R.Z.; Li, Y.W.; Su, Y.L.; Li, A.X. Major surfome and secretome profile of *Streptococcus agalactiae* from Nile tilapia (*Oreochromis niloticus*): Insight into vaccine development. *Fish Shellfish Immunol.* **2016**, *55*, 737–746. [[CrossRef](#)] [[PubMed](#)]
88. Opiyo, M.A.; Jumbe, J.; Ngugi, C.C.; Charo-Karisa, H. Dietary administration of probiotics modulates non-specific immunity and gut microbiota of Nile tilapia (*Oreochromis niloticus*) cultured in low input ponds. *Int. J. Vet. Sci. Med.* **2019**, *7*, 1–9. [[CrossRef](#)]
89. Xia, Y.; Wang, M.; Gao, F.; Lu, M.; Chen, G. Effects of dietary probiotic supplementation on the growth, gut health and disease resistance of juvenile Nile tilapia (*Oreochromis niloticus*). *Anim. Nutr.* **2020**, *6*, 69–79. [[CrossRef](#)] [[PubMed](#)]
90. Liu, H.; Wang, S.; Cai, Y.; Guo, X.; Cao, Z.; Zhang, Y.; Liu, S.; Yuan, W.; Zhu, W.; Zheng, Y.; et al. Dietary administration of *Bacillus subtilis* HAINUP40 enhances growth, digestive enzyme activities, innate immune responses and disease resistance of tilapia, *Oreochromis niloticus*. *Fish Shellfish Immunol.* **2017**, *60*, 326–333. [[CrossRef](#)] [[PubMed](#)]
91. Abarike, E.D.; Cai, J.; Lu, Y.; Yu, H.; Chen, L.; Jian, J.; Tang, J.; Jun, L.; Kuebutornye, F.K.A. Effects of a commercial probiotic BS containing *Bacillus subtilis* and *Bacillus licheniformis* on growth, immune response and disease resistance in Nile tilapia, *Oreochromis niloticus*. *Fish Shellfish Immunol.* **2018**, *82*, 229–238. [[CrossRef](#)] [[PubMed](#)]
92. Xia, Y.; Lu, M.; Chen, G.; Cao, J.; Gao, F.; Wang, M.; Liu, Z.; Zhang, D.; Zhu, H.; Yi, M. Effects of dietary *Lactobacillus rhamnosus* JCM1136 and *Lactococcus lactis* subsp. *lactis* JCM5805 on the growth, intestinal microbiota, morphology, immune response and disease resistance of juvenile Nile tilapia, *Oreochromis niloticus*. *Fish Shellfish Immunol.* **2018**, *76*, 368–379. [[CrossRef](#)] [[PubMed](#)]
93. Luo, X.; Liao, G.; Liu, C.; Jiang, X.; Lin, M.; Zhao, C.; Tao, J.; Huang, Z. Characterization of bacteriophage HN48 and its protective effects in Nile tilapia *Oreochromis niloticus* against *Streptococcus agalactiae* infections. *J. Fish Dis.* **2018**, *41*, 1477–1484. [[CrossRef](#)]
94. Abarike, E.D.; Jian, J.; Tang, J.; Cai, J.; Yu, H.; Chen, L. Traditional Chinese Medicine Enhances Growth, Immune Response, and Resistance to *Streptococcus agalactiae* in Nile Tilapia. *J. Aquat. Anim. Health* **2019**, *31*, 46–55. [[CrossRef](#)]
95. Tavares, G.C.; Carvalho, A.F.; Pereira, F.L.; Rezende, C.P.; Azevedo, V.A.C.; Leal, C.A.G.; Figueiredo, H.C.P. Transcriptome and Proteome of Fish-Pathogenic *Streptococcus agalactiae* Are Modulated by Temperature. *Front. Microbiol.* **2018**, *9*, 2639. [[CrossRef](#)] [[PubMed](#)]
96. Zhang, Z.; Li, Y.; Hu, M.; Yu, A. Genome reanalysis to decipher resistome, virulome, and attenuated characters of attenuated *Streptococcus agalactiae* strain HZAUSC001. *Microb. Pathog.* **2020**, *147*, 104416. [[CrossRef](#)]