Egyptian Journal of Aquatic Biology & Fisheries
Zoology Department, Faculty of Science,
Ain Shams University, Cairo, Egypt.
ISSN 1110 – 6131
Vol. 26(5): 39 – 51 (2022)
www.ejabf.journals.ekb.eg

Tilapia Lake Virus (TiLV): the aetiological agent of Tilapia Lake Virus Disease (TiLVD): A review article

Saleh Ahmed Rabeh
National Institute of Oceanography and Fisheries, Cairo, Egypt.
dr.salehrabeh@hotmail.com

ARTICLE INFO
Article History:
Received: July 23, 2022
Accepted: Aug. 13, 2022
Online: Sept. 9, 2022

Keywords:
Tilapia Lake Virus (TiLV), Aquaculture, Tilapia Lake Virus, Disease (TiLVD)

ABSTRACT

Due to their tolerance for high-density aquaculture and relative disease resistance, Tilapia is an important global food source. The economic impact of tilapia on worldwide trade is estimated at $7.5 billion U.S. dollars (USD) annually. Since 2009, tilapia aquaculture has been threatened by mass die-offs in farmed fish in Israel and Ecuador by a novel orthomyxo-like virus called Tilapia Lake Virus (TiLV). Subsequently, the disease was reported in tilapia culture regions worldwide that pose a threat to the global tilapia industry, which not only provides inexpensive dietary protein but also is a major employer in the developing world. Epidemiological surveys indicated that 37% of Egyptian fish farms were affected in 2015 with an average mortality rate of 9.2% and a potential economic impact of around US$ 100 million. Although two published papers claimed that TiLV is present in Egypt, it is worth mentioning that such results have not been confirmed yet. The issue is of interest because tilapia is a popular fish representing many Egyptian exports.

INTRODUCTION

Tilapines, comprising more than 100 species, are the second most import group of farmed fish worldwide after carp. Worldwide trade of tilapia has huge economic impact as an industry that brings in approximately 4.5 million metric tons of product and $7.5 billion annually (**FAO, 2014**). This industry is a major employer in China, Egypt, Thailand, Philippines, Indonesia, Laos, Costa Rica, Colombia, Ecuador and Honduras. The United States as the lead importer (**Bacharach *et al.*, 2016**). According to **FAO (2017)** global production of tilapia is estimated at 6.4 million metric tons (MMT), with the top three producers in 2015 being the People's Republic of China (1.78 MMT), Indonesia (1.12 MMT) and Egypt (0.88 MMT). Bangladesh, Vietnam and the Philippines are other leading producers. Also, Tilapia are very important for ecological systems (**Eyngor *et al.*, 2014**) as they are beneficial in algae and mosquito control, habitat maintenance for shrimp farming and an important wild capture species (**Bacharach, *et al.*, 2016**). Thus, the spread of TiLV has global impact in both economic losses to farmers and fishers, cause significant mortality up to 90% (**Dong *et al.*, 2017**) and ecological settings. Tilapia lake virus (TiLV), an emerging disease of tilapia, was first reported in Israel in 2011 (**Eyngor *et al.*, 2014**). Subsequently, the disease was reported in tilapia

culture regions worldwide, including Asia, Africa, South America **(OIE, 2018)** and most recently in the USA in 2019 (**Ahasan *et al*., 2020**).

The disease associated with TiLV infection is currently known under two different names, syncytical hepatitis of tilapia (SHT) as first referred to by **Ferguson *et al*. (2014)** and tilapia lake virus disease (TiLVD) as in the OIE technical disease card (**OIE, 2017**). Duration of survival outside the host has not been determined; however horizontal, waterborne spread has been demonstrated under experimental conditions (**Eyngor *et al*., 2014**).

This review summarizes the information currently available on TiLV. This included the aetiological agent, host factors, disease patterns and risk factors, pathology and diagnostic tests, and socio economic impact.

1. Classification of TiLV

Tilapia lake virus (TiLV) or *Tilapia tilapinevirus*, is a negative-sense, single-stranded RNA virus (**Eyngor *et al*., 2014**). It belongs to Group V of the Baltimore Classification System of viruses. It is the only species in the monotypic genus Tilapinevirus, which in turn is the only genus in the family Amnoonviridae. **Pulido *et al*. (2019)** proposed two genetic clades of TiLV (Israeli and Thai clades) based on multilocus sequence phylogenetic analysis (MLSA) of 8305 nucleotides of 5 TiLV genomes.

2. Discovery and geographical distribution

A novel RNA virus was first discovered and identified in 2014 and termed tilapia lake virus (TiLV) when Kinneret Lake experienced a major noticeable decline in tilapia catch quantities (**Eyngor *et al*., 2014**).

Subsequently, scientific publications have reported identification of TiLV from samples collected in Ecuador (**Ferguson *et al*., 2014** and **Bacharach *et al*., 2016**), Egypt (**Fathi *et al*., 2017** and **Nicholson *et al*., 2017**), Colombia (**Tsofack *et al*., 2017**), Thailand (**Dong *et al*., 2017** and **Surachetpong *et al*., 2017**), India (**Behera *et al*., 2018**), Indonesia (**Koesharyani *et al*., 2018**), and Malaysia (**Amal *et al*., 2018**). Currently TiLV has been reported in 16 countries **(Fig. 1)**. This number is continuing to rise due to improved diagnostic assays and surveillance activities around the world (**Surachetpong *et al*., 2020**). A partial genome from Thailand showed relatively high variation (around 97% nucleotide identities) to strains from Israel (**Dong *et al*., 2017**). Reports of mortality in tilapia in Ghana and Zambia in 2016 have not been attributed to TiLV but the available information did not indicate that the presence of the virus has been investigated. Thus, a lack of thorough investigation of all mortality incidents means that the geographic distribution of TiLV may be wider than currently.

Fig. 1. Geographical distribution of TiLV-infected countries. Sixteen countries across four different continents are affected by the virus. Red color shows TiLV-infected countries (After **Surachetpong *et al*., 2020**)

In Egypt, tissue samples from 7 farms affected by 'summer mortality' were tested. Sequence analysis yielded a TiLV sequence with 93% homology to the published TiLV sequence described from Israel (**Fathi *et al*., 2017** and **Nicholson *et al*., 2017**).

3. Shape, size and structure

Electron microscopy has revealed tilapia lake virus to be an enveloped icosahedral particle (**Eyngor et al., 2014**) that is 55–100 nm in diameter (**Jansen *et al*.,2019**) **(Figs. 2&3)**.TiLV is described to be an orthomyxo-like virus it may share similar structural features, like surface glycoproteins and a helical nucleocapsid (**Bacharach *et al*., 2016**).

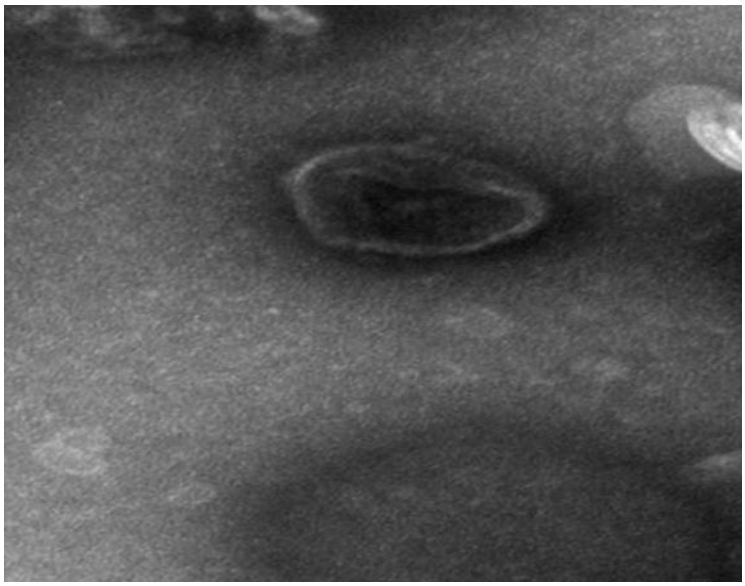

Fig. 2. Transmission electron micrograph of a TiLV particle shows envelope virion at the size of 80 nm

(After Surachetpong *et al*., 2020)

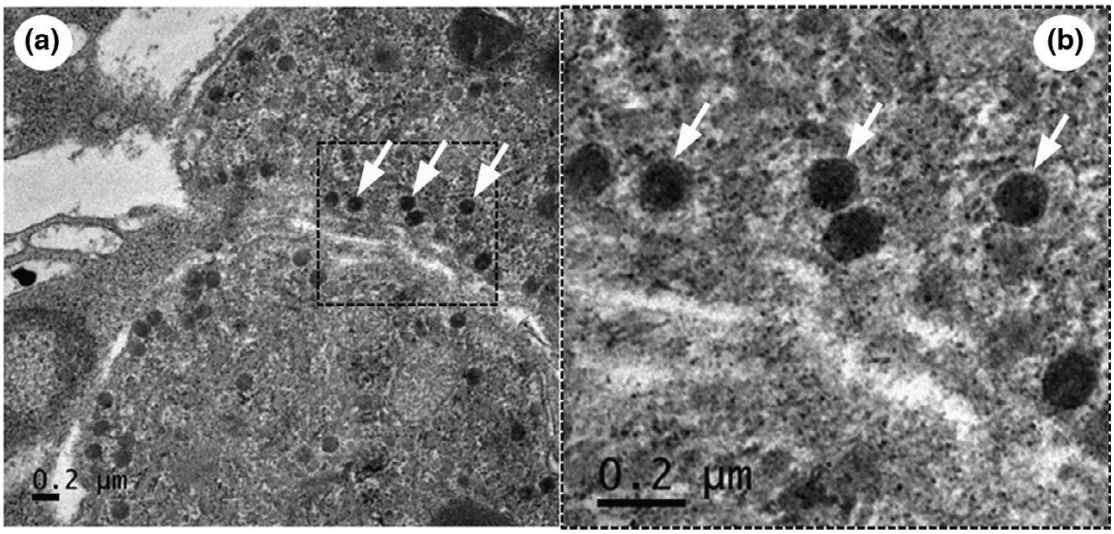

Fig.3. Transmission electron micrographs of TiLV-infected fish liver tissue showing cytoplasmic viral particles (white arrows) at low (a) and high (b) magnification. The electron micrograph in (b) is a magnification of the box outlined in black in (a), and the same 3 example virions (80–90 nm diameter) are indicated with white arrows in both electron micrographs (Images by H.T. Dong).

The RNA strand includes ten viral genomic segments with open reading frames (ORF) which encode for ten proteins (**Surachetpong *et al.*, 2017**). The genome's total size is 10.323kb and each of the ten segments range in size from 465 to 1,641 nucleotides (**Bacharach *et al.*, 2016**). The first segment is the largest and has minimal homology with the influenza C virus PB1 subunit. The remaining nine segments show no homology with other known viruses, though their genome organization is consistent with that of other orthomyxoviruses (**Eyngor *et al.*, 2014**). Comparative genome analysis of TiLV from various global populations of tilapia has indicated that the genome segments have geographically influenced genetic variation (**Dong *et al.*, 2017**). Thirteen nucleotides that are present in all segments are also included in the TiLV 5' and 3' noncoding termini, which give TiLV resemblance to two other orthomyxoviruses: Isavirus and Thogoto (**Bacharach *et al.*, 2016**).

4. Transmission

4.1. Reservoir

Both wild and farmed infected fish populations are the only established reservoirs of infection. The original source of TiLV is not known.

4.2. Modes of transmission

The virus has been found in fresh and preserved tilapia and found to be transmitted through direct horizontal transmission by cohabitation or transfer of live aquatic animals (**Tsofack *et al.*, 2017**) **(Fig.4)**. It was suggested that the eye, brain and liver are likely to contain highest concentrations of TiLV and thus solid and liquid waste are likely to be contaminated (**OIE, 2018**).

TiLV enters fish through oral or direct gills exposure and distributes systemically to other internal organs (spleen, liver, kidney and gonads). The virus spreads horizontally and vertically via infected mucus or faeces, and fertilized eggs to other naïve fish (**Surachetpong *et al.*, 2020**). Faeces of Tilapia were suggested as a possible source of horizontal transmission and non-lethal mode of sampling for TiLV detection (**Pierezan *et al.*, 2019**).

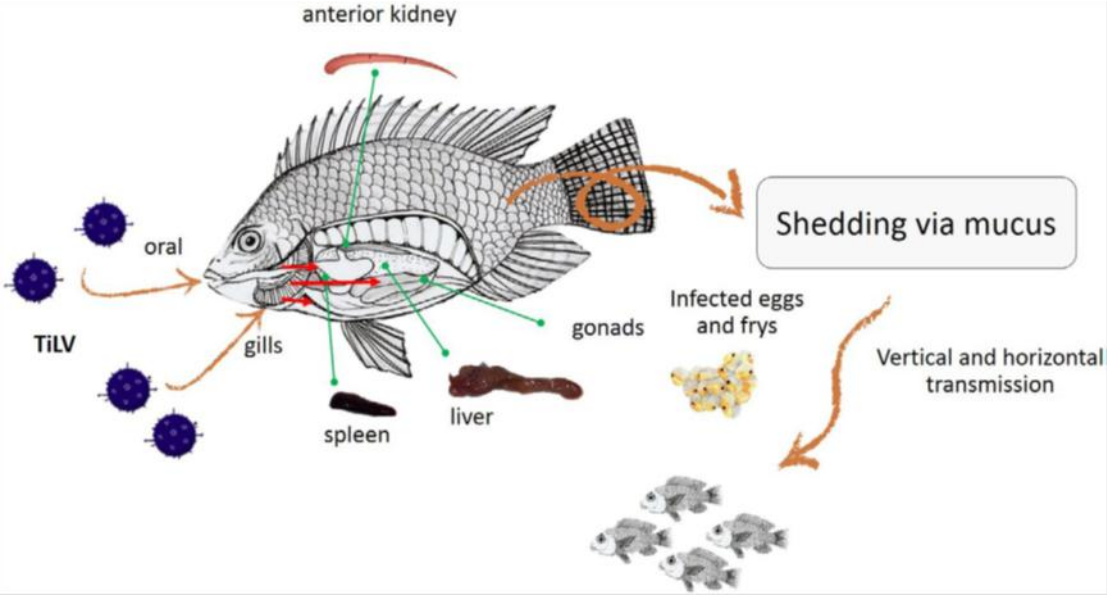

Fig. 4. Mechanism of TiLV pathogenesis spread and virus

5. Risk factors

Disease has been associated with transfer between farms and thus may be associated with stress (**Ferguson *et al.*, 2014** and **Dong *et al.*, 2017**). Other risk factors (temperature, salinity, etc.) have not been identified as potential risk factors.

6. Control

Till now there is no evidence that there are practices to limit viral spread in an infected farm (**OIE, 2018**). However, restriction of the movement of live tilapines between farms or fisheries is thought to limit the spread of the viral disease to new species, as well as generic biosecurity measures to minimise spread via clean practices and sanitizing equipment in these areas should also be implemented. Due to their lipid membrane, viral particles have been found to be sensitive to organic solvents (ether and chloroform) (**Eyngor *et al.*, 2014**).

7. Host range

7.1. Susceptible and resistant species

Mortalities attributed to TiLV have been observed in wild tilapia *Sarotherodon (Tilapia) galilaeus*, farmed Tilapia *Oreochromis niloticus* and commercial hybrid Tilapia (*O. niloticus* X *O. aureus*) (**Ferguson *et al.*, 2014**; **Eyngor *et al.*, 2014** and **Bacharach *et al.*, 2016**). To date only tilapines have been shown to be susceptible. It is possible that other species will be found to be susceptible.

A study of **Jaemvimol *et al*. (2019)** revealed the possibility of disease induction in giant gourami (Osphronemus goramy, while failed to induce disease in snakeskin gourami (*Trichogaster pectoralis*), iridescent shark (*Pangasianodon hypophtthalmus*), walking catfish (*Clarias macrocephalus*), striped snake head fish (*Channa striata*), climbing perch (*Anabas testudineus*), and common carp (Cyprinus carpio) by injection of TiLV.

Co-cultivated grey mullet (*Mugil cephalus*) and carp (*Cyprinus carpio*) have not shown mortality during disease outbreaks in Israel (**Eyngor *et al*., 2014**). Similarly, cocultivated grey mullet and thin-lipped mullet (*Liza ramada*) were found to be unaffected during Egyptian outbreaks (**Fathi *et al*., 2017**) and cocultivated Indian Major Carps; rohu (*Labeo rohita*), catla (*Catla Catla*), mrigal (*Cirrhinus mrigala*)), milk fish (*Chanos chanos*) and pearl spot (*Etroplus suratensis*) were unaffected in India (**Behera *et al*., 2018**). However, TiLV has also been detected in other fish species without clinical signs, such as wild river carp (*Barbonymus schwanenfeldii*) (**Abdullah *et al*., 2018**) and giant gourami (*Osphronemus goramy*) (**Chiamkunakorn *et al*., 2019**).

There is some evidence that certain genetic strains of tilapia are resistant. One strain of tilapia (genetically male tilapia) incurred a significantly lower level of mortality (10-20%) compared with other strains (**Ferguson *et al*., 2014**).

Waiyamitra *et al*. (2021) investigated the susceptibility of Mozambique tilapia (*O. mossambicus*) to TiLV infection by comparing TiLV infection in red hybrid and Mozambique tilapia, as well as comparing the infectivity and biology of the infection in Mozambique tilapia after exposure to both low and high concentrations of TiLV. Regardless of the challenge dose, TiLV can cause clinical signs and mortality in Mozambique tilapia, though to different degrees of virus replication and severity.

7.2. Affected life stage

Fingerlings were mainly affected during the outbreak reported by **Ferguson *et al*. (2014)** and **Dong *et al*. (2017)**. The latter reported approximately 90% mortality in red tilapia fingerlings within one months of stocking into cages. Mortality just over 9% in medium to large sized Nile perch was notes by **Fathi *et al*. (2017)**. Other reports have not commented on different levels of mortality by life stage (**Eyngor *et al*., 2014**).

8. Clinical sings

8.1. Host tissues and infected organs

The eyes, brain and liver are the main organs where pathology is observed (**Eyngor *et al*., 2014**).

8.2. Gross observations and macroscopic lesions

Gross lesions included ocular alterations, including opacity of the lens and in advanced cases ruptured lens. Other lesions included skin erosions, haemorrhages in the leptomeninges and congestion of the spleen (**Eyngor *et al*., 2014**).

8.2. Microscopic lesions and tissue abnormality

Histologic lesions have been observed in the brain, eye and liver (**Eyngor *et al*., 2014**). Lesions in the brain included oedema, focal haemorrhages in the leptomeninges,

and capillary congestion in both the white and grey matter and neural degeneration. Foci of gliosis and occasional perivascular cuffs of lymphocytes have been detected. Ocular lesions included ruptured lenticular capsule and cataractous changes. Foci of hepatocellular swelling were observed. The spleen was hyperplastic, with proliferating lymphocytes. Melano-macrophage centers (MMCs) were increased in size and number in both the liver and the spleen. The presence of an orthomyxo-like virus within diseased hepatocytes was confirmed by Transmission electron microscopy and thus confirmed earlier reports of syncytial hepatitis (**Del-Pozo *et al*., 2016**).

9. Diagnostic methods

9.1. Definition of suspicion

High levels of mortality in tilapine species, along with ocular alterations (opacity of the lens or more severe pathology), should be considered suspicious of TiLV. On post-mortem skin erosions, haemorrhages in the leptomeninges and moderate congestion of the spleen and kidney may be observed.

9.2. Presumptive test methods

According to **Eyngor *et al*. (2014)** TiLV can be cultured in primary tilapia brain cells or in an E-11 cell line, inducing a cytopathic effect at 5-10 days. The optimal conditions for culturing TiLV were described by **Tsofack *et al*. (2017)**.

9.3. Confirmatory test methods

A not fully validated test was carried out by (**Eyngor *et al*., 2014**) where a PCR primer set has been designed and a reverse transcriptase (RT) PCR has been developed. **Tsofack *et al.* (2017)** has published a more highly sensitive, nested RT-PCR and is suitable for the detection of TiLV in clinical cases. Recently a semi-nest RT-PCR with improved detection sensitivity (7.5 viral copies per reaction) over the nested RT-PCR, has been published (**Dong *et al*., 2017**).

The presently available one step reverse transcription (RT)-PCR assay **(Eyngor *et al*., 2014)**, although highly specific, is of limited sensitivity when applied to clinical samples. Accordingly, **Tsofack *et al*. (2017)** described a highly sensitive, nested RT-PCR and semi-nested RT-PCR **(Dong *et al*., 2017)** assay for TiLV detection from clinical specimens. However, validation of these three methods is suggested (**OIE, 2018**).

Piamsomboon & Wongtavatchai (2021) compared the TiLV detection using these three RT-PCR methods in fish species from the water source where TiLV has been detected. They revealed that Semi-nested RT-PCR detected 9 positives, while the nested and one-step RT-PCR produced 7 and 4 positives, respectively.

Recently, **Taengphu *et al*. (2020)** recommend the use of their newly established segment 1 PCR method as an alternative tool for TiLV diagnosis and in active surveillance programs in the laboratories where quantitative PCR is not accessible. Compared the TiLV detection using these three RT-PCR methods in fish species from the water source where TiLV has been detected.

As recommended by the World Organization for Animal Health (OIE), the confirmatory test methods for TiLV detection are RT-PCR (**Eyngor *et al*., 2014**), nested RT-PCR (**Tsofack *et al*., 2017**). In addition, the TiLV detection was extended to farmed

barramundi locating in close proximity to the TiLV-PCR positive wild fish. Several studies have described specific and sensitive PCR based diagnostic of TiLV disease **(Table 1)**.

Table 1. Diagnosis methods of TiLV

Diagnosis method	References
PCR	**Mugimba *et al*., 2018**
RT- PCR	**Dong *et al*., 2017; Fathi *et al*., 2017; Nicholson *et al*., 2017; Surachetpong *et al*., 2017; Tsofack *et al*., 2017; Amal *et al*., 2018**
Nested RT- PCR	**Tsofack *et al*., 2017**
Semi-nested PCR	**Koesharyani *et al.* (2018)**
SYBR green-based RT-qPCR	**Tattiyapong, *et al*.,2017**
TaqMan probe-based RT-qPCR	**Waiyamitra *et al*. 2018**
LAMP - RT	**Phusantisampan, 2019**
Cell line (E-11)	**Eyngor *et al*., 2014;Tsofack *et al*., 2017; Soni *et al.*, 2018**
Cell culture	**Mugimba *et al*., 2018**
ELISA	**Jansen *et al*., 2019;Hu *et al*., 2020; Tattiyapong *et al*., 2020**
iELISA	**Hu *et al*., 2020; Tattiyapong *et al*., 2020**
ISH	**Dong *et al*. 2017**
UHTS	**Bacharach *et al*. 2016**
Nanopore sequencing	**Delamare-Deboutteville *et al*., 2021**

ELISA enzyme-linked immune sorbent assay, iELISA Indirect enzyme-linked immunosorbent assay, RT-PCR- Reverse Transcriptase polymerase chain reaction, TEM- Transmission electron microscopy, UHTS - Unbiased high-throughput sequencing, MS- Mass spectrometry, ISH- in situ hybridization, RT-qPCR- Reverse Transcriptase quantitative polymerase chain reaction RT-LAMP- Reverse transcription loop-mediated isothermal amplification.

10. Control methods

Currently, no published methods have been shown to be effective in limiting the impact of an outbreak on an infected farm. It has been suggested that breeding for resistance or the development of a vaccine may offer the long term prospects for managing the disease (**Ferguson *et al*., 2014**). A breeding program would need to select and test a range of different strains of tilapia with a view to finding those least susceptible.

10.1. Treatment

- Using approved chemicals to prevent fish infected with TiLV from secondary bacterial and parasitic infections.
- Using common disinfectants against Tilapia Lake Virus (TiLV) **(Table, 2)**.
- Using of immune-modulators and immune-stimulators to enhance the immune system of the fish.

10.2. Vaccination

As TiLV is affecting fingerlings (fish in the 10, 20, 30 gram range) some of these fish are too small for injection or are labour intensive for injection, so the commercial solution will need to be an immersion vaccine, or an oral vaccine.

Table 2. Common disinfectants against Tilapia Lake Virus (TiLV), After **Aich *et al*. (2021)**

Disinfectant	Dose (ppm)	Effective temperature (^{O}C)	Duration of exposure (min)	Effect on Virus	Reference
Virkon®	5000	28	1	Virus inactivation	**Jaemwimol *et al*., 2019**
H_2O_2	300	28	10	Veridical effect	
Iodine	2.5	28	10	Veridical effect	
Formaline	80	28	60	Veridical effect	
NaOCl	10	28	10	Veridical effect	
PVD	50a	-	30	Veridical effect	**Soto *et al*., 2019**
Chlorine	20	-	30	Veridical effect	**Soto *et al*., 2019**

[a] Free iodine concentration

CONCLUSION

- TiLVD can be considered as a transboundary fish disease (TFD).
- Development of effective vaccine against this virus is a must.
- Extensive work may be directed in the line of developing SPF tilapia's or selectively bred resistant tilapia.
- TiLV segment 1 is a promising gene candidate for studying the genetic diversity of this virus
-The currently available information on TiLV highlight the importance of international collaboration for knowledge generation regarding the virus itself and its implications.

REFERENCES

Abdullah, A.; Ramly, R.; Ridzwan, M.S.M.; Sudirwan, F.; Abas, A.; Ahmad, K.; Murni, M. and Kua, B.C. (2018). First detection of tilapia lake virus (TiLV) in wild river carp (*Barbonymus schwanenfeldii*) at Timah Tasoh Lake, Malaysia. Journal of Fish Diseases, 41(9): 1459-1462.

Ahasan, M.S.; William, K.; Cem, G.; Brenda, P.; Win, S.; Pamela, N.; Al-Hussinee, L.; Subramaniam, K. and **Waltzek, T.B**. (2020). Genomic characterization of Tilapia lake virus isolates recovered from moribund Nile Tilapia (*Oreochromis niloticus*) on a farm in the United States. Microbiology Resource Announcements, 9(4): 1-2.

Aich, N.; Paul, A.; Choudhury, T.G. and **Saha, H**. (2021). Tilapia Lake Virus (TiLV) disease: Current status of understanding. Aquaculture and Fisheries, **7**(1): 7 – 17.

Amal, M.N.A.; Koh, C.B.; Nurliyana, M.; Suhaiba, M.; Nor-Amalina, Z.; Santha, S. ;Diyana-Nadhirah, K.P.; Yusof, M.T. ;Ina-Salwany, M.Y. and **Zamri-Saad, M**. (2018**).** A case of natural co-infection of tilapia lake virus and Aeromonas veronii in a Malaysian red hybrid tilapia (**Oreochromis niloticus** x *O. mossambicus*) farm experiencing high mortality. Aquaculture, 485: 12-16.

Bacharach, E.; Mishra, N.; Briese, T.; Zody, M.C.; Tsofack, J.E.K.; Zamostiano, R.; Berkowitz, A.; Ng, J.; Nitido, A.; Corvelo, A.; Toussaint, N.C.; Abel Nielsen, S.C.; Hornig, M.; Del Pozo, J.; Bloom, T.; Ferguson, H.; Eldar, A. and **Lipkin, W.I**. (2016). Characterization of a novel orthomyxo-like virus causing mass die-offs of Tilapia. mBio., **7**(2): e00431-16.

Behera, B.K.; Pradhan, P.K.; Swaminathan, T.R.; Sood, N.; Paria, P.; Das, A. ;Verma, D.K.; Jena, J.K.; Lal, K.K.; Yadav, M.K.; Sood, N.; Parida, P.K.; Pradhan, P.K.; Paria, P.; Kumar, R. and **Swaminathan, T.R**. (2018). Emergence of tilapia lake virus associated with mortalities of farmed Nile Tilapia, *Oreochromis niloticus* (Linnaeus 1758) in India. Aquaculture, 484: 168-174.

Chiamkunakorn, C.; Machimbirike, V.I.; Senapin, S.; Khunrae, P.; Dong, H.T. and **Rattanarojpong, T**. (2019). Blood and liver biopsy for the non-destructive screening of tilapia lake virus. Journal of Fish Diseases, 42(11): 1629-1636.

Delamare-Deboutteville, J.; Taengphu, S.; Gan, H.M.; Kayansamruaj, P.; Debnath, P.P.; Barnes , A.; Wilkinson, S.; Kawasaki, M.C.; Mohan, V.; Senapin, S. and **Dong, H.T.** (2021). Rapid genotyping of Tilapia Lake Virus (TiLV) using nanopore sequencing. J. Fish Dis., 00: 1-12.

Del-Pozo, J.; Mishra, N.; Kabuusu, R.; Cheetham, S.; Eldar, A.; Bacharach, E.; Lipkin, W.I. and **Ferguson, H.W.** (2016). Syncytial hepatitis of Tilapia (*Oreochromis niloticus* L.) is associated with orthomyxovirus- like virions in hepatocytes. Veterinary Pathology, 54(1): 154-160. https://doi.org/10.1177/0300985816658100

Dong, H.T.; Siriroob, S.; Meemetta, W.; Santimanawong, W.; Gangnonngiw, W.; Pirarat, N.; Khunrae, P.; Rattanarojpong, T.; Vanichviriyakit, R. and **Senapin, S. (**2017). Emergence of tilapia lake virus in Thailand and an alternative semi-nested RT-PCR for detection. Aquaculture, 476: 111-118.

Eyngor, M.; Zamostiano, R.; Tsofack, J.E.K.; Berkowitz, A.; Bercovier, H.; Tinman, S.; Lev, M.; Hurvitz, A.; Galeotti, M.; Bacharach, E. and **Eldar, A.** (2014). Identification of a novel RNA virus lethal to tilapia. Journal of Clinical Microbiology, 52(12): 4137-46.

FAO (2014). The state of world fisheries and aquaculture. Food and Agriculture Oraganization of the United Nations (Vol. 2014).https://doi.org/92-5-105177-1

FAO (2017). Global Food and Agriculture Organization of the United Nations (FAO). http://www.fao.org/fishery/statistics/global-production/en

Fathi, M.; Dickson, C.; Dickson, M.; Leschen, W.; Baily, J.; Muir, F.; Ulrich, K. and **Weidmann, M.** (2017). Identification of Tilapia Lake Virus in Egypt in Nile tilapia affected by 'summer mortality' syndrome. Aquaculture, 472: 430-432.

Ferguson, H.W.; Kabuusu, R.; Beltran, S.; Reyes, E.; Lince, J.A. and **Del Pozo, J.** (2014). Syncytial hepatitis of farmed tilapia, *Oreochromis niloticus* (L.): a case report. Journal of Fish Diseases, 37: 583-589.

Jaemwimol, P.; Sirikanchana, K.; Tattiyapong, P.; Mongkolsuk, S. and **Surachetpong, W.** (2019). Veridical effects of common disinfectants against tilapia lake virus. Journal of Fish Diseases, 42(10): 1383-1389.

Jansen, M.D.; Dong, H.T. and **Mohan, C.V. (2019).** Tilapia Lake Virus: A threat to the global tilapia industry. Reviews in Aquaculture, **11**(3): 725-739.

Hu, H., Zeng, W.; Wang, Y.; Wang, Q.; Bergmann, S. M.; Yin, J. and **Liu, C.** (2020). Development and application of a recombinant protein-based indirect ELISA for detection of anti-tilapia lake virus IgM in sera from tilapia. Aquaculture, 520, 734-756.

Koesharyani, I.; Gardenia, L.; Widowati, Z.; Khumaira, K. and **Rustianti, D.** (2018). Studi kasus infeksi Tilapia lake virus (TiLV) pada ikan nila (*Oreochromis niloticus*). Jurnal Riset Akuakultur, 13: 85-92.

Mugimba, K.K.; Chengula, A.A.P.; Wamala, S.; Mwega, E.D.; Kasanga, C.J.; Byarugaba, D.K.; Mdegela, R.H.; Tal, S.; Bornstein, B.; Dishon, A.; Mutoloki, S.; David, L.; Evensen, Ø. and **Munang'andu, H.M.** (2018). Detection of tilapia lake virus (TiLV) infection by PCR in farmed and wild Nile tilapia (*Oreochromis niloticus*) from Lake Victoria. J Fish Dis., 41: 1181-1189.

Nicholson, P.; Fathi, M.A.; Fischer, A.; Mohan, E.; Schieck, C. ;Mishra, N.; Heinimann, A.; Frey, J.; Wieland, B. and **Jores, J.** (2017). Detection of Tilapia Lake Virus in Egyptian fish farms experiencing high mortalities in 2015. J Fish Dis., 40: 1925-1928.

OIE (2017). Tilapia Lake Virus (TiLV), Philippines." World Organization for Animal Health (OIE). Immediate Notification. Disease notification report 25278, 23/11/2017., [https://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?page_refer=MapFullEventReport&reportid=25278]

OIE (2018). Technical disease cards: Tilapia Lake Virus (TiLV) – A Novel Orthomyxo-Like Virus. https://www.oie.int/fileadmin/Home/eng/Internationa_Standard_Setting/docs/pdf/A_TiLV_disease_card.pdf

Phusantisampan, T.; Tattiyapong, P.; Mutrakulcharoen, P.; Sriariyanun, M. and **Surachetpong, W.** (2019). Rapid detection of tilapia lake virus using a one-step reverse transcription loop-mediated isothermal amplification assay. Aquaculture, 507: 35-39.

Piamsomboon, P. and **Wongtavatchai, J.** (2021). Detection of Tilapia Lake Virus (TiLV) in healthy fish from the pre-existing disease environment using different RT-PCR methods. Turkish Journal of Fisheries and Aquatic Sciences, 21: 205-209.

Pierezan, F.; Yun, S.; Surachetpong, W. and **Soto, E.** (2019). Intragastric and intracoelomic injection challenge models of tilapia lake virus infection in Nile tilapia (*Oreochromis niloticus* L.) fingerlings. Journal of Fish Diseases, 42(9): 1301-1307.

Pulido, H.L.L.; Chìo, M.M.; Chaparro, A.L.H.; Dong, H.T. and **Senapin, S.** (2019). Tilapia lake virus TiLV from Peru is genetically close to the Israeli isolates. Aquaculture, 510: 61-65.

Soni, P.; Pradhan, P. K.; Swaminathan, T. R. and **Sood, N.** (2018). Development, characterization and application of a new epithelial cell line from caudal fin of *Pangasianodon hypophthalmus* (Sauvage 1878). Acta Tropica, 182: 215–222.

Soto, E.; Yun, S. and **Surachetpong, W. (2019).** Susceptibility of Tilapia Lake Virus to buffered povidone-iodine complex and chlorine. Aquaculture, 512: 734342. https://doi.org/10.1016/j.aquaculture.2019.734342

Surachetpong, W.; Janetanakit, T.; Nonthabenjawan, N.; Tattiyapong, P.; Sirikanchana, K. and **Amonsin, A.** (2017). Outbreaks of Tilapia Lake Virus Infection, Thailand, 2015-2016. Emerging Infectious Diseases, 23(6): 1031-1033.

Surachetpong, W.; Roy, S.R.K. and **Nicholson, P.** (2020). Tilapia lake virus: The story so far. J Fish Dis., 43:1115-1132.

Taengphu, S.; Sangsuriyabc, P.; Phiwsaiyaab, K.; Debnathd, P.P.; Delamare-Debouttevillee, J. ;Mohane, C.V.; Dong, H.T. and **Senapinab, S**. (2020). Genetic diversity of tilapia lake virus genome segment 1 from 2011 to 2019 and a newly validated semi-nested RT-PCR method. Aquaculture, 526: 735423.

Tattiyapong, P.; Sirikanchana, K. and **Surachetpong, W.** (2017). Development and validation of a reverse transcription quantitative polymerase chain reaction for tilapia lake virus detection in clinical samples and experimentally challenged fish. Journal of Fish Diseases, 41(2): 255–261.

Tattiyapong, P.; Dechavichitlead, W.; Waltzek, T.B. and **Surachetpong, W.** (2020). Tilapia develop protective immunity including a humoral response following exposure to tilapia lake virus. Fish & Shellfish Immunology, 106: 666-674.

Tsofack, J.E.K.; Zamostiano, R.; Watted, S.; Berkowitz, A.; Rosenbluth, E.; Mishra, N.; Briese, T.; Lipkin, W.I.; Kabuusu, R.M. and **Ferguson, H**. (2017). Detection of tilapia lake virus in clinical samples by culturing and nested reverse transcription-PCR. Journal of clinical microbiology, 55(3): 759-767.

Waiyamitra, P.; Tattiyapong, P.; Sirikanchana, K.; Mongkolsuk, S.; Nicholson, P. and **Surachetpong, W.** (2018). A TaqMan RT-qPCR assay for tilapia lake virus (TiLV) detection in tilapia. Aquaculture, 497: 184-188.

Waiyamitra, P.; Piewbang, C.; Techangamsuwan ,S.; Liew, W.C. and Surachetpong W. (2021).Infection of Tilapia tilapinevirus in Mozambique Tilapia (*Oreochromis mossambicus*), a Globally Vulnerable Fish Species. Viruses.;13(6):1104.