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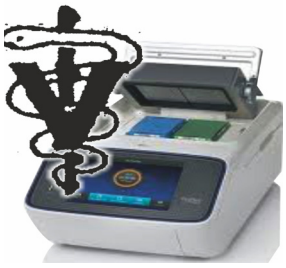

# A Review of *Streptococcus agalactiae* and its Antimicrobial Resistance Potentials in Nigerian Tilapia Fish Production

Adetona Mutiat Adenike[1], Akorede Farhan Rhidor[2,3], Lawal Zainab Damilola[4], Omotoso Rodiat Olabisi[5], Ghali Muhammad Ibraheem[1] and Odetokun Ismail Ayoade[1]

[1]Department of Public Health and Preventive Medicine, Faculty of Veterinary Medicine, University of Ilorin, Nigeria, P.M.B 1515, Ilorin, Kwara State, Nigeria

[2]Department of Veterinary Pharmacology and Toxicology, Faculty of Veterinary Medicine, Usman Danfodio University, Sokoto, P.M.B. 2346, Sokoto, Sokoto State, Nigeria

[3]Afana Animal Futures, Zaria, Kaduna, Nigeria

[4] Department of Theriogenology and Production, Faculty of Veterinary Medicine, University of Ilorin, P.M.B 1515, Ilorin, Kwara State, Nigeria

[5]Department of Statistics, Faculty of Physical sciences, University of Ilorin, P.M.B 1515, Ilorin, Kwara State, Nigeria

Correspondence: madetona9@gmail.com, **Phone: +2348168823157**

## ABSTRACT

Antimicrobial resistance (AMR) is a significant global threat to public health, undermining the stability of aquaculture, food security, and human health. Annually, bacterial AMR is directly responsible for over one million deaths and associated with more than 3 million deaths globally, especially in Sub-Saharan Africa. In Nigeria, aquaculture production is a major contributor to food security, and *Oreochromis niloticus* (tilapia) is one of the most cultured fish species in the country. *Streptococcus agalactiae* (*S. agalactiae*) has been identified as a major pathogen in tilapia farming. It is a zoonotic bacterium that has been reported to cause severe economic losses in tilapia production. The pathogen demonstrates increasing resistance to commonly used antibiotics and has the potential to transfer resistant determinants across populations. This review provides a comprehensive study of *S. agalactiae* in tilapia farming in Nigeria. It explores the nature of bacterial infection in Nigeria, its epidemiology, risk factors, and potential for global resistance. It also describes effective mitigation strategies to prevent infection and the spread of *S. agalactiae* resistance in tilapia farming in Nigeria. The prevalence of *S. agalactiae* in tilapia worldwide ranges from 5% to 60%, resulting in mass mortalities of up to 90%. Overstocking, poor biosecurity, and water quality management are among the risk factors for the infection, and effective mitigation strategies for AMR include the use of non-antibiotic alternatives, farm management and biosecurity implementation, and policy recommendations. This review recommends implementing a One-Health framework to mitigate the spread of this infection.

**Keywords**: *Streptococcus agalactiae*, Antimicrobial resistance, Tilapia, Nigeria, Risk factors, One-Health.

## INTRODUCTION (Background and rationale)

Tilapia farming in global aquaculture is amongst the most significant, with a substantial contribution to economic development, food security and nutrition [1]. Over the past decades, its production has spiked exponentially, with Asia and Africa in the leads [2]. Nigeria's aquaculture sector is one of the leading fast-growing farm businesses, yielding an increase in global production contribution from 0.07% in 1995 to 0.21% in 2022 and placing Nigeria in second place in Africa's aquaculture production [3]. The economic importance of Nile tilapia in Nigeria's aquaculture system is described by its nature and characteristics (adaptability, growth rate, nutritional value) making it an ideal species for small and commercial-scale farming operations in the country. The most common Genetically Improved Farmed Tilapia (GIFT) strain in circulation contributes to income generation and food security [4].

Nigerian tilapia farmers have adopted various production systems, ranging from traditional earthen ponds to modern cage culture systems, to meet the growing demand. These systems can support the commercial production of fish in large volumes, with some farms reaching up to 32 tonnes of annual tilapia production, demonstrating the sector's potential for scaling and improved production [5]. This intensification in production, however, does not come without consequences. Intensification in tilapia farming has been reported to create favourable conditions for disease incidence and outbreaks of bacterial infections [6]. Notable among these bacterial infections affecting tilapia production is Streptococcosis, a zoonotic systemic infection that primarily affects juveniles (though it affects other life stages as well), resulting in high mortality rates during outbreaks. This infection has been reported across tilapia farms globally, with *S. agalactiae* as the leading strain causing outbreaks [7]. *S. agalactiae,* also referred to as the group B *streptococcus* (GBS), is the most recognised pathogen of importance in tilapia production. It's known for its morphological and genetic diversity, distinct host-pathogen infection system and immune responses [6]. The clinical manifestations vary from acute septicaemia and meningoencephalitis to chronic granulomatous inflammation in humans and animals. Affected tilapia exhibit signs of septicaemia and severe systemic inflammation, causing multiple organ dysfunction and widespread systemic damage [8].

Outbreaks often occur in tropical regions, particularly during hot, dry seasons when temperatures exceed 27°C. The pathogen is known for its resistance to multiple antibiotics and its ability to persist in both the fish's aquatic environment and tissues for long periods, making it difficult to control and requiring robust strategies for immediate detection, treatment, and long-term prevention [7]. Thus, with the advent and increased threat of antimicrobial resistance (AMR), this pathogen represents a global concern with significant implications for aquaculture sustainability and public health [8]. This phenomenon (AMR) is particularly concerning in developing countries, where stringent regulatory frameworks and surveillance systems for AMR are inadequate [9]. In Nigeria, it is compounded by factors like access to fish health professionals, inadequate diagnostic facilities, regulation of antimicrobial products, and public knowledge of antimicrobial stewardship [8]. Misuse of antibiotics for growth promotion and prophylaxis in Nigerian aquaculture operations, and antibiotic-laden effluent discharged from fish farms into surrounding environments, contribute to selection pressure for resistant genes and create ecological and public health implications [10].

In 2019, the World Health Organisation (WHO) estimated the incidence of drug-resistant infections to surpass diabetes and cancer, resulting in a 10 million increase in death rates by 2050 annually [11]. AMR has been listed among the top ten threats to global health, food security, and economic development, and over the years, organisations such as the European Centre for Disease Prevention and Control (2018) and the WHO (2017) have documented the increasing prevalence of resistant bacteria, including *S. agalactiae* [12;13]. Almost 40% of WHO member countries that have developed AMR National action plans compliant with the One Health perspective do not have aquaculture included as a critical component in these plans [14]. Meanwhile, heavy use of antimicrobials in agriculture creates a pathway for the transmission of AMR from groundwater to animals and humans through ingestion. When these resistant pathogens cause infection, they are difficult to treat and often relapse, resulting in high morbidity and mortality and an increased economic burden [15].

## Isolation and Identification of *S. agalactiae*

### Sampling and detection techniques

Isolation and identification of *S. agalactiae* require sampling approaches that minimise contamination and maximise pathogen recovery. Plate 1 below provides a representation of the

steps involved in culture and isolation of the pathogen from tilapia fish. Sampling protocols typically involve tissue collection from affected fish, with emphasis on the brain, kidney, spleen, liver, and heart (which harbour the highest concentrations of the bacteria) [16]. The sample collection step is crucial for successful isolation. Tilapia is sampled as soon as possible following death (to promote bacterial viability and prevent post-mortem overgrowth) with sterile instruments disinfected with 70% ethanol before collection, and processed immediately or stored at 4°C for no more than 24 hours before processing [17]. The gold standard for *S. agalactiae* isolation is culture-based. *S. agalactiae* is a gram-positive, beta-haemolytic, catalase-negative, facultative anaerobe, a coccus bacterium that can grow at a wide range of temperatures (preferring 35-37°C) [18]. The culture-based method of isolation typically uses selective and differential media to enhance recovery, suppress competing bacterial growth, and facilitate identification [10]. Blood agar, Edward agar (supplemented with 5% sheep blood), and tryptone soy agar are excellent for culturing *S. agalactiae* in the laboratory [19]. Other media that enhance recovery of the pathogen from samples in the lab and suppress growth of gram-negative bacteria are Columbia nalidixic acid agar and modified Todd-Hewitt broth. The bacterium is incubated at 37°C for 24-48 hours [20].

Following culture, biochemical tests are performed to identify bacteria. Biochemical identification relies on tests such as the catalase test, the CAMP (Christie-Atkins-Munch-Petersen) test (which detects CAMP production), the esculin hydrolysis test, and the oxidase test. The organism is also Gram-positive, appearing as purple cocci in chains under the microscope [19]. Commercial identification systems such as API 20 Strep, Vitek 2, and Phoenix are also employed for *S. agalactiae* identification [21]. They offer standardised protocols and reliable identification, showing good concordance with molecular identification methods compared to the traditional biochemical tests [21]. Another rapid identification technique is Fluorescent in situ hybridisation (FISH), an advanced method that uses fluorescent probes to bind specific ribosomal RNA sequences of bacteria, which can be visualised directly [7]. FTA elute card, combined with visual colourimetric loop-mediated isothermal amplification (FTA-e/LAMP) involving DNA extraction, can also be used to visually confirm *S. agalactiae* in aquaculture settings [22].

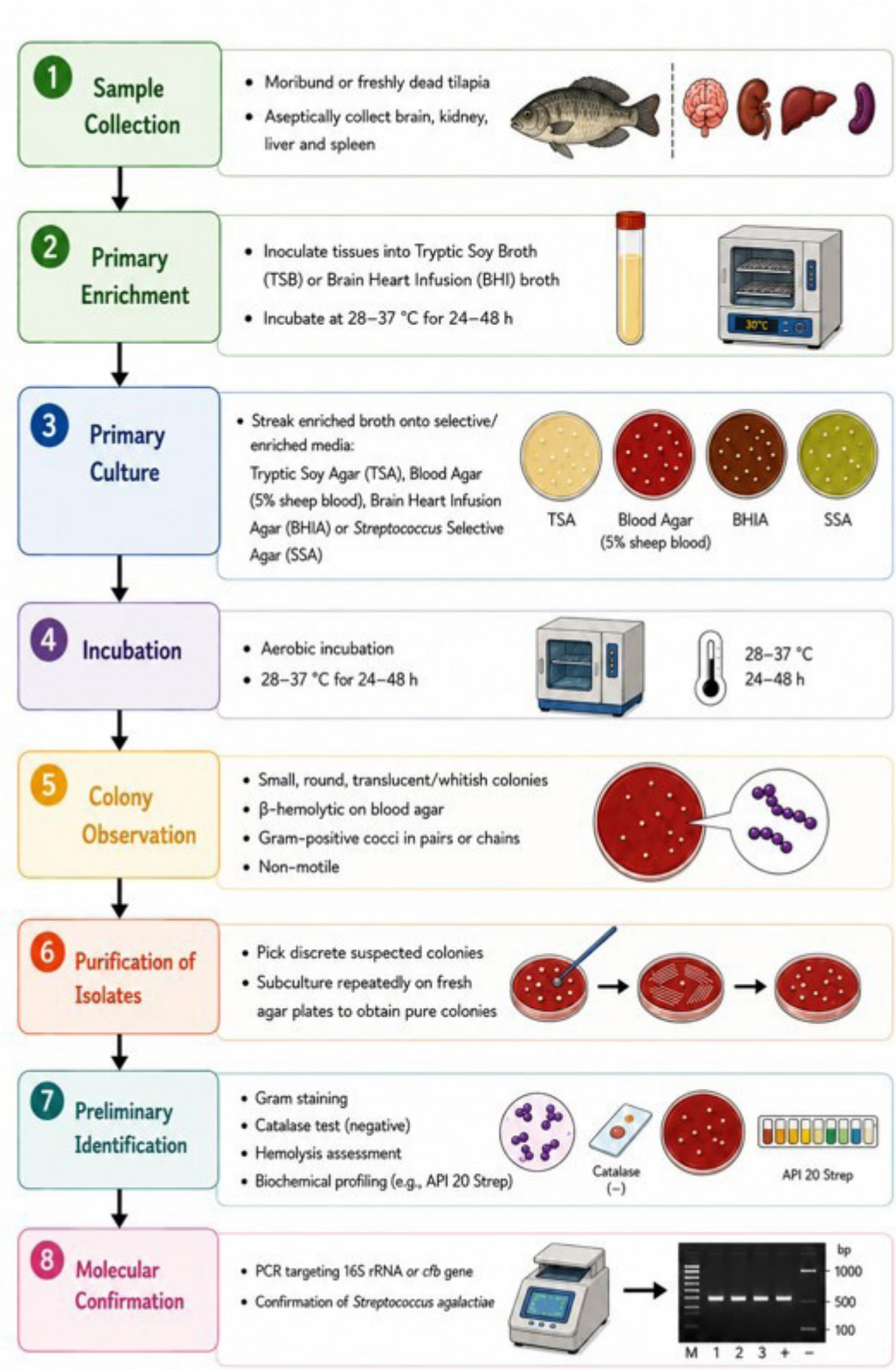


**Plate 1:** Culture and isolation of *Streptococcus agalactiae* from tilapia fish

**Advances in Molecular Diagnostics**

The pathogenicity of *S. agalactiae* is mediated by a range of virulence factors, including capsular polysaccharides (CPS), surface proteins (e.g., alpha- and beta-haemolysins, pili, and *srr* proteins), and immune evasion mechanisms. The organism is classified into ten distinct serotypes (Ia, Ib, II–IX) based on capsular polysaccharide composition, with serotypes Ia, Ib, II, III, and V [23]. Historically, diagnosis of *S. agalactiae* has relied on conventional culture-based and biochemical methods, which, despite their established sensitivity, suffer from prolonged turn-around times and limited discriminatory power for epidemiological purposes [24]. Molecular diagnosis for *S. agalactiae* detection commonly targets genes that have been shown to provide excellent specificity (like 16S rRNA gene, *cfb* gene, *fbsA* gene and *cspA* gene) through techniques like Polymerase chain reaction (PCR) assays that target species-specific genes, offering rapid and accurate results [25]. DNA-sequencing techniques like whole genome sequencing reveal extensive genetic diversity within *S. agalactiae* populations, with distinct lineages existing, and proteomic and molecular fingerprinting techniques to provide accurate diagnosis, efficient tracking and understanding of epidemiological patterns [26]. Over the decades, molecular techniques for diagnosing this bacterium have been developed and refined. These technologies enable not only rapid pathogen identification but also capsular typing, virulence gene profiling, antimicrobial resistance genotyping, and molecular epidemiological surveillance [27]. They include Polymerase chain reaction (PCR), quantitative real-time PCR (qPCR), and next-generation sequencing (NGS), with revolutionised speed, sensitivity, and specificity of detection and characterisation [28].

Polymerase chain reaction (PCR) is a thermocyclic *in vitro* DNA amplification technique developed by Kary Mullis in 1983 [29]. It is designed to exponentially replicate a defined target sequence using thermostable DNA polymerase, oligonucleotide primers, deoxynucleotide triphosphates, and a template DNA [28]. A standard PCR cycle involves three steps: denaturation (94–98°C) to separate double-stranded DNA; annealing (50–65°C) for primer binding to complementary sequences; and extension (72°C) for DNA synthesis by Taq or other thermostable polymerases [30]. In the diagnosis of *S. agalactiae*, PCR enables the specific detection of conserved chromosomal targets or virulence genes (producing PCR products) without the need for viable bacteria. After 25–40 cycles, products are visualized by gel electrophoresis with ethidium bromide or SYBR Safe staining,

permitting qualitative detection of the target amplicon [30]. Real-time PCR, or quantitative PCR (qPCR), is a modification of conventional PCR that monitors DNA amplification in real time by detecting fluorescent reporter signals. Unlike the conventional PCR, qPCR enables quantification of the initial template amount and eliminates the need for post-amplification gel electrophoresis, reducing assay time, contamination risk, and technical complexity [29]. Two principal chemistries are employed: intercalating dyes (SYBR Green) and sequence-specific fluorescent probes (TaqMan, Molecular Beacons, Scorpions) [31]. The TaqMan-based qPCR is the most widely used format for diagnostic applications. It typically makes use of a dual-labelled probe bearing a 5' fluorophore and a 3' quencher. The probe hybridises to the target sequence between the primer binding sites, and during extension, the 5'-3' exonuclease activity of Taq polymerase cleaves the probe, releasing the fluorophore from the quencher, generating a signal proportional to amplicon quantity. The cycle threshold (Ct) value, at which fluorescence crosses a defined threshold, is inversely proportional to initial template concentration [31].

Sequencing techniques that provide more precise, accurate genetic information about the pathogen exist. First-generation Sanger dideoxy sequencing, developed by Frederick Sanger in 1977, employs chain-terminating dideoxynucleotide triphosphates (ddNTPs) to generate a nested series of labelled DNA fragments. These fragments, resolved by capillary electrophoresis, will produce a sequence read of 500–1,000 base pairs [32]. This process can be applied for definitive species identification via 16S rRNA gene sequencing, multilocus sequence typing (MLST), and characterisation of antimicrobial resistance gene alleles in *S. agalactiae* diagnosis [24]. MLST was introduced to enable the diagnosis of bacterial isolates by sequencing their housekeeping genes. For *S. agalactiae*, the genes *adhP*, *atr*, *glcK*, *glnA*, *pheS*, *sdhA*, and *tkt* are used to generate sequence types (STs) and clonal complexes (CCs) that enable fine-resolution epidemiological discrimination [24]. Next-generation and third-generation sequencing techniques, such as Illumina short-read sequencing and Long-read sequencing (Oxford Nanopore and PacBio), offer the possibility of producing billions of reads from a single experiment [33]. Illumina sequencing-by-synthesis (SBS) technology generates massively parallel short reads (75–300 bp) from fragmented, adapter-ligated DNA libraries. Billions of reads can be generated per run on platforms such as the HiSeq, NovaSeq, and MiSeq, enabling compre-

hensive genomic characterisation at decreasing cost [27]. Whole-genome sequencing on the Illumina platform for *S. agalactiae* enables simultaneous determination of sequence type, serotype (via in silico cps typing), resistance genotype, virulence gene content, and core genome phylogeny from a single sequencing experiment [33]. Third-generation sequencing platforms like Oxford Nanopore Technologies (ONT) and Pacific Biosciences (PacBio) can generate long reads (>10 kb, up to >100 kb for Nanopore technology). They enable the *de novo* assembly of closed, complete bacterial genomes and resolve repetitive elements, mobile genetic elements, and plasmids that are typically fragmented in short-read assemblies [34].

**Diagnostic challenges and reliability**

One major issue in the reliability of identification tests is the possibility of false-negative results, which may be caused by several factors, such as sample degradation, PCR inhibitors, or bacterial cell death during sample transportation and storage [35]. False-negative outcomes may cause missed diagnoses and delayed treatment, which may cost a lot of lives and economic loss. The creation of internal controls and sample processing procedures that reduce the impact of inhibitors has gained more importance to guarantee the reliability of the diagnosis [6]. *S. agalactiae* identification methods include the hydrolysis of sodium Hippurate, the CAMP reaction, pigment production, and antibiotic disk susceptibility, immunological tests such as Lancefield's classical precipitin test, immunofluorescence staining, counter immunoelectrophoresis, and coagglutination [36]. The CAMP test is considered an important confirmatory test for the identification of *S. agalactiae*, which is low-cost and easy to perform [36], but its specificity and sensitivity have been questioned [37]. The reliability of this test in fish isolates has been questioned, with some studies reporting CAMP-negative strains that are confirmed as *S. agalactiae* by molecular methods [35].

Another key issue that may cause misdiagnosis of the pathogen is contamination during sample collection, transportation, or processing [21]. Cross-contamination of samples may occur at various points throughout the diagnostic process, from field sampling to laboratory testing [36]. To reduce the risk of contamination, stringent quality control standards, such as negative controls, sterile sampling, and appropriate laboratory procedures, are necessary [38]. Proper sampling methods and the use of proper sample containers and media of transport should be strictly adhered to by field personnel to ensure

the integrity of the sample [39]. Environmental interference can significantly affect the diagnostic accuracy of aquaculture samples, particularly when multiple bacterial species are present. This may reduce the visibility of *S. agalactiae* colonies on culture media due to competing bacteria, and other bacteria may interfere with PCR amplification [38]. Environmental conditions, including pH, temperature, and organic matter content, can also influence diagnostic performance, posing challenges even when using selective media and highly specific PCR reactions [39].

Due to the genetic diversity of *S. agalactiae* strains, diagnostic assays and validation may also pose a challenge [17]. The difference in traits (genetic/phenotypic) of strains associated with fish to those associated with humans could influence the performance of diagnostic tests that have been initially designed to work with human clinical isolates [21]. To maintain high accuracy and reliability, diagnostic protocols should be regularly validated and updated. The presence of dead bacteria or bacterial DNA in samples can contaminate samples and confuse interpretation as positive PCR results [39]. Therefore, quality assurance and standardisation of diagnostic procedures are very important, especially in resource-restricted environments where access to quality control materials and proficiency tests might be scarce [40]. Standardised operating procedures and quality control programs are indispensable in guaranteeing reproducible and trustworthy diagnostic outcomes [17]. The table (1) below provides a summary of common problems that contribute to diagnostic challenges in *S. agalactiae* culture/molecular detection.

**Table 1.** Common problems/issues in culture/molecular detection of *S. agalactiae*

| Problem | Diagnostic challenge | Causative factor | Mitigation strategy | Diagnostic method affected | References |
|---|---|---|---|---|---|
| Sample validity | False results (false-negative/ false-positive) | Sample degradation, contamination, improper sample handling, poor storage or transport medium/containers | Proper sample collection process, storage medium, optimised aseptic protocol during sample processing and use of control strains for confirmation | Culture, molecular | Haenen *et al.* [6]; Abd El Atty *et al*. [21]; Zhou *et al*. [35]; Nair *et al*. [38] |
| CAMP test reliability | Misidentification of bacteria (some strains, despite being present in the sample, test negative) | Sensitivity and specificity of the test | Complemented by other tests and confirmation using molecular methods | Culture | Zhou *et al*. [35]; Boyanov *et al.* [36]; Ballard *et al.* [37] |
| Detection of non-viable genetic material | PCR/qPCR interference | PCR inhibitors, unsuitable pH, presence of non-viable DNA or dead bacteria | Optimised PCR conditions, quality control programs | Molecular | Nair *et al.* [38]; Leigh *et al.* [40] |
| Primer/probe design challenges | False interpretation, missed detection | Genetic diversity, mutation, and reduced sensitivity/performance of the diagnostic platform | Robust primer designs, assay validation | Molecular | Algammal *et al.* [17]; MacAulay *et al.* [39] |

**Key:** CAMP = Christen-Atkins-Munch-Peterson; PCR = Polymerase chain reaction; qPCR = real-time/ quantitative PCR; DNA = deoxyribonucleic acid.

**Prevalence and Distribution of *S. agalactiae*: Global and regional prevalence**

The global and regional prevalence of *S. agalactiae* in tilapia farming has been documented extensively, with significant variations observed between geographical regions and different environmental conditions [41]**.** In regions producing the highest volume of tilapia annually (Asian countries like China, Thailand, Malaysia and Indonesia), high prevalence rates exceeding 50% in affected farms were reported [42]. Latin American countries like Brazil, Colombia, Ecuador report prevalence rates ranging between 15% to 60%, with higher rates observed in areas with intensive farming systems and during periods of environmental stress [43]. A study reported a high prevalence of 86.7% of *S. agalactiae* alone and 4.85% for mixed infection of *S. agalactiae* and *S. iniae* in Thailand [26] and in Mexico, outbreaks with mortality rates reaching 68% and 80% have been recorded [7]. Recently, mass mortality (50 – 90%) resulting from *S. agalactiae* outbreaks has been reported in Taiwan, Bangladesh, India and Egypt [44 - 47].

In Africa, varying reports from different tilapia production systems in Egypt have been reported. Prevalence rates of 5% - 20% in different small-scale farming systems and higher rates exceeding 26.2% in intensive cage culture systems were documented [17]. The resistance profile of this pathogen has also shown 7% prevalence of isolates showing resistance to ampicillin and erythromycin in Egypt [19]. In Ghana, serotype distribution, virulence and AMR potential of *S. agalactiae* isolated from tilapia farmed in Lake Volta recovered 32 isolates [25]. In Australia, *S. agalactiae* strains recovered were the same as the ST-260 sequence type isolated from tilapia in Latin America and ST-261 type isolated in the United States, China, Ghana and Israel [48]. Specific *S. agalactiae* data from Nigerian tilapia farms are limited. Despite Nigeria's status as a major aquaculture producer in Africa, evidence to suggest the presence of the pathogen on the Nigerian tilapia farms is lacking. However, anecdotal reports from fish farmers in intensive systems and veterinarians during the typical dry season that favour the pathogen's proliferation indicate streptococcal infections exist. True prevalence reports on the *S. agalactiae* isolated from tilapia farms in Nigeria are limited.

Variations in regional prevalence rates can be attributed to several factors: import of infected broodstock into South America, rapid introduction to other American regions through trade networks and extension of tilapia farming in other regions. Dissemination is also facilitated significantly by climate changes, farming prac-

tices, fish genetics and availability of pathogen introduction pathways [6]. Tropical regions with water temperatures consistently exceeding 25°C favours *S. agalactiae* proliferation and the genetic diversity of tilapia populations may also influence susceptibility [49]. Temporal dynamics of *S. agalactiae* prevalence show higher rates during periods of environmental stress, such as drought, temperature fluctuations or poor water quality [4].

**Risk factors for *S. agalactiae* infection in tilapia farming**

A summary of risk factors for *S. agalactiae* infection has been provided in Plate 2 below. The most prominent risk factor for *S. agalactiae* infections in tilapia is the stocking density. Positive correlation between stocking density and disease incidence has been demonstrated [50]. Physiological stress resulting from high stocking density (exceeding 50 fish per cubic meter) compromises fish immune function, promoting susceptibility to infections. The dense population also facilitates pathogen transmission through increased contact between the fish and higher bacterial load in the water [51]. The water temperature also plays an important role; temperature fluctuations and consistently high temperatures represent critical environmental risks [52]. *S. agalactiae* shows the ability to thrive at temperatures between 25-30°C (the preferred temperature range for tilapia culture and this gives room for outbreaks to occur), especially during hot and dry seasons when the temperature is ≥27°C or sudden temperature changes that trigger stress in fish [53]. Therefore, it is important to note that climate change and the increase in global temperature variations may exacerbate the risk of infection and expand the geographical range of *S. agalactiae* infections on tilapia farms globally [9].

Another important risk factor is nutrition. Nutritional stress and feed quality issues contribute a great deal to fish susceptibility to this infection [54]. Contaminated feed may harbour pathogenic bacteria, a lack of essential nutrients in fish fed low-quality feed can compromise the immune capability and overfeeding results in water quality deterioration [55]. Poor water quality conditions (including low dissolved oxygen levels, high ammonia concentrations and organic matter overloads) create favourable conditions for bacterial growth and compromise fish health by triggering stress [56]. Farms with poor water exchange channels and effluent management accumulate organic matter from leftover feed, fish waste, etc., to create anaerobic conditions that promote pathogen proliferation and toxic metabolite production [56].

Biosecurity on the farm limits outbreaks, failures facilitate pathogen introduction and spread within and between farms [57]. Unrestricted personnel movement between farms, movement of infected animals (in this case, fish), contaminated equipment/materials, and inadequate disinfection protocols can introduce pathogens from infected farms to uninfected farms [57]. Poor fish handling triggers stress (stress during grading, transport, health assessment, etc.), which could cause disease [56]. The presence of other pathogens or concurrent infections exacerbates the risk of *S. agalactiae* infections. It results in immune suppression and tissue damage, providing entry portals for bacterial, parasitic, or viral infections, which create a predisposing condition for *S. agalactiae* establishment and proliferation [3].

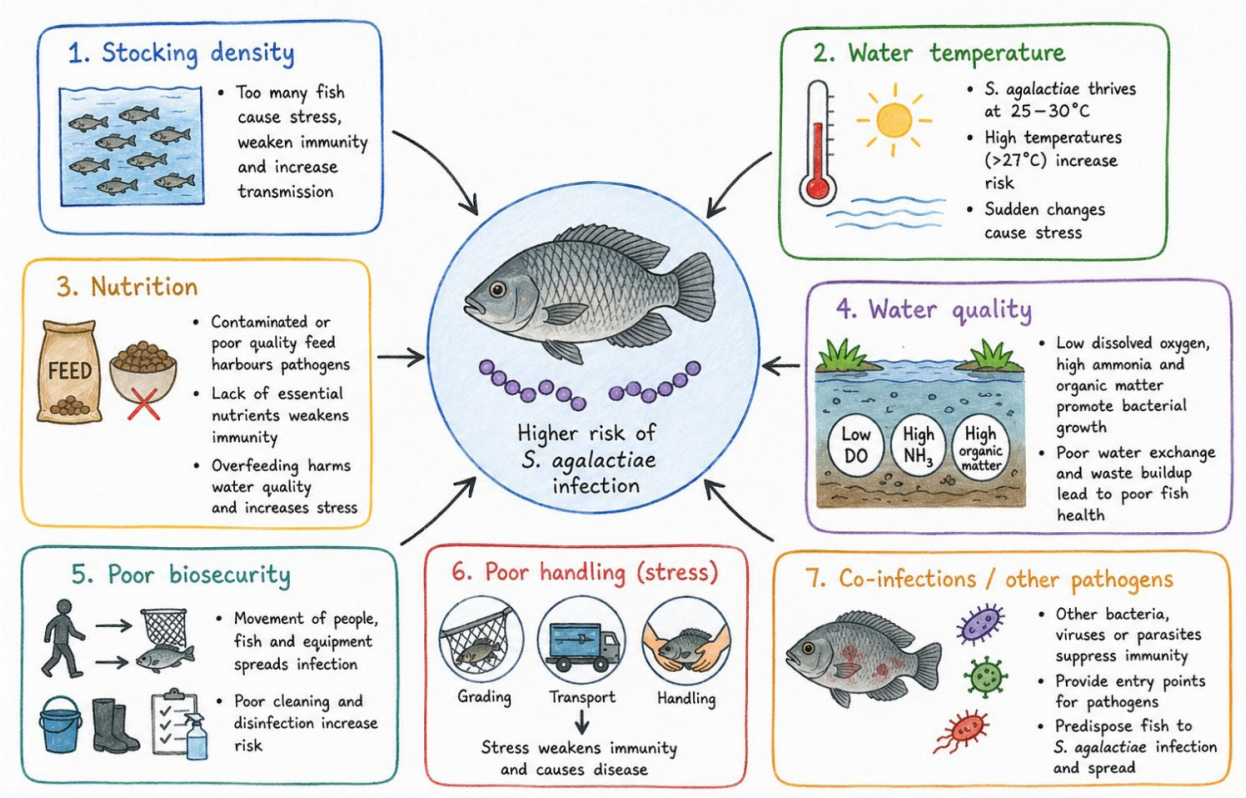


**Plate 2:** Risk factors for *S. agalactiae* infection in tilapia farming

### Temporal trends and outbreak history

Multiple pathways support the spread of *S. agalactiae* through tilapia populations. The infected fish shed the bacteria into the surrounding water, and direct transmission between fish, vertical broodstock transmission, transmission in water, and contact with equipment foster its dissemination. Entry portals include the gills, skin abrasions and gastrointestinal tract of the fish. The bacteria colonise and

proliferate by adhering to epithelial cells using proteins and adhesins [7]. Seasonal patterns of *S. agalactiae* infection in tilapia farming show consistent trends across different geographical regions, with peak incidence typically occurring during warmer months and periods of environmental stress [49].

In tropical regions, the dry season (December to March) is typically associated with higher disease incidence due to increased water temperatures, reduced water levels, and concentrated fish populations. During this period, evaporation rates increase, leading to higher concentrations of organic matter and potentially harmful substances in pond water. The combination of thermal stress and poor water quality creates ideal conditions for *S. agalactiae* proliferation and fish immune suppression [49;58]. Low temperatures present different challenges that can also contribute to *S. agalactiae* outbreaks. It can lead to rapid changes in water chemistry, temperature fluctuations, low feed intake, metabolism, and growth performance that can stress fish and introduce pathogens [58]. Heavy rainfall and flooding events can facilitate pathogen spread between farms and water bodies, while sudden influxes of freshwater can cause osmotic stress that compromise fish immune function [59]. The seasonal variation in disease incidence requires adaptive management strategies that account for changing environmental conditions throughout the year.

**Antimicrobial resistance in *S. agalactiae*: Resistance profiles and patterns**

*S. agalactiae* has demonstrated high resistance patterns to common antibiotics in circulation with 95% of isolates showing resistance to ampicillin and erythromycin [60]. Resistance patterns of *S. agalactiae* from tilapia farming systems revealed concerning trends in development and dissemination. This was observed in Vietnamese isolates showing widespread multidrug resistance, particularly nalidixic acid, neomycin, oxacillin, oxytetracycline, and β-lactam/β-lactamase inhibitor combinations [61]. In Egypt, 35.7% of the isolates exhibited resistance to 7 different classes of antimicrobials and carried resistant genes (*pbp1A*, *tetM*, and *ermA* genes) [17]. Nineteen (19) out of 21 isolates recovered from tilapia in Egypt possessed virulence-associated genes (*fbsA, cfb,* and *pbp1A/ponA*) as 100%, 76%, and 52%, respectively with multidrug resistance pattern (cefotaxime, trimethoprim-sulfamethoxazole, ceftriaxone, tetracycline, gentamicin, penicillin and high resistance against ampicillin and erythromycin [19]. Kirby-Bauer disk diffusion assay of *S. agalactiae* (serotype Ia) isolated from Lake Volta in Ghana yielded fewer alarming results. The majority of the isolates were susceptible to

the tested antibiotics (oxytetracycline, erythromycin, florfenicol, enrofloxacin, ampicillin and amoxicillin) except gentamicin [25].

Antibiotic resistance genes detected in the isolates include *aac* (6′)-*Ib-cr* (35.7%), *bla*CTX (23.8%), *qnr*S (19%), *ampC* (16.7%), *floR* (14. 3%), *sul*1, *tet*A, and *van*.C1 (2.4%). This high level of resistance to commonly used antibiotics reflects the selective pressure exerted by widespread and often inappropriate use of these drugs [60]. Multidrug resistance (MDR) has become increasingly common in *S. agalactiae* isolates from tilapia, with many strains showing resistance to three or more different antibiotic classes [62]. The predominant antibiotic-resistant types showed resistance to multiple antibiotics, including penicillin, chloramphenicol, sulfamethoxazole-trimethoprim, ceftriaxone, tetracycline, cefotaxime, erythromycin, and ampicillin [63]. This pattern of multidrug resistance severely limits treatment options and may require combination therapy [62]. Multi-Antibiotic Resistance index (MAR) of aquaculture-related bacteria (11,274 isolates) for 40 countries, of which are mostly low- and middle-income countries present high AMR levels [64]. This pattern is particularly concerning for Nigeria, as a developing country with an expanding aquaculture sector, where regulatory oversight and antimicrobial stewardship programs may be less developed.

**Mechanisms of resistance**

The pathogen has a polysaccharide capsule that masks its antigens, making it difficult for the host's immune system to detect it. It can form biofilms on the mucosal surface and other tissues, providing itself with a shield from the host's immune system and antibiotic treatments [65]. Horizontal gene transfer of resistance between the bacterial species in aquatic environments is of great concern in the development of AMR in tilapia farming [66]. Transposons and integrons are mobile genetic elements that promote the transmission of resistance genes not only among *S. agalactiae* populations but also to other bacterial species found in the aquatic environment [67]. The pathogen's mechanism of resistance is also related to its ability to alter binding proteins, modify ribosomal targets, and express efflux pumps. The presence of virulence genes also contributes to its resistance mechanisms [17]. The *cfb* gene encodes for the CAMP factor, a pore-forming toxin that induces red blood cell lysis. It also possesses resistance determinants such as *pbp 1A, aac* (6') aph (2''), *tetM, ermA, tetK,* and *ermB* genes that promote the development of MDR [17].

The development of tetracycline resistance in

*S. agalactiae* has been attributed to both chromosomal mutations and the acquisition of resistance genes through horizontal gene transfer [66]. The widespread use of oxytetracycline as a feed additive and therapeutic agent has exerted strong selective pressure on the development of resistance [66]. Macrolide antibiotics, including erythromycin and azithromycin, have shown variable patterns of resistance in *S. agalactiae* isolates [67]. The resistance to macrolides is typically mediated by methylation of the ribosomal target site or by efflux pumps that actively remove the antibiotic from bacterial cells [67]. Cross-resistance among macrolides means that resistance to one drug in this class often confers resistance to others, limiting therapeutic options when macrolide resistance is present [67]. Global temporal trend, including past, present, and contemporary findings, indicate that the pathogen harbours antibiotic resistance genes (ARGs) conferring resistance to 9 antibiotic classes, predominantly macrolide, tetracycline, aminoglycoside, and lincosamide resistance genes [68].

**Link to Human and Environmental Health**

*S. agalactiae* is a zoonotic organism and leading cause of severe infections in human populations, often resulting in life-threatening conditions such as sepsis, pneumonia, and meningitis [69]. The dual-way pathogenicity of zoonotic pathogens establishes a direct route of AMR gene transfer between aquatic and terrestrial systems, and this may undermine the clinical treatment of humans [70]. The AMR gene flow to the environment takes place in various ways, such as water released by aquaculture plants, fish products contaminated with toxins and direct interaction between aquaculture employees and infected fish [71]. The aquatic environment can serve as a reservoir for resistant bacteria and genes, which can persist in sediments and water for extended periods and spread when favourable environmental conditions influence them [64]. In Plate 3, the interconnectedness of this relationship and the resultant effects on One-Health is elucidated.

The threat of *S. agalactiae* from tilapia farming to One Health is possible due to interactions among humans, fish, and environmental health [72]. AMR genes introduced into tilapia farming systems have the potential to reach human populations through different pathways, such as foodborne, occupational (during handling), and environmental (effluent management) [72]. The economic consequences of this interconnectedness are significant, as the potential loss of effective antimicrobials in human medicine due to the development of resistance in aquaculture environments is a major public health issue, far beyond the direct effects on the profitability of

fish farming [72].

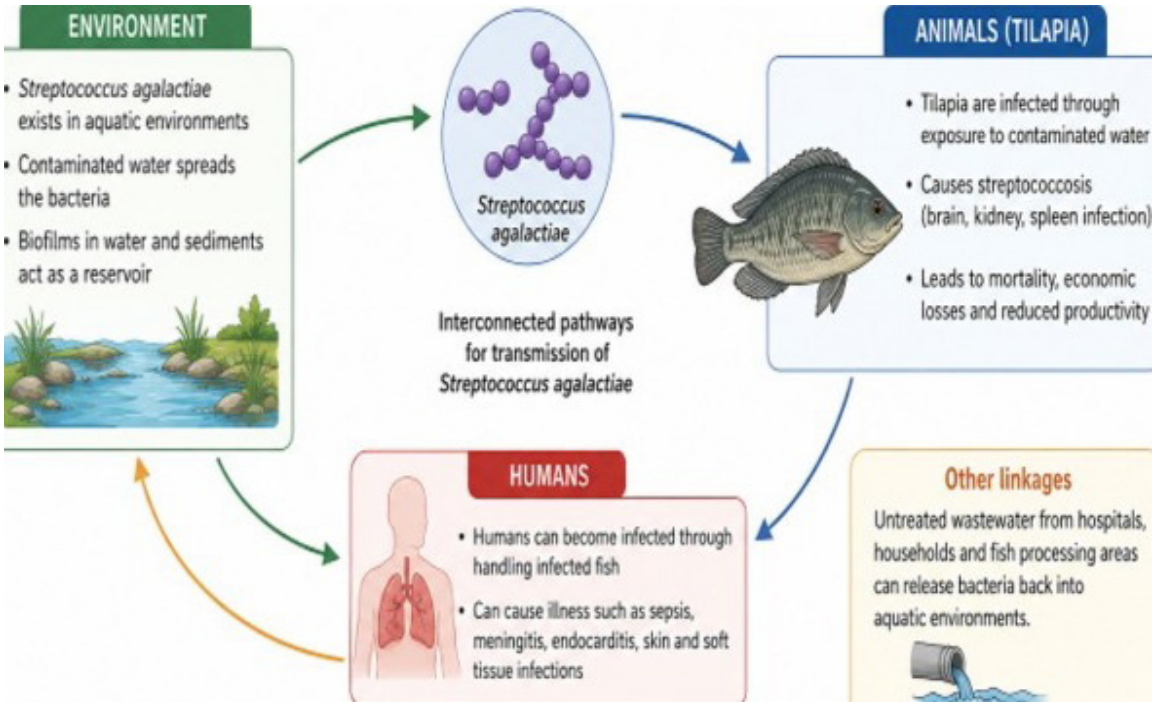


**Plate 3:** One health link of *S. agalactiae* infection in animal (tilapia), human and environment

**Mitigation strategies for AMR in Nigerian tilapia farming**

**Prudent use of antimicrobials**

Prudent antimicrobial use is a fundamental strategy for AMR mitigation. Stewardship protocols for prophylactic versus therapeutic use must be clearly defined to minimise selective pressure for resistance development [73]. Antibiotic use for prophylaxis should be limited to high-risk situations with scientific justification, while therapeutic use should be reserved strictly for confirmed bacterial infections [74]. Distinguishing these two approaches is essential because antibiotics are commonly used for prevention and growth promotion. While it potentially serves the intended prophylactic purpose, it also creates a sustained condition that favours the emergence of resistant bacterial populations [73]. Therefore, ensuring antibiotic stewardship on the farms should incorporate farm personnel training that emphasises the importance of completing the treatment course, record keeping, identification of potential trends and risk factors for AMR [71].

A stewardship approach in tilapia production, therefore, requires more than simply telling farmers to use fewer drugs. It involves better diagnosis, better treatment records, stronger oversight of what is used on farms, and training of personnel to recognise disease early and respond appropriately [75].

Many alternatives to antibiotics in aquaculture work indirectly by preventing disease and improving management rather than by acting as simple substitutes for antimicrobials [75]. This is important for AMR mitigation in tilapia farming, as it goes beyond restricting products to redesigning production systems to make disease events less frequent and antibiotic dependence less routine.

**Non-antibiotic interventions**

Disease prevention is another crucial part of mitigation. At present, antibiotics remain the most widely used preventive measure, but other strategies have been reported [76]. Vaccination is a promising strategy for preventing *S. agalactiae* infection in tilapia. Studies have tested the efficacy of various vaccines (formalin-killed, oral/feed-based) in protecting against multiple serotypes of *S. agalactiae*. Although the efficacy of these vaccines varies with challenge conditions and vaccine formulation, success has been recorded in some studies [77;78]. It has also been reported that commercially available vaccines against *S. agalactiae*, already in use in tilapia production systems, are now an established part of disease control in major tilapia-production settings [79]. Furthermore, formalin-inactivated vaccination in broodstock can produce strong immune responses and sustained protection when booster timing is optimised [80]. These findings support the view that vaccines are no longer only experimental tools in tilapia health management, but practical disease-prevention measures with direct relevance to AMR reduction.

In addition, evidence for vaccine efficacy is also increasingly convincing. A single commercial vaccine dose protected Nile tilapia from 15 to 300 days post-vaccination, with relative survival values ranging from 67% to 94% across challenge points [79]. Similarly, a study found that booster vaccination at 4 or 12 weeks after the primary dose prolonged adequate protection in broodstock for at least 28 weeks [80]. Beyond biological efficacy, vaccination also appears economically meaningful. Vaccination against *S. agalactiae* in Nile tilapia farms could be economically advantageous under realistic mortality scenarios, which strengthens the argument that vaccines should be treated as both a health and production intervention [81]. In AMR terms, this matters because every successful vaccination program helps reduce the number of outbreaks that would otherwise prompt antimicrobial treatment.

It has been demonstrated that a phytogenic feed additive improved feed conversion, immune response, and survival after *S. agalactiae* challenge in Nile tilapia [84]. However, medicinal plant derivatives, categorised among the more promising alternatives to on-farm antibiotic use in aquacul-

ture, should be used with care [75]. They should be viewed mainly as supportive measures rather than as stand-alone replacements for sound husbandry, vaccination, and biosecurity [75]. Their value lies in strengthening the overall health status of the fish and reducing susceptibility, not in functioning as a simple one-for-one substitute for antibiotics. Probiotics and prebiotics, employing strains of beneficial bacteria in various forms for prophylactic and therapeutic purposes, have also been extensively studied [82]. These alternatives to antibiotics enhance fish health by modulating gut microbiome, promoting growth rates and improving immune responses [63].

*Lactobacillus* species, *Bacillus subtilis*, and *Enterococcus faecium* have shown particularly promising results, competitively excluding pathogenic bacteria, producing antimicrobial compounds, and enhancing the host immune response [82].

**Farm management and biosecurity**

Farm management and biosecurity are equally central to AMR mitigation because outbreaks of Streptococcosis are strongly shaped by production stress. Biosecurity and farm management practices like ensuring water quality parameters (dissolved oxygen levels >5 mg/L, temperature 26-30°C, pH 6.5-8.0, and ammonia levels <0.5 mg/L), appropriate filtration and aeration systems, optimal stocking density, disinfection and hygiene protocols maintain fish health and reduce the incidence of *S. agalactiae* outbreaks [85]. Implementing all-in/all-out production systems can reduce the accumulation of pathogens on the farm, and rigorous hygiene protocols mitigate transmission of this pathogen [85]. Use of footbaths and hand-washing stations, proper disposal of dead fish and contaminated material, help prevent environmental contamination and reduce pathogen reservoirs [17]. On the contrary, practices like aggressive handling, high stocking density, and poor water quality have been identified as management-related stressors that increase disease vulnerability across the tilapia value chain [85]. This means that water management is not merely a routine technical issue: it is part of disease control. Stable and suitable water conditions, reduced crowding, and reduced handling stress help maintain immune competence and make outbreaks less likely, thereby reducing the perceived need for antimicrobial intervention.

Biosecurity should also be treated as a whole-farm and whole-value-chain system rather than a single set of isolated practices. Disease risk in tilapia production can enter through hatcheries, nurseries, live fish movement, equipment, and multiple contact points along the production val-

ue chain [85]. Similarly, alternatives to antibiotics work best when combined with better farm management. In practical terms, this supports other measures such as quarantining incoming stock, cleaning and disinfecting nets and equipment, controlling the movement of personnel and materials, properly disposing of mortalities, and stricter hygiene routines around production units. These steps do not directly kill resistant bacteria as an antibiotic might. Instead, they reduce opportunities for pathogen introduction, amplification, and spread, which is often the more sustainable strategy [75].

**Policy and regulatory framework**

A policy and regulatory framework that effectively address challenges of resistance development must be incorporated in national action plans on AMR. International organisations, such as the World Organisation for Animal Health (WOAH), Food and Agriculture Organisation (FAO) and World Health Organisation (WHO), have a crucial role in the provision of technical guidance and coordination of AMR mitigation strategies [86]. Harmonisation of national regulations with international standards facilitates and ensures that these strategies are consistent with best global practices [87]. Capacity building programs for regulatory personnel, extension workers and stakeholders in the production system are also essential for effective policy implementation [87]. These frameworks should encourage economic incentives for responsible aquaculture practices and establish reference laboratories in countries like Nigeria, where limited information on *S. agalactiae* in tilapia is a bottleneck.

**Conclusion and recommendations**

This review presents a comprehensive analysis of *S. agalactiae* infection in tilapia, revealing pathogenic pathways, risk factors for infection and AMR in tilapia aquaculture. *S. agalactiae* is a significant pathogen in tilapia farming with documented resistance to multiple classes of antimicrobials like erythromycin and neomycin. Practical recommendations for AMR mitigation that can be incorporated into the production system have been highlighted to include (implementing antimicrobial stewardship at farm level, using antibiotic alternatives such as probiotics, phytogenic compounds, and prebiotics, preventing outbreaks through strict hygiene and biosecurity protocols, nutrition, minimising fish handling stress, and strengthening regulatory mechanisms). Success in AMR mitigation strategies on tilapia farms requires sustained commitment from government agencies, tilapia industry stakeholders, research institutions, and international agencies that employ the One-Health approach to ensure that tilapia produc-

tion goals do not compromise environmental and human health outcomes. The strategy could be facilitated through working collaboratively to achieve food security, environmental protection, and health preservation.

## REFERENCES

1. Arumugam, M., Jayaraman, S., Sridhar, A., *et al*. (2023). Recent advances in tilapia production for sustainable developments in Indian aquaculture and its economic benefits. *Fishes*, 2023;8(4):176. doi:10.3390/fishes8040176

2. El-Sayed, A.M. and Fitzsimmons, K. (2023). From Africa to the world—The journey of Nile tilapia. *Rev Aquac*., 2023;15(S1):6-21. doi:10.1111/raq.12738

3. Abd El-Hack, M.E., El-Saadony, M.T., Nader, M.M., *et al*. (2022). Effect of environmental factors on growth performance of Nile tilapia (Oreochromis niloticus). *Int J Biometeorol*. 2022;66(11):2183-2194. doi:10.1007/s00484-022-02347-6

4. Okon, E.M., Falan, B.M., Solaja, S.O. *et al*. (2021). Systematic review of climate change impact research in Nigeria: Implication for sustainable development. *Heliyon*, 2021;7(9):e07941. doi:10.1016/j.heliyon.2021.e07941

5. Miao, W. (2020).Trends of aquaculture production and trade: Carp, tilapia and shrimp. *Asian Fish Sci*., 2020;33S. doi:10.33997/j.afs.2020.33.S1.001

6. Haenen, O.L.M., Dong, H.T., Hoai, T.D., *et al*. (2023). Bacterial diseases of tilapia, their zoonotic potential and risk of antimicrobial resistance. *Rev Aquac*.,15(S1):154-185. doi:10.1111/raq.12743

7. Sule, H.A., Saba, A.O. and Yu, C.Y. (2025). Streptococcosis in Aquaculture: Advances, Challenges, and Future Directions in Disease Control and Prevention.

8. Laith, A.A., Abdullah, M.A., Nurhafizah, W.W.I. *et al*. (2019). Efficacy of live attenuated vaccine derived from the *Streptococcus agalactiae* on the immune responses of *Oreochromis niloticus*. *Fish Shellfish Immunol*., 90:235-243. doi:10.1016/j.fsi.2019.04.052

9. Thornber, K., Bashar, A., Ahmed, MdS, *et al*. (2022). Antimicrobial resistance in aquaculture environments: Unravelling the complexity and connectivity of the underlying societal drivers. *Environ Sci Technol*., 56(21):14891-14903. doi:10.1021/acs.est.2c00799

10. Alhaji, N.B., Maikai, B.V. and Kwaga, J.K.P. (2021). Antimicrobial use, residue and resistance dissemination in freshwater fish farms of North-central Nigeria: One health implications. *Food Control*., 130:108238. doi:10.1016/j.foodcont.2021.108238

11. Morrison, L. and Zembower, T.R. (2020). Antimicrobial resistance. *Gastrointest Endosc Clin N Am*., 30(4):619-635. doi:10.1016/j.giec.2020.06.004

12. Christaki, E., Marcou, M. and Tofarides, A. (2020). Antimicrobial resistance in bacteria: Mechanisms, evolution and persistence. *J Mol Evol*., 88(1):26-40. doi:10.1007/s00239-019-09914-3

13. Kariyawasam, R.M., Julien, D.A., Jelinski, D.C. *et al*. (2022). Antimicrobial resistance (AMR) in COVID-19 patients: A systematic review and meta-analysis (November 2019–June 2021). *Antimicrob Resist Infect Control*., 11(1). doi:10.1186/s13756-022-01085-z

14. Caputo, A., Bondad-Reantaso, M.G., Karunasagar, I. *et al*. (2023). Antimicrobial resistance in aquaculture: A global analysis of literature and national action plans. *Rev Aquac*., 15(2):568-578. doi:10.1111/raq.12741

15. Tang, K.W.K., Millar, B.C. and Moore, J.E. (2023). Antimicrobial Resistance (AMR). *Br J Biomed Sci*. *Institute of Biomedical Science (IBMS)*, 80. doi:10.3389/bjbs.2023.11387

16. Girijan SK, Krishnan R, Maniyappan K, Pillai D. (2023). Isolation and identification of

*Streptococcus agalactiae* in cage-cultured green chromide *Etroplus suratensis* in Kerala, India. *Dis Aquat Organ*, 154:1-6.

17. Algammal, A.M., Mabrok, M., Almessiry, B.K. *et al*. (2025). Unraveling the pathogenic potential, virulence traits, and antibiotic resistance genes of multidrug-resistant *Streptococcus agalactiae* strains retrieved from Nile tilapia. *BMC Microbiol*., 25(1):629. doi:10.1186/s12866-025-04405-6

18. Mohamed Amira, El Asely, A., Abdel Gwad Eman, Abbass A, Shaheen Adel. (2023). Prevalence of Streptococcosis-related mortalities in farmed Nile tilapia (*Oreochromis niloticus*) at different life stages. *Egyptian Journal of Veterinary Sciences*, 54(5):949-964. doi:10.21608/ejvs.2023.207062.1494

19. Alazab, A., Sadat, A. and Younis, G. (2022). Prevalence, antimicrobial susceptibility, and genotyping of *Streptococcus agalactiae* in Tilapia fish (*Oreochromis niloticus*) in Egypt. *J Adv Vet Anim Res*., 9(1):95-103. doi:10.5455/javar.2022.i573

20. Lusta, M., Voronkova, O., Finkova, O. *et al*. (2023). Microbiological monitoring of antibiotic resistance of strains of *Streptococcus agalactiae* among pregnant women. *Regul Mech Biosyst*., 14(2):208-212. doi:10.15421/022331

21. Abd El Atty, M.H., Hafez, H.M. and Anwar, M.A. (2024). Comparative study between the Vitek2 and API 20 STEP regarding the identification of medically relevant *Streptococci*. *Ain Shams Med J*. 75(2):323-330. doi:10.21608/asmj.2024.259440.1194

22. Pepey, E., Taukhid, T., Keck, N. *et al*. (2021). Application of the FTA elute card coupled with visual colorimetric loop-mediated isothermal amplification for the rapid diagnosis of *Streptococcus agalactiae* in farmed tilapia (*Oreochromis niloticus*). *J Fish Dis*., 44(5):505-512. doi:10.1111/jfd.13337

23. Alsharksi, A.N., Sirekbasan, S., Gürkök-Tan, T. and Mustapha, A. (2024). From tradition to innovation: Diverse molecular techniques in the fight against infectious diseases. *Diagnostics*, 14(24):2876. doi:10.3390/diagnostics14242876

24. Chen, Y., Liu, L., Liu, J. *et al*. (2024). Serotype distribution, antimicrobial resistance, and molecular characterization of group B *Streptococcus* isolates from chinese pregnant woman. *The Journal of Maternal-Fetal & Neonatal Medicine*, 37(1). doi:10.1080/14767058.2023.2295805

25. Duodu, S., Ayiku, A., Adelani, A. *et al*. (2024). Serotype distribution, virulence and antibiotic resistance of *Streptococcus agalactiae* isolated from cultured tilapia *Oreochromis niloticus* in Lake Volta, Ghana. *Dis Aquat Organ*., 158:27-36. doi:10.3354/dao03780

26. Delannoy, C.M.J., Samai, H. and Labrie, L. (2021). *Streptococcus agalactiae* serotype IV in farmed tilapia. *Aquaculture*, 544:737033. doi:10.1016/j.aquaculture.2021.737033

27. Pervez, M.T., Hasnain, M.J., Abbas, S.H., Moustafa, M.F., Aslam, N. and Shah, S.S.M. (2022). A Comprehensive review of performance of next-generation sequencing platforms. *Biomed Res Int*., 2022(1). doi:10.1155/2022/3457806

28. Teatero, S., Ramoutar, E., McGeer, A. *et al*. (2016). Clonal complex 17 Group B *Streptococcus* strains causing invasive disease in neonates and adults originate from the same genetic pool. *Sci Rep*., 6(1):20047. doi:10.1038/srep20047

29. Poyart, C., Tazi, A., Réglier-Poupet, H. *et al*. (2007). Multiplex PCR assay for rapid and accurate capsular typing of group B *Streptococci*. *J Clin Microbiol*., 45(6):1985-1988. doi:10.1128/JCM.00159-07

30. Gigaulia, P., Sapre, S., Gontia-Mishra, I., Chouksey, P., Singh Rathore, G. and Singh, M. (2021). Introduction to PCR and Molecular Markers. *Professional Books Publisher.*, 95-121.

31. Heid, C.A., Stevens, J., Livak, K.J. and Williams, P.M. (1996). Real time quantitative PCR. *Genome Res*., 6(10):986-994. doi:10.1101/gr.6.10.986

32. Sanger, F., Air, G.M., Barrell, B.G. *et al*.

(1977).Nucleotide sequence of bacteriophage φX174 DNA. *Nature*, 265(5596):687-695. doi:10.1038/265687a0

33. Polonis, K., Blommel, J.H., Hughes, A.E.O., Spencer, D., Thompson J.A. and Schroeder, M.C. (2025). Innovations in short-read sequencing technologies and their applications to clinical genomics. *Clin Chem*., 71(1):97-108. doi:10.1093/clinchem/hvae173

34. Cuber, P., Chooneea, D., Geeves, C. *et al*. (2023). Comparing the accuracy and efficiency of third generation sequencing technologies, Oxford Nanopore Technologies, and Pacific Biosciences, for DNA barcode sequencing applications. *Ecol Genet Genom*., 28:100181. doi:10.1016/j.egg.2023.100181

35. Zhou, J., Zhang, L., Zhang, Y. *et al*. (2023). Analysis of molecular characteristics of CAMP-negative *Streptococcus agalactiae* strains. *Front Microbiol*., 14. doi:10.3389/fmicb.2023.1189093

36. Boyanov, V.S., Alexandrova, A.S. and Gergova, R.T. (2025). Diagnostic challenges and genotypic characteristics of CAMP-negative phenotype GBS isolates. *J Pure Appl Microbiol*., 19(4):2779-2787. doi:10.22207/JPAM.19.4.16

37. Ballard, M.B., Mercado-Evans, V., Marunde, M.G., Nwanosike, H., Zulk, J. and Patras, K.A. (2021). Group B *Streptococcus* CAMP factor does not contribute to interactions with the vaginal epithelium and is dispensable for vaginal colonization in mice. *Microbiol Spectr*., 9(3). doi:10.1128/Spectrum.01058-21

38. Nair, V.A., Sumithra, T.G., Reshma, K.J. and Sanil, N.K. (2023). Development of a novel multiplex-PCR technology for simultaneous detection of five major aquaculture pathogens. *Indian J Exp Biol*. Published online 2023. doi:10.56042/ijeb.v61i05.857

39. MacAulay, S., Ellison, A.R., Kille, P. and Cable, J. (2022). Moving towards improved surveillance and earlier diagnosis of aquatic pathogens: From traditional methods to emerging technologies. *Rev Aquac*., 14(4):1813-1829. doi:10.1111/raq.12674

40. Leigh, W.J., Zadoks, R.N., Costa, J.Z., Jaglarz, A. and Thompson, K.D. (2020). Development and evaluation of a quantitative polymerase chain reaction for aquatic *Streptococcus agalactiae* based on the *groEL* gene. *J Appl Microbiol*., 129(1):63-74. doi:10.1111/jam.14556

41. Abdallah, E.S.H., Metwally, W.G.M., Abdel-Rahman, M.A.M., Albano, M. and Mahmoud, M.M. ( 2024). *Streptococcus agalactiae* infection in Nile Tilapia (*Oreochromis niloticus*): A Review. *Biology (Basel)*. *Multidisciplinary Digital Publishing Institute (MDPI)*. 13(11). doi:10.3390/biology13110914

42. Zhang, D., Liu, Z., Ren, Y. *et al*. (2022). Epidemiological characteristics of *Streptococcus agalactiae* in tilapia in China from 2006 to 2020. *Aquaculture*., 549:737724. doi:10.1016/j.aquaculture.2021.737724

43. Fyrand, K., Xu, C. and Evensen, Ø. (2024). Characterization of *Streptococcus agalactiae* 1a isolated from farmed Nile tilapia (Oreochromis niloticus) in North America, Central America, and Southeast Asia. *Fish Shellfish Immunol*., 154. doi:10.1016/j.fsi.2024.109919

44. Sudpraseart, C., Wang, P. and Chen, S. (2021). Phenotype, genotype and pathogenicity of *Streptococcus agalactiae* isolated from cultured tilapia (*Oreochromis* spp.) in Taiwan. *J Fish Dis*., 44(6):747-756. doi:10.1111/jfd.13296

45. Preenanka, R., Safeena, M.P., Vidhya, B. and Sumithra, T. (2024). Impact of salinity on *Streptococcus agalactiae* and health parameters of *Oreochromis niloticus* during streptococcosis outbreaks. *The Microbe*, 5:100167. doi:10.1016/j.microb.2024.100167

46. Abdallah, E.S.H., Metwally, W.G.M., Abdel-Rahman, M.A.M., Albano, M. and Mahmoud, M.M. (2024). Isolation and characterization of *Streptococcus agalactiae* inducing mass mortalities in cultured Nile tilapia (*Oreochromis niloticus*) with trials for disease control using zinc oxide nanoparticles and ethanolic leaf extracts of some medicinal plants. *BMC Vet Res*., 20(1). doi:10.1186/s12917-024-04298-z

47. Rahman, M., Rahman, M., Monir, M. *et*

*al*. (2021). Isolation and molecular detection of *Streptococcus agalactiae* from popped eye disease of cultured tilapia and Vietnamese koi fishes in Bangladesh. *J Adv Vet Anim Res*., 8(1):1. doi:10.5455/javar.2021.h480

48. Kawasaki, M., Delamare-Deboutteville, J., Bowater, R.O. *et al*. (2018). Microevolution of *Streptococcus agalactiae* ST-261 from Australia indicates dissemination via imported tilapia and ongoing adaptation to marine hosts or environment. Published online 2018. doi:10.1128/AEM

49. Lusiastuti, A.M., Suhermanto, A., Hastilestari, B.R. *et al*. (2024). Impact of temperature on the virulence of *Streptococcus agalactiae* in Indonesian aquaculture: A better vaccine design is required. *Vet World*. Published online March 22, 2024:682-689. doi:10.14202/vetworld.2024.682-689

50. Hassan, H.U., Ali, A., Wattoo, J. *et al*. (2025). Effects of stocking density on growth performance, digestive enzyme activity, biochemical composition, and economic efficiency of a new strain of *Oreochromis niloticus* reared in cage culture system at Sindh Pakistan. *Food Sci Nutr*., 13(1). doi:10.1002/fsn3.4238

51. Hassan, S., Abdel-Rahman, M., Mansour, E.S. and Monir, W. (2020). Isolation, phenotypic characterization and antibiotic susceptibility of prevalent bacterial pathogens implicating the mortality of cultured Nile tilapia, *Oreochromis niloticus*. *Egyptian Journal for Aquaculture*., 10(1):23-43. doi:10.21608/eja.2020.25437.1017

52. Paredes-Trujillo, A. and Mendoza-Carranza M. (2022). A systematic review and meta-analysis of the relationship between farm management, water quality and pathogen outbreaks in tilapia culture. *J Fish Dis*., 45(10):1529-1548. doi:10.1111/jfd.13679

53. Djumanto, D., Peristiwady, T., Itani, G., Cadiz, P.N., Hasan, S.O.D. and Simanjuntak, C.P.H. (2025). Proceedings of the 3rd International Seminar on Fish and Fisheries Sciences (ISFFS 2025). Vol 51. (Djumanto D, Peristiwady T, Itani G, Cadiz PN, Hasan ODS, Simanjuntak CPH, eds.). *Atlantis Press International* BV; 2025. doi:10.2991/978-94-6463-942-1

54. Liu, Y., Huang, E., Xie, Y. *et al*. (2023). The effect of dietary lipid supplementation on the serum biochemistry, antioxidant responses, initial immunity, and motor pathway of juvenile tilapia (*Oreochromis niloticus*). *Fishes*., 8(11):535. doi:10.3390/fishes8110535

55. Naiel, M.A.E., Fawzy, R.M., Zaki, E.M., Abo-Elmatty, A.M.A., El-Tarabily, K.A. and Negm, S.S. (2024). Pumpkin seed extract attenuates toxic effects of ochratoxin-contaminated diets in terms of growth performance, antioxidant and immunological indices of Nile tilapia. *Aquac Rep*., 38:102300. doi:10.1016/j.aqrep.2024.102300

56. Hussain, G. (2025). Tilapia Farming: Breeding Plans, Mass Seed Production, and Aquaculture Technologies. *Academic Press*; 2025.

57. Brunt, M.W., Ritter, C., Renaud, D.L., LeBlanc, S.J. and Kelton, D.F. (2025). Perceived barriers to implementation of biosecurity best management practices for control of *Salmonella Dublin* on dairy farms: A focus group study. *J Dairy Sci*., 108(5):5233-5243. doi:10.3168/jds.2024-25676

58. Said, A.A., Reda, R.M., Metwally, M.M.M. and El-Hady, H.M.A. (2023). The contribution of *Coriandrum sativum* in enhancing *Oreochromis niloticus* rearing at sub-optimal temperatures: Effect on growth, health status, survival rate and resistance to *Aeromonas Veronii*. *BMC Vet Res*., 19(1):254. doi:10.1186/s12917-023-03809-8

59. Zornu, J., Tavornpanich, S., Brun, E. *et al*. (2023). Understanding tilapia mortalities and fish health management in Lake Volta: A systematic approach. *Front Sustain Food Syst*., 7. doi:10.3389/fsufs.2023.1249898

60. Liu, K., Zhang, L., Gu, X. *et al*. (2022). The prevalence, molecular characterization and antimicrobial resistance profiling of *Streptococcus agalactiae* isolated from clinical mastitis cases on large dairy farms in China. *Journal of Dairy Research*., 89(1):75-79. doi:10.1017/

S0022029922000152

61. Nhinh, D.T., Hoa, D.T., Trinh, T.T. *et al*. (2026). Distribution of serotypes, virulence genes, and antimicrobial resistance of *Streptococcus agalactiae* isolated in tilapia farms in Vietnam over 2022–2024. *Aquaculture*., 615:743636. doi:10.1016/j.aquaculture.2026.743636

62. Maniyappan, K.K., Girijan, S.K., Krishnan, R., Gopan, A. and Pillai, D. (2025). Assessing multi-drug resistance in *Streptococcus agalactiae* infecting farmed Nile Tilapia: Findings from Kerala, India. *Microb Pathog*., 205:107666. doi:10.1016/j.micpath.2025.107666

63. Melo-Bolívar, J.F., Ruiz Pardo, R.Y., Hume, M.E. *et al*. (2019). Establishment and characterization of a competitive exclusion bacterial culture derived from Nile tilapia (*Oreochromis niloticus*) gut microbiomes showing antibacterial activity against pathogenic *Streptococcus agalactiae*. *PLoS One*, 14(5):e0215375. doi:10.1371/journal.pone.0215375

64. Reverter, M., Sarter, S., Caruso, D. *et al*. (2020). Aquaculture at the crossroads of global warming and antimicrobial resistance. *Nat Commun*., 11(1):1870. doi:10.1038/s41467-020-15735-6

65. Lannes-Costa, P.S., Oliveira, J.S.S., Silva Santos, G. and Nagao, P.E. (2021). A current review of pathogenicity determinants of *Streptococcus* sp. *J Appl Microbiol*., 131(4):1600-1620. doi:10.1111/jam.15090

66. Ager, E.O., Carvalho, T., Silva, E.M., Ricke, S.C. and Hite, J.L. (2023). Global trends in antimicrobial resistance on organic and conventional farms. *Sci Rep*., 13(1):22608. doi:10.1038/s41598-023-47862-7

67. Shivaee, A., Kazemi, F., Navidifar, T. *et al*. (2025). Global trends in macrolide and lincosamide resistance in *Streptococcus* species: A comprehensive systematic review and meta-analysis. *BMC Infect Dis*., 25(1):1577. doi:10.1186/s12879-025-11884-5

68. Fang, H., Li, D., Wei, X. *et al*. (2025). Global dynamics of *Streptococcus agalactiae* antibiotic resistance: Past, present, and future. *J Hazard Mater*., 499:140199. doi:10.1016/j.jhazmat.2025.140199

69. Abdulaziz, A. and Vikram, A. (2024). A review of group b *Streptococcus* (GBS) in young adults: Emerging concerns beyond the neonatal population. *Journal of medical and clinical case reports*. Published online August 17, 2024. doi:10.61615/JMCCR/2024/AUG027140817

70. Tang, K.W.K., Millar, B.C. and Moore, J.E. (2023). Antimicrobial Resistance (AMR). *Br J Biomed Sci*. *Institute of Biomedical Science (IBMS)*, 80. doi:10.3389/bjbs.2023.11387

71. Lulijwa, R., Rupia, E.J. and Alfaro, A.C. (2020). Antibiotic use in aquaculture, policies and regulation, health and environmental risks: A review of the top 15 major producers. *Rev Aquac*., 12(2):640-663. doi:10.1111/raq.12344

72. Milijasevic, M., Veskovic-Moracanin, S., Babic Milijasevic, J., Petrovic, J. and Nastasijevic, I. (2024). Antimicrobial resistance in aquaculture: Risk mitigation within the one health context. *Foods*, 13(15):2448. doi:10.3390/foods13152448

73. Garza, M., Mohan, C.V., Brunton, L., Wieland, B. and Häsler, B. (2022). Typology of interventions for antimicrobial use and antimicrobial resistance in aquaculture systems in low- and middle-income countries. *Int J Antimicrob Agents*., 59(1):106495. doi:10.1016/j.ijantimicag.2021.106495

74. Paul, R. AMR in Aquaculture: Enhancing Indian Shrimp Exports through Sustainable Practices and Reduced Antimicrobial Usage. *Research paper* 209, South Centre, Geneva

75. Bondad-Reantaso, M.G., MacKinnon, B., Karunasagar, I. *et al*. (2023). Review of alternatives to antibiotic use in aquaculture. *Rev Aquac*., 15(4):1421-1451. doi:10.1111/raq.12786

76. Dien, L.T., Ky, L.B., Huy, B.T. *et al*. (2022). Characterization and protective effects of lytic bacteriophage pAh6.2TG against a

pathogenic multidrug-resistant *Aeromonas hydrophila* in Nile tilapia (*Oreochromis niloticus*). *Transbound Emerg Dis*., 69(4). doi:10.1111/tbed.14321

77. Pholchamat, S., Vialle, R., Luang-In, V. *et al*. (2024). Evaluation of the efficacy of MONTANIDE™ GR01, a new adjuvant for feed-based vaccines, on the immune response and protection against *Streptococcus agalactiae* in oral vaccinated Nile tilapia (*Oreochromis niloticus*) under laboratory and on-farm conditions. *Fish Shellfish Immunol*., 149:109567. doi:10.1016/j.fsi.2024.109567

78. Hayat, M., Mohd Yusoff, M.S., Samad, M.J. *et al*. (2021). Efficacy of feed-based formalin-killed vaccine of *Streptococcus iniae* stimulates the gut-associated lymphoid tissues and immune response of red hybrid tilapia. *Vaccines (Basel)*. 9(1):51. doi:10.3390/vaccines9010051

79. Queiróz, G.A., de, Silva TMF and Leal, C.A.G. ( 2024). Duration of protection and humoral immune response in Nile tilapia (*Oreochromis niloticus* L.) vaccinated against *Streptococcus agalactiae*. *Animals*, 14(12):1744. doi:10.3390/ani14121744

80. Piamsomboon, P., Jantrakajorn, S., Tanpichai, P. and Wongtavatchai, J. (2024). Implementation of formalin-inactivated *Streptococcus agalactiae* vaccine in Nile *tilapia Oreochromis niloticus* broodstock: Efficient vaccination regime, antibody response, and immune-related genes. *Egypt J Aquat Res*., 50(4):558-565. doi:10.1016/j.ejar.2024.09.006

81. Delphino, M.K.V.C., Barone, R.S.C., Leal, C.A.G, Figueiredo, H.C.P., Gardner, I.A. and Gonçalves, V.S.P. (2019). Economic appraisal of vaccination against *Streptoccocus agalactiae* in Nile tilapia farms in Brazil. *Prev Vet Med*., 162: 131-135. doi:10.1016/j.prevetmed.2018.12.003

82. Amenyogbe, E., Chen, G., Wang, Z., Huang, J., Huang, B. and Li, H. (2020). The exploitation of probiotics, prebiotics and synbiotics in aquaculture: present study, limitations and future directions: A review. *Aquaculture International*., 28(3):1017-1041. doi:10.1007/s10499-020-00509-0

83. Murray, C.J., Ikuta, K.S., Sharara, F. *et al*. (2022). Global burden of bacterial antimicrobial resistance in 2019: A systematic analysis. *The Lancet*., 399(10325):629-655. doi:10.1016/S0140-6736(21)02724-0

84. Gruber, C., Ocelova, V., Kesselring, J.C. and Wein, S. (2025). Phytogenic feed additives as a sustainable alternative to antibiotics: enhancing growth and disease resistance in Nile tilapia (*Oreochromis niloticus*). *Animals*, 15(3):380. doi:10.3390/ani15030380

85. MacKinnon, B., Debnath, P.P., Bondad-Reantaso, M.G., Fridman, S., Bin, H. and Nekouei, O. (2023). Improving tilapia biosecurity through a value chain approach. *Rev Aquac*., 15(S1):57-91. doi:10.1111/raq.12776

86. Dauda, A.B., Nababa, A.S., Oshoke, J.O. *et al*. (2024). Antibiotics use and awareness of risks associated with antimicrobial resistance among fish farmers in Katsina State, Nigeria. Published online 2024. doi:10.2139/ssrn.4767228

87. Frumence, G., Mboera, LE.G., Sindato, C. *et al*. (2021). The governance and implementation of the national action plan on antimicrobial resistance in Tanzania: A qualitative study. *Antibiotics*, 10(3):273. doi:10.3390/antibiotics10030273