Tilapia lake virus:
Understanding the host immunity and challenges for vaccine development

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Department of Veterinary Microbiology and Immunology, Faculty of Veterinary Medicine, Kasetsart University, Bangkok, THAILAND

Outline

Introduction: Tilapia Lake Virus (TiLV)

TiLV immunology

Vaccine development and challenges

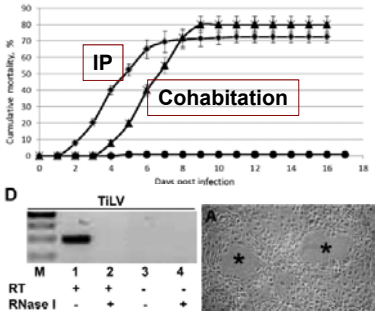
JCM
Journals.ASM.org

Identification of a Novel RNA Virus Lethal to Tilapia

Marina Eyngor,[a] Rachel Zamostiano,[b] Japhette Esther Kembou Tsofack,[b] Asaf Berkowitz,[a] Hillel Bercovier,[c] Simon Tinman,[d] Menachem Lev,[e] Avshalom Hurvitz,[f] Marco Galeotti,[g] Eran Bacharach,[b] Avi Eldar[a]

Department of Poultry and Fish Diseases, The Kimron Veterinary Institute, Bet Dagan, Israel[a]; Department of Cell Research and Immunology, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Tel Aviv, Israel[b]; The Hebrew University-Hadassah Medical School, Jerusalem, Israel[c]; Department of Animal Facility, Faculty of Life Sciences, Bar Ilan University, Ramat Gan, Israel[d]; Ein Gev Fisheries, Kibbutz Ein Gev, Israel[e]; Dan Fish Farms, Kibbutz Dan, Upper Galilee, Israel[f]; Department of Food Science, Section of Veterinary Pathology, University of Udine, Udine, Italy[g]

Tilapia Lake Virus
TiLV

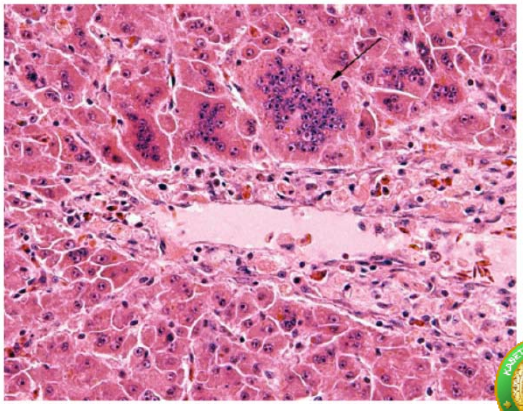


Syncytial hepatitis of farmed tilapia, *Oreochromis niloticus* (L.): a case report

Journal of Fish Diseases 2014, 37, 583–589

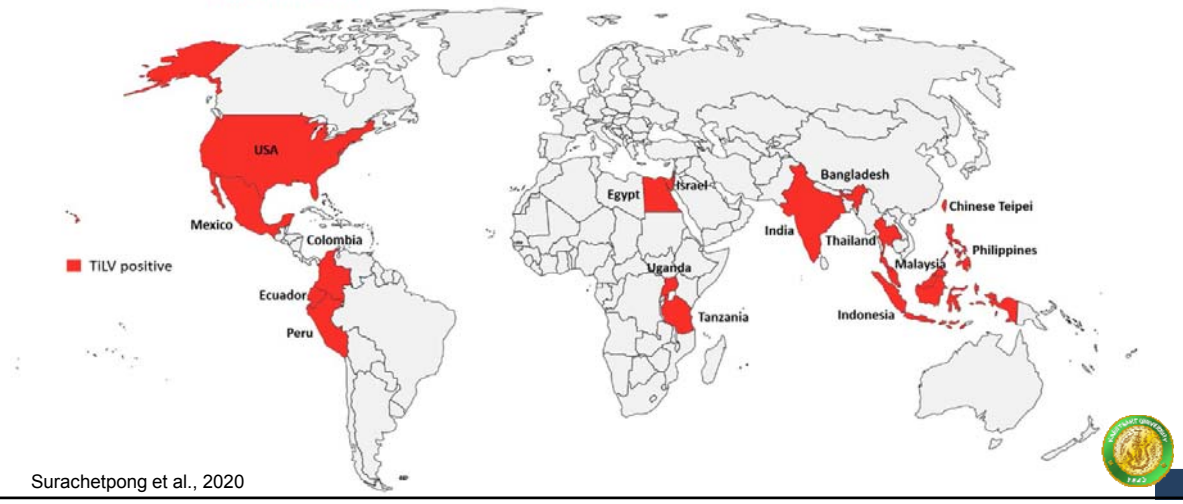
H W Ferguson[1], R Kabuusu[1], S Beltran[2], E Reyes[2], J A Lince[2] and J del Pozo[3]

1 Marine Medicine Programme, School of Veterinary Medicine, St George's University, St George, Grenada
2 Produmar S.A., Guayaquil, Ecuador
3 Department of Pathology, Royal (Dick) School of Veterinary Medicine, University of Edinburgh, Edinburgh, Scotland, UK



Geographical distribution of tilapia lake virus (TiLV)

1116 WILEY Journal of Fish Diseases SURACHETPONG ET AL.



Surachetpong et al., 2020

Malaysia

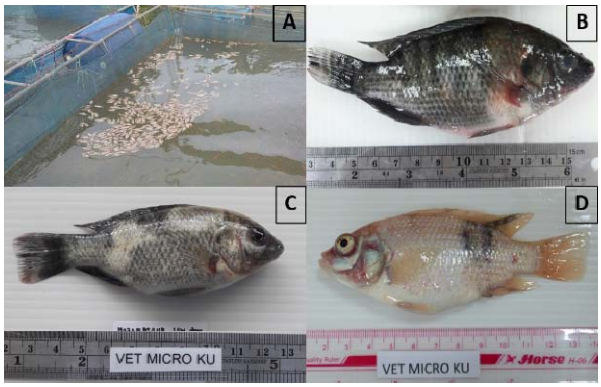
- 50-80 g
- >80% mortality

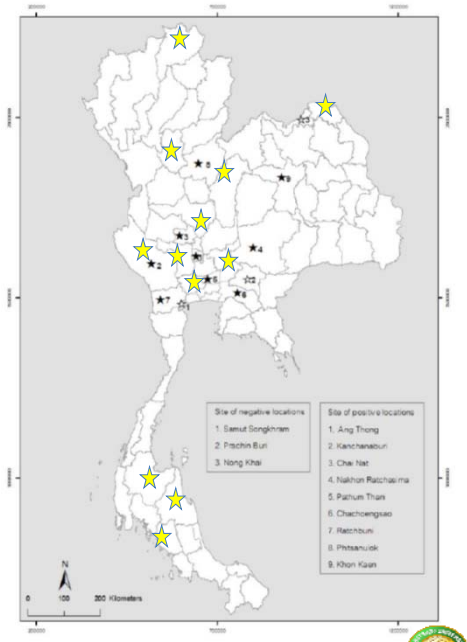
Peru

- 300-400 g
- >40% mortality

Outbreaks of TiLV in Thailand (2015-16)

- From 32 outbreaks of high mortality → 22 are TiLV positive
- Bacteria, parasite and virus





Surachetpong et al., 2017

Emerging Infectious Diseases • www.cdc.gov/eid • Vol. 23, No. 6, June 2017

Susceptible fish species for TiLV

- Wild tilapia
- Hybrid tilapia

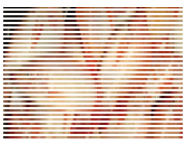
(*O. niloticus* × *O. aureus hybrids*)

- Nile tilapia (*O. niloticus*)
- Red tilapia (*Oreochromis* spp.)
- Grey tilapia (*O. niloticus x O. aureus*)

Sarotherodon galilaeus

Tilapia zilli

Oreochromis aureus



Aquaculture 497 (2018) 462–468

Contents lists available at ScienceDirect

Aquaculture

journal homepage: www.elsevier.com/locate/aquaculture

Susceptibility of important warm water fish species to tilapia lake virus (TiLV) infection

Phitchaya Jaemwimol[a], Pattarasuda Rawiwan[a,b], Puntanat Tattiyapong[a,b], Pattrawut Saengnual[c], Attapon Kamlangdee[d], Win Surachetpong[a,b,*]

Cyprinus carpio

Trichogaster pectoralis

Barbodes gonionotus

Lates calcarifer

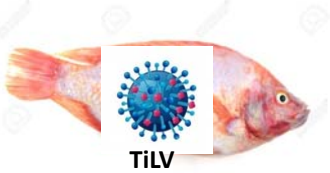
Anabas testudineus

Clarias macrocephalus

Pangasianodon hypophthalmus

Chana striata

Most important warm water fish species are resistant to tilapia lake virus (TiLV) infection



TiLV

Susceptible

***Oreochromis* sp.**

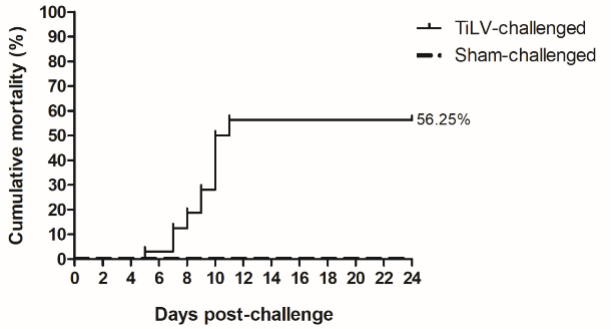
TiLV

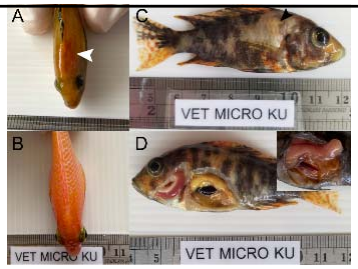
Susceptible

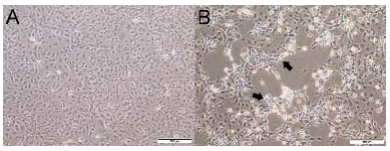
Osphronemus goramy

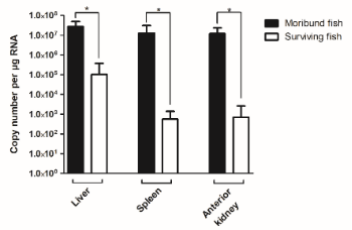
TiLV can infect ornamental African cichlids

- **High mortality, virus detected in tissues**







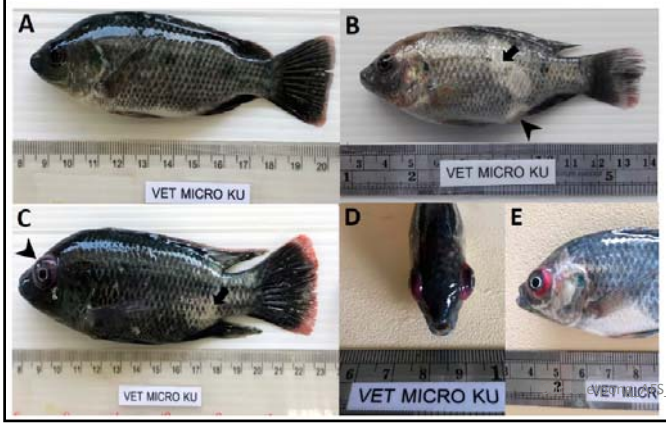


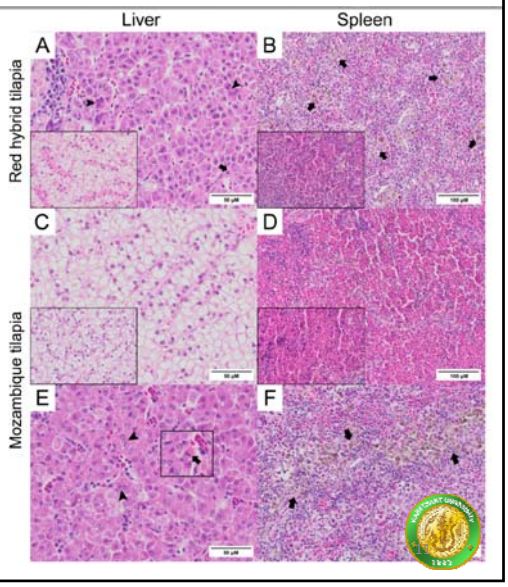
Yamkasem et al., 2021 (under review)

viruses
MDPI
Article

Infection of *Tilapia tilapinevirus* in Mozambique Tilapia (*Oreochromis mossambicus*), a Globally Vulnerable Fish Species

Pitchaporn Waiyamitra [1], Chutchai Piewbang [2,3], Somporn Techangamsuwan [2,3], Woei Chang Liew [4,†] and Win Surachetpong [1,*]





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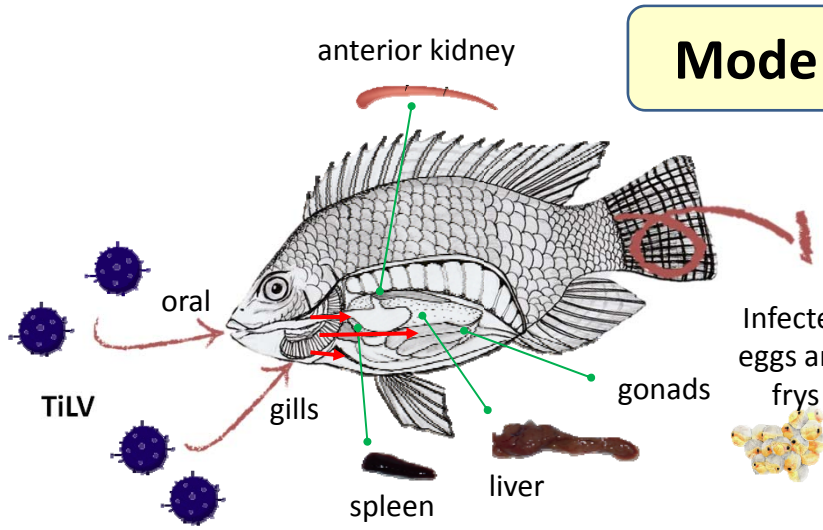
Mode of transmission



Shedding via mucus

Vertical and horizontal transmission

Surachetpong et al., 2020

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Fish and Shellfish Immunology 116 (2021) 115–123

Contents lists available at ScienceDirect

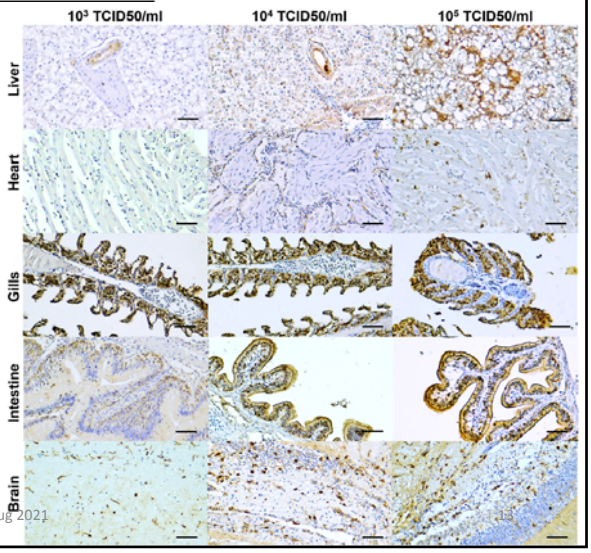
Fish and Shellfish Immunology

journal homepage: www.elsevier.com/locate/fsi

Tilapia lake virus immunoglobulin G (TiLV IgG) antibody: Immunohistochemistry application reveals cellular tropism of TiLV infection

Chutchai Piewbang [a,b,1], Puntanat Tattiyapong [c,1], Somporn Techangamsuwan [a,b,*], Win Surachetpong [c,**]

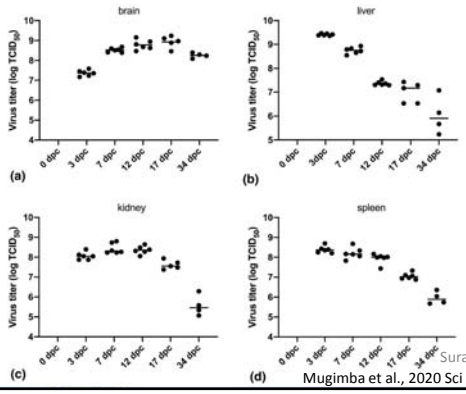
- TiLV localization in intestines, gills, liver, spleen, kidneys
- Endothelial cells and circulating lymphocytes



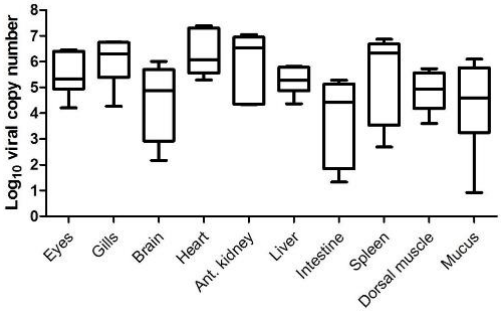
TiLVD is a systemic disease..... the virus is detected in most tissues.

	Viral loads (copies/µg of total RNA)					
Sample No.	Gills	Liver	Brain	Heart	Anterior kidney	Spleen
1	2.2×10^5	1.7×10^5	3.4×10^5	6.3×10^5	1.6×10^5	2.3×10^4
2	3.1×10^5	6.3×10^3	1.3×10^6	3.9×10^5	3.1×10^5	2.8×10^4

Tattiyapong et al., 2018 J Fish Dis.



Mugimba et al., 2020 Sci Reports.



Yamkasem et al., 2021 Trans Emerg Disease

Thammatorn et al., 2019 J Fish Diseases

Outline

Introduction: Tilapia Lake Virus (TiLV)

TiLV immunology

Vaccine development and challenges

Evidence of protective immunity against TiLV

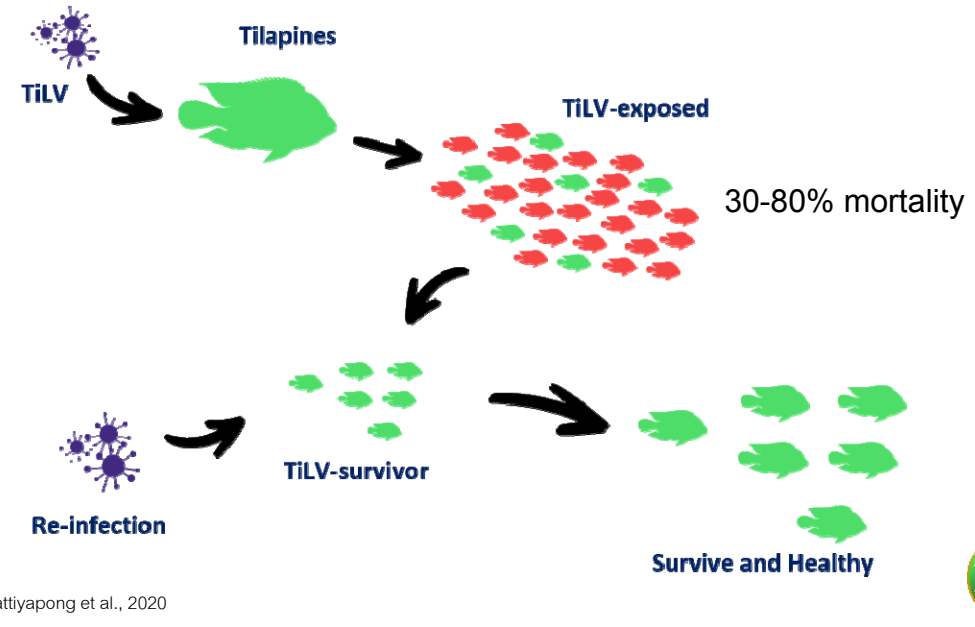
- Once the initial wave of mortality ceased, no more outbreaks were recorded in the same pond. (Eyngor et al., 2014)
- The existence of fish that survived the TiLV-induced disease strongly suggests that an effective immune response against this pathogen can be mounted. This has important applications for future disease containment strategies. (Eyngor et al., 2014) such as vaccine development.

Repeat TiLV exposure do not cause disease and mortality in tilapia

Experiment	Group	Challenge method	No. of fish	Survival rate (%)	
				1st challenge	2nd challenge
1	Naïve	Intraperitoneal injection	20	**25 (5/20)**	100 (5/5)
			20	**35 (7/20)**	100 (7/7)
2		Cohabitation	15	**40 (6/15)**	100 (6/6)
			15	**66 (10/15)**	100 (10/10)
3	Previous TiLV exposure	Intraperitoneal injection	20	**100 (20/20)**	
			20	**100 (20/20)**	
4		Cohabitation	15	**100 (15/15)**	
			15	**100 (15/15)**	

Tattiyapong et al., 2020 Fish & Shellfish Immunology

Protective immunity against TiLV



Eyngor et al., 2014, Tattiyapong et al., 2020

Fish and Shellfish Immunology 106 (2020) 666–674

Contents lists available at ScienceDirect

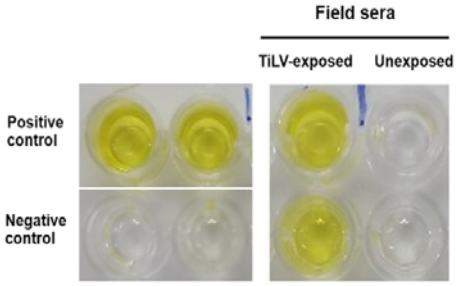
Fish and Shellfish Immunology

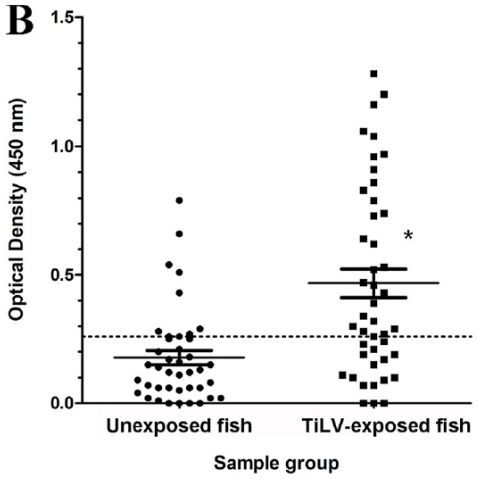
journal homepage: www.elsevier.com/locate/fsi

Full length article

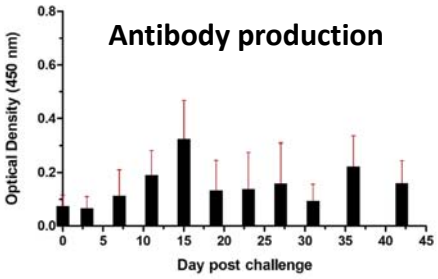
Tilapia develop protective immunity including a humoral response following exposure to tilapia lake virus

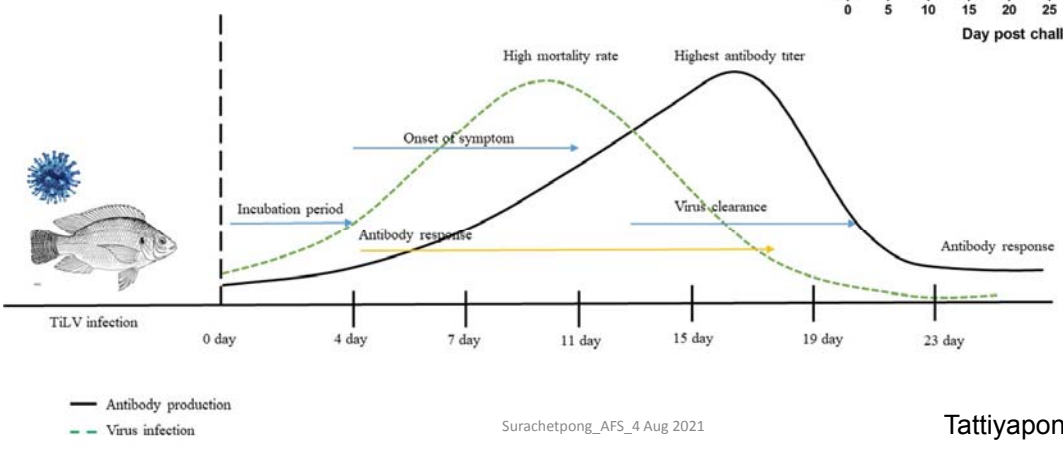
Puntanat Tattiyapong [a,b], Worawan Dechavichitlead [a,b], Thomas B. Waltzek [c,d], Win Surachetpong [a,b,*]





Detection of antibody in serum of TiLV-infected fish

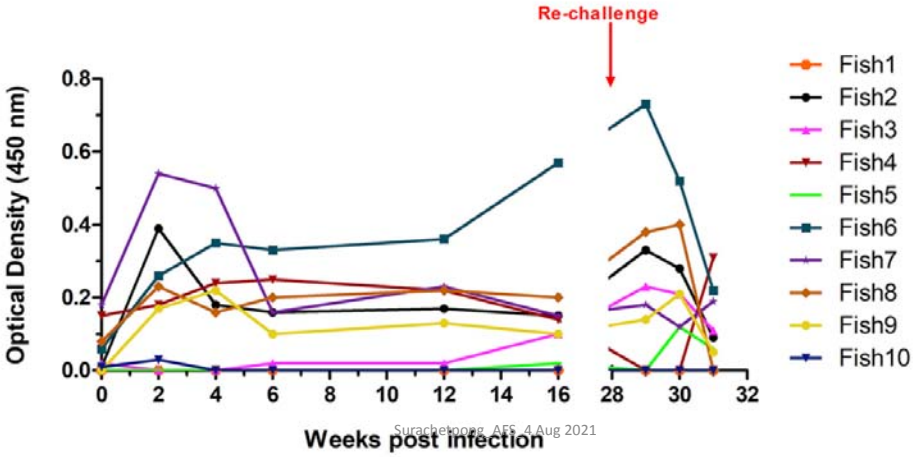




Tattiyapong et al., 2020

Antibody is the key immune response to control TiLV

- Rapid antibody response within 2 weeks
- Antibody persists for 4-5 months with anamnestic response.

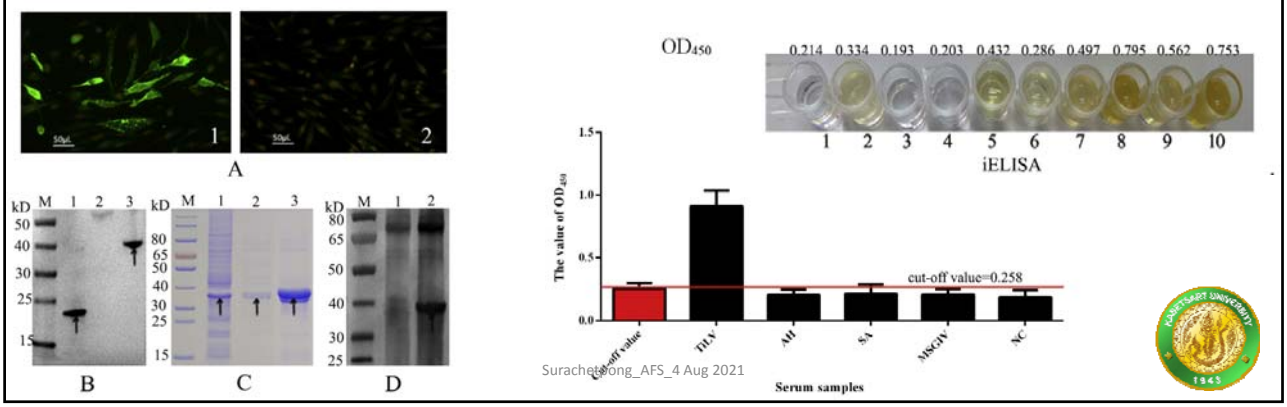


Contents lists available at ScienceDirect
Aquaculture
journal homepage: www.elsevier.com/locate/aquaculture

Development and application of a recombinant protein-based indirect ELISA for detection of anti-tilapia lake virus IgM in sera from tilapia

Huzi Hu[a,c], Weiwei Zeng[a,*], Yingying Wang[a], Qing Wang[a], Sven M. Bergmann[b], Jiyuan Yin[a], Yingying Li[a], Xiaoyu Chen[d], Caixia Gao[a,c], Defeng Zhang[a], Chun Liu[a], Yan REN[a], Cunbin Shi[a]

- Segment 8 of TiLV



Outline

Introduction: Tilapia Lake Virus (TiLV)

TiLV immunology

Vaccine development and challenges

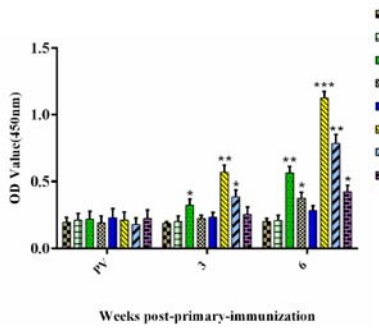
vaccines

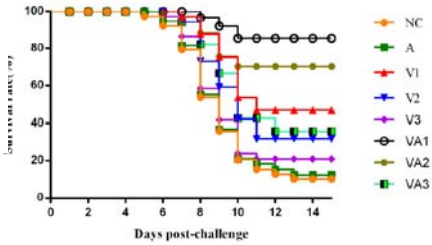
MDPI

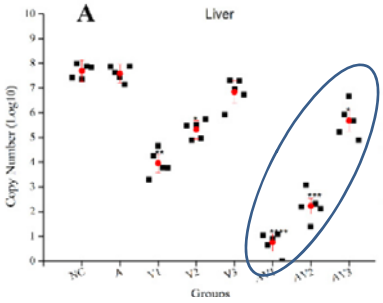
Article

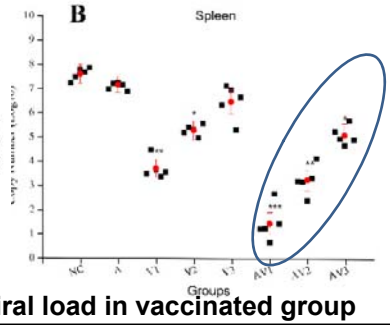
Cell Culture-Derived Tilapia Lake Virus-Inactivated Vaccine Containing Montanide Adjuvant Provides High Protection against Viral Challenge for Tilapia

Weiwei Zeng [1,2,*], Yingying Wang [2], Huzi Hu [2], Qing Wang [2,*], Sven M. Bergmann [3], Yahui Wang [2], Bo Li [2], Yuefeng Lv [2], Hua Li [1], Jiyuan Yin [1] and Yingying Li [2]









Higher antibody in antigen+adjuvant

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Less TiLV viral load in vaccinated group

Aquaculture 539 (2021) 736654

Contents lists available at ScienceDirect

Aquaculture

journal homepage: www.elsevier.com/locate/aquaculture

Potency and efficacy of VP20-based vaccine against tilapia lake virus using different prime-boost vaccination regimens in tilapia

Weiwei Zeng [a,*], Yingying Wang [b], Xiaoyu Chen [d], Qing Wang [b,**], Sven M. Bergmann [c], Ying Yang [a], Yahui Wang [b], Bo Li [b], Yuefeng Lv [b], Hua Li [a], Wensheng Lan [e]

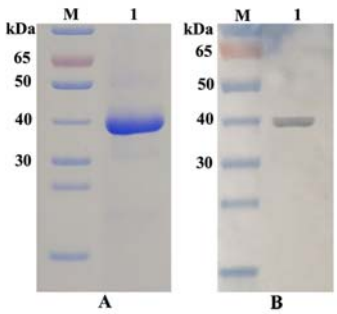
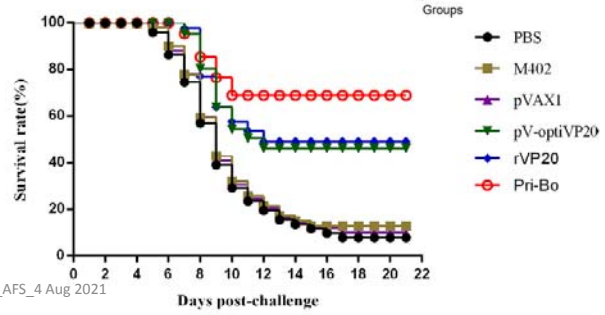
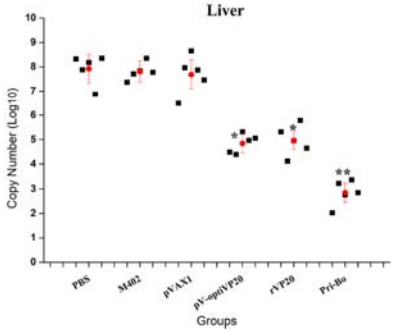


Fig. 3. Expression of recombinant VP20 in transfected in Tilapia cells.Indirect immunofluorescence assay using the VP20-specific polyclonal antibody of cells containing (A) pVAX1 (B) pV-VP20 and (C) pV-optiVP20.





Surachetpong_AFS_4 Aug 2021

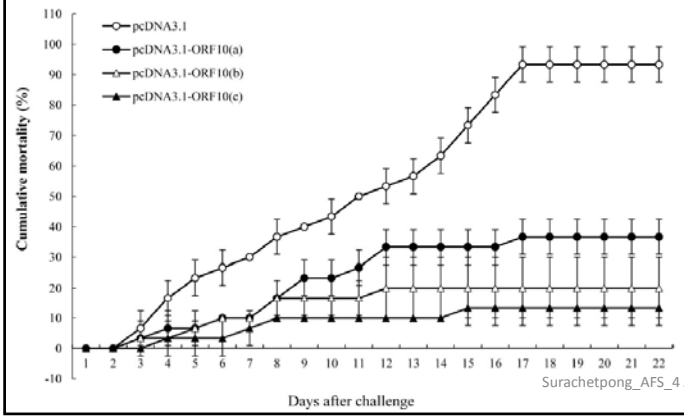
Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 28 July 2021 doi:10.20944/preprints202107.0626.v1

Article

A high efficacy DNA vaccine against tilapia lake virus in Nile tilapia (*Oreochromis niloticus*)

Nai-tong Yu[1*], Wei-wei Zeng[2], Jian-hua Wang[1], Yu-liang Zhang[1], Xiu-chun Zhang[1], Zhi-xin Liu[1]

- TiLV ORF10



Vaccinated tilapia	Cumulative mortality (death/total)	RPS%
pcDNA3.1–ORF10 (c)	13.33% (4/30)	85.72
pcDNA3.1–ORF10 (b)	20.00% (6/30)	78.57
pcDNA3.1–ORF10 (a)	36.67% (11/30)	60.71
pcDNA3.1	93.33% (28/30)	-

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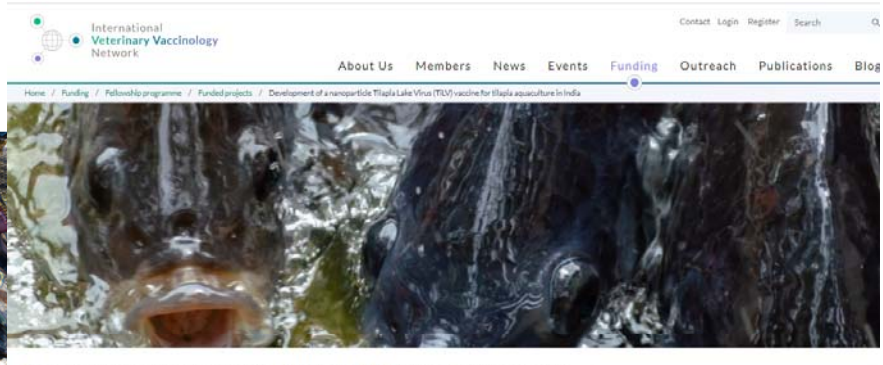
New grant will help develop vaccine delivery system for tilapia lake virus

By Shem Oirere
April 2, 2021

of 50 percent or more.



Development of a nanoparticle Tilapia Lake Virus (TiLV) vaccine for tilapia aquaculture in India

Project summary

Aquaculture is the fastest growing food-production sector globally, with over 1 billion people relying on fish as their major protein source. Tilapia (*Oreochromis sp.*) is a major trade commodity for many low to middle-income countries (LMIC), with its production estimated to be around 6.4 million tons per annum (FAO, 2017). The hardiness of tilapia, its adaptability to various production systems and its rapid growth, makes it an excellent fish species for aquaculture. Intensification of tilapia farming has promoted severe disease outbreaks, however, resulting in high mortalities and economic hardship for tilapia farmers. Tilapia Lake Virus (TiLV), a highly virulent and contagious novel orthomyxo-like virus has recently been associated with disease outbreaks in tilapia aquaculture, resulting in massive mortalities in both wild and cultured tilapines. First reported in Israel in late 2009, TiLV-related disease outbreaks have now been reported across Asia, Africa, and North and South America. In Indian tilapia aquaculture, the virus is associated with mortality levels around 80-90%. Vaccination has proven a successful tool for controlling viral diseases in aquaculture, with most vaccines delivered by intraperitoneal (IP) injection. Many tilapia farmers will not vaccinate by injection once the fish have been moved onto the farm and would prefer fish to be vaccinated in the hatchery. It is difficult to inject small fish, however. Also TILV tends to affect small fingerlings, some of which are too small to inject. Alternative vaccine delivery methods, such as oral or

Researchers involved in this project

Dr Sreeja Lakshmi
Kerala University of Fisheries and Ocean Studies, India

Dr Preetham Elumalai
Kerala University of Fisheries and Ocean Studies, India

Dr Kim Thompson
Moredun Research Institute, UK

Dr David Smith
Moredun Research Institute, UK

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Take home message

- **Tilapia Lake virus** is threatening global tilapia aquaculture
- Fish survive TiLV infection develop **protective immunity** that prevents subsequent infection
- Different TiLV-prototype vaccines are under developed
- **Vaccine** and biosecurity will be important tools to reduce the disease

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Thank you for your attention

fvetwsp@ku.ac.th