FAO/ASTF Project: GCP/RAF/510/MUL:

Enhancing capacity/risk reduction of emerging Tilapia Lake Virus (TiLV) to African tilapia aquaculture: Intensive Training Course on TiLV

4-13 December 2018. Kisumu, Kenya

in cooperation with Kenya Marine Fisheries Research Institute (KMFRI) and Kenya Fisheries Service (KeFS)

Session:

Tilapia Lake Virus: causative, agent, distribution, epidemiology

Win Surachetpong (DVM, PhD, DTBVP, CertAqV)

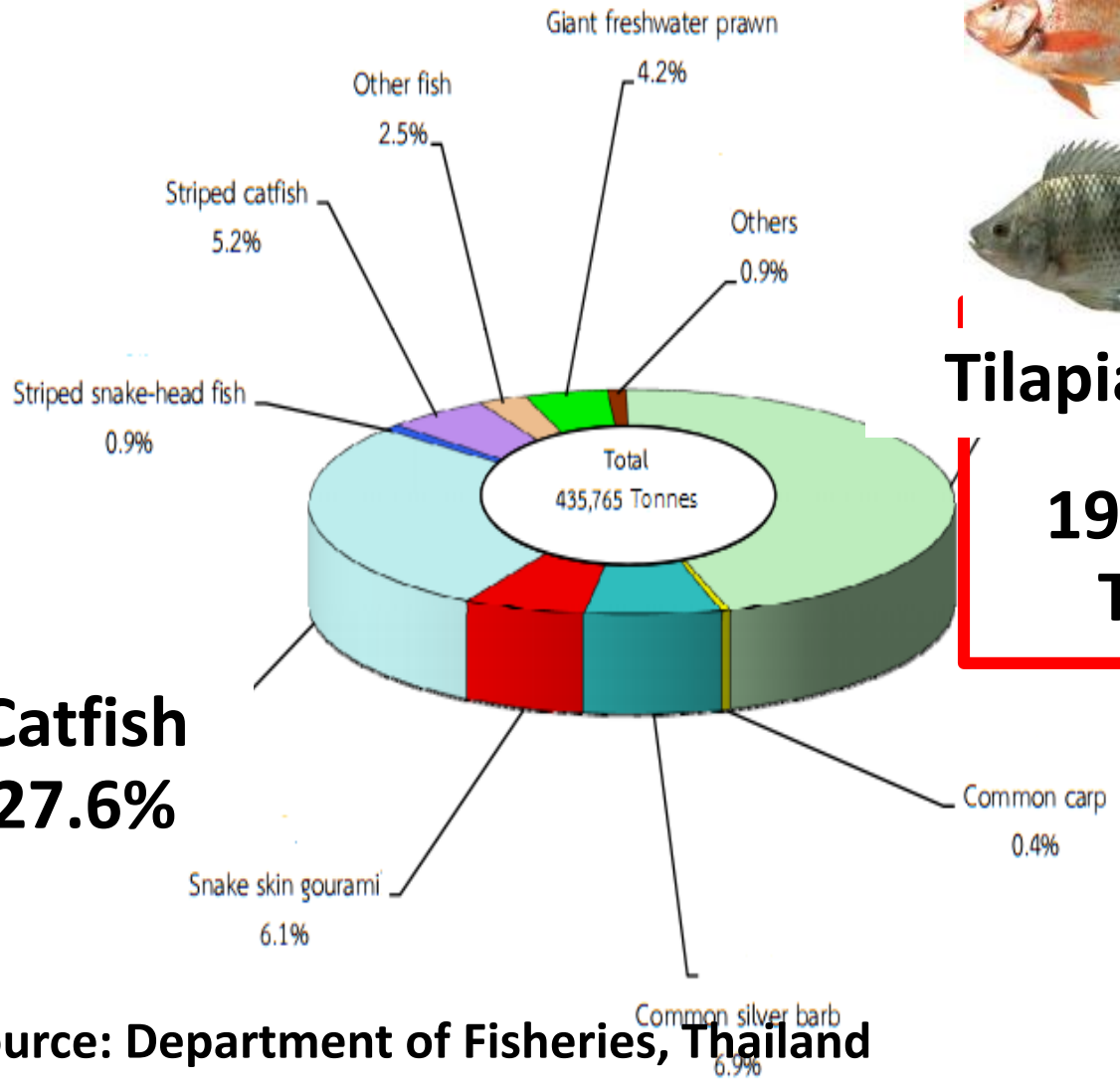
fvetwsp@ku.ac.th

Food and Agriculture Organization of the United Nations

Faculty of Veterinary Medicine, Kasetsart University

- One of the oldest Vet school in Thailand
- 140 Faculty, 750 Vet students
- 4 teaching hospitals, with >700 cases/day

Tilapia is the main aquaculture species in Thailand



Tilapia 45.3%

197,401 Tons

Catfish 27.6%

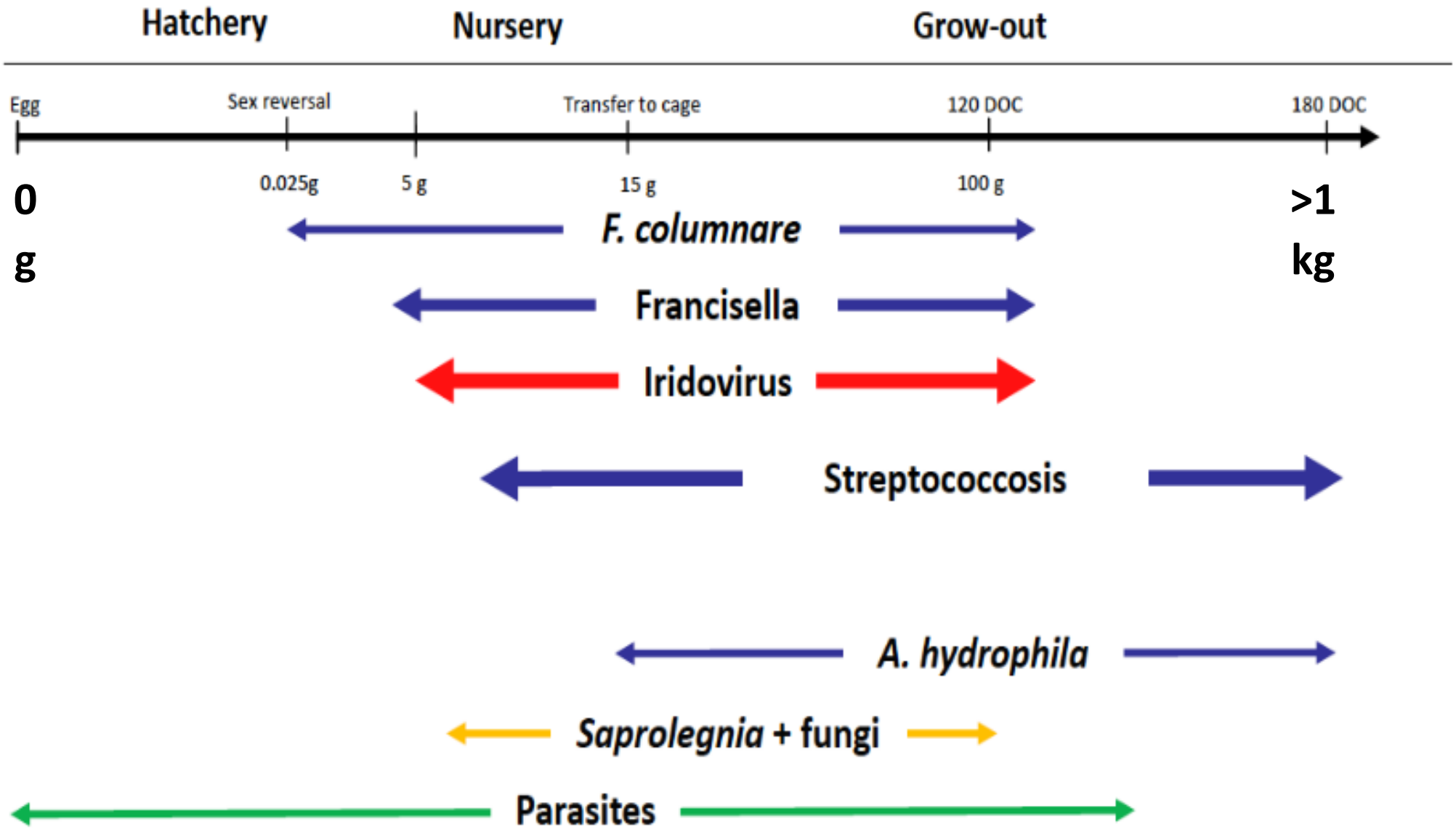
Source: Department of Fisheries, Thailand

Tilapia aquaculture, THAILAND

Identification of a new emerging disease

"to open Pandora's box" means to perform an action that may seem small or innocent, but that turns out to have severely detrimental and far-reaching negative consequences. (Wikepedia)

Infectious diseases of tilapia



Massive mortality in red hybrid tilapia


Win Surachetpong


Win Surachetpong


Win Surachetpong


Win Surachetpong

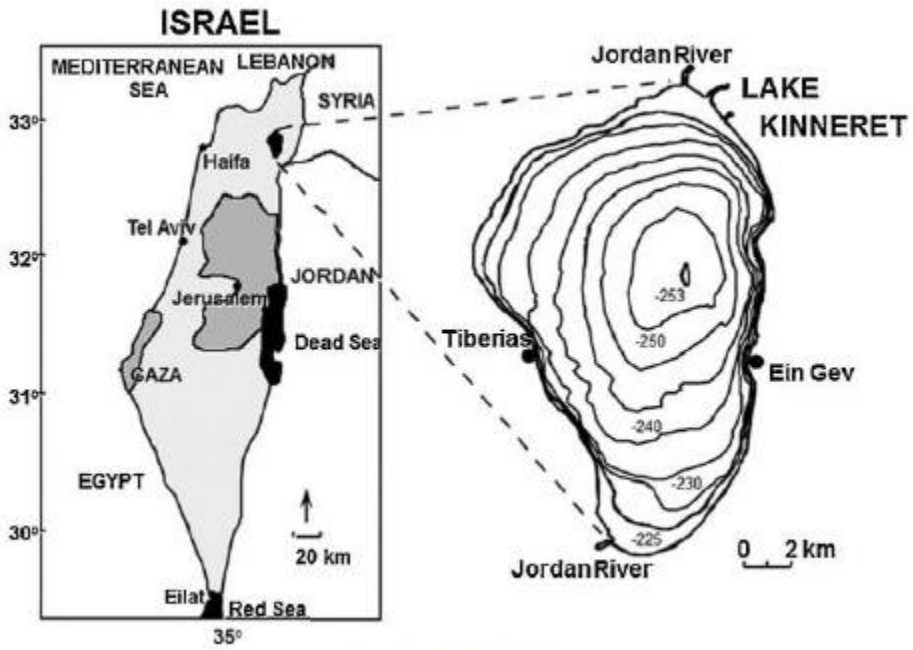
High mortality in culture tilapia


Daily mortality above 1-2% for 3 consecutive days
60-100 fish/cage Fish size 20 g
No history of high mortality since stocking
Win Surachetpong

The start – farmed stocks

- Summer of 2009: massive mortality events in all tilapia fish farming areas
- "Wave of mortality spreading from cage to cage "
- No apparent cause

- Significant decrease of *Sarotherodon galilaeus*



Parparov & Gal, 2012

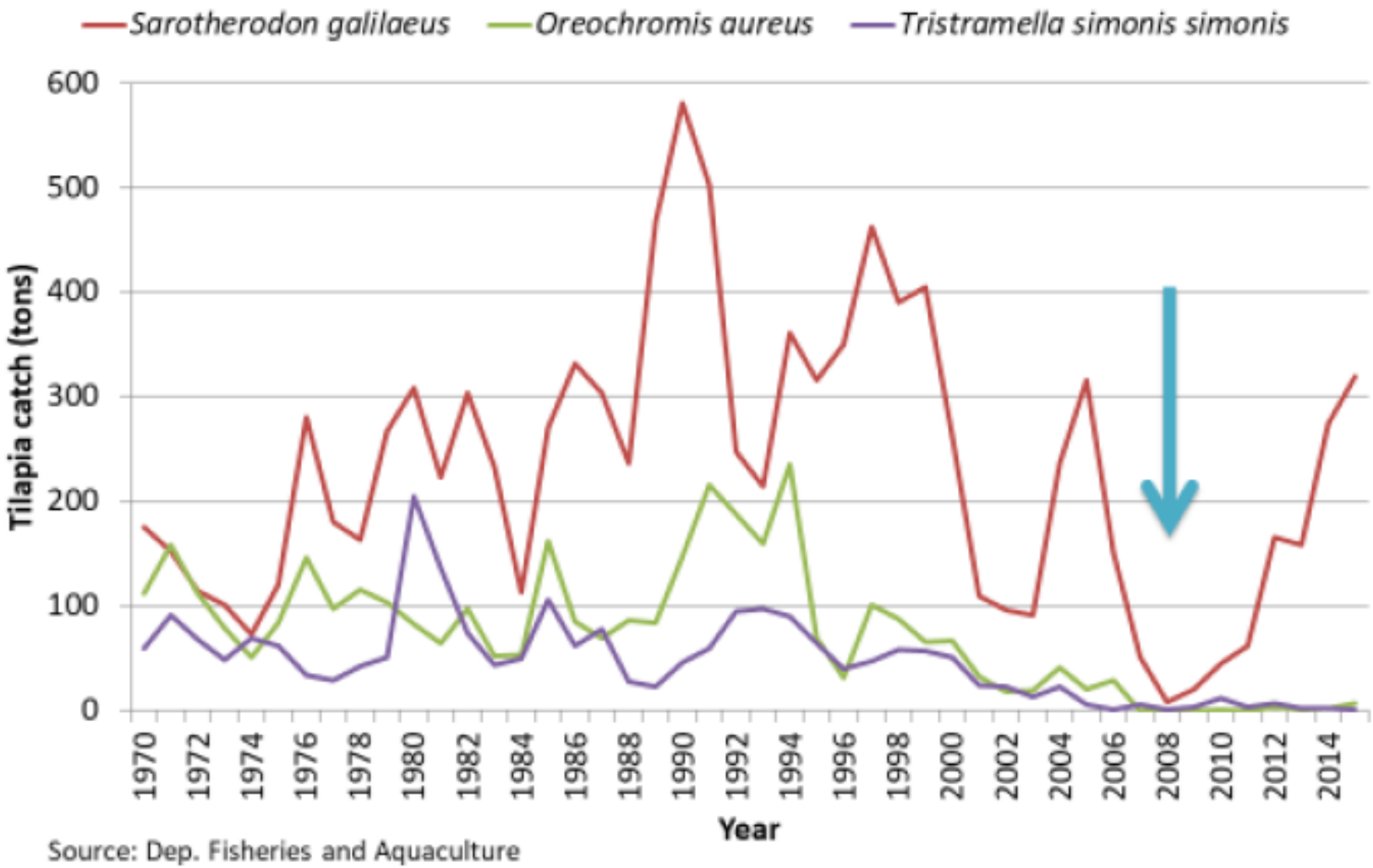
JCM
JournalsASM.org

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 catch – Lake Kinneret



Source: Dep. Fisheries and Aquaculture

Courtesy of: N. Davidovich, Ministry of Agricultrure and Rural Development

JCM Journals.ASM.org

December 2014 Volume 52 Number 12 Journal of Clinical Microbiology p. 4137–4146

Identification of a Novel RNA Virus Lethal to Tilapia

Tilapia Lake Virus: TiLV

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]

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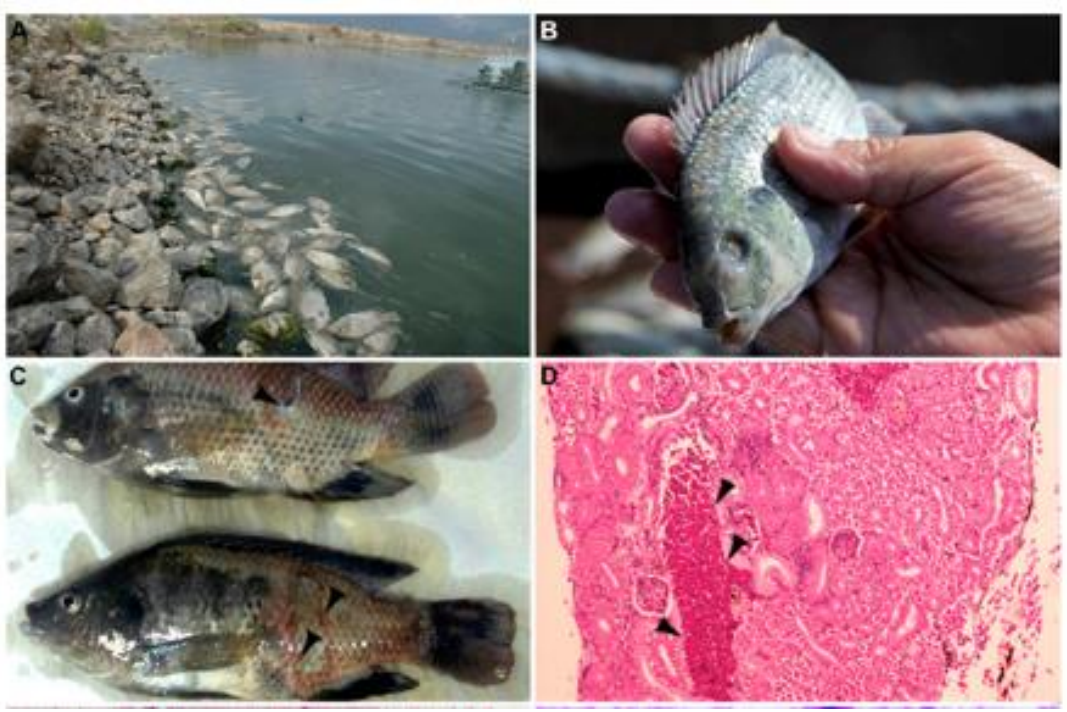
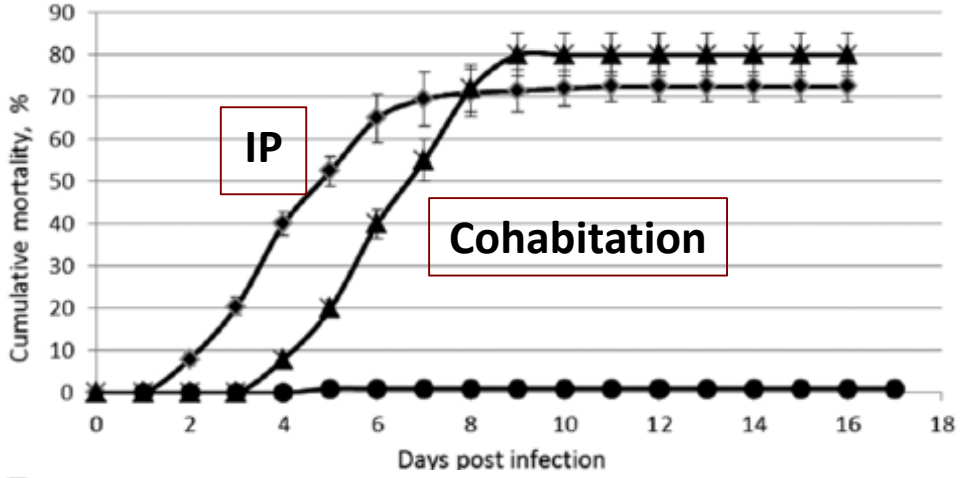
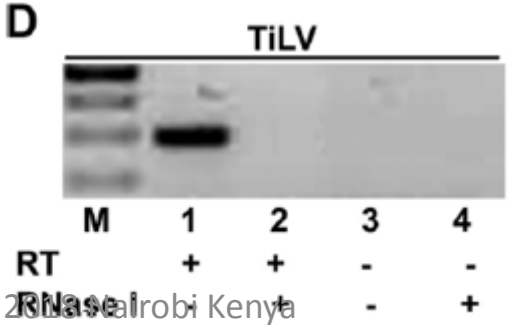
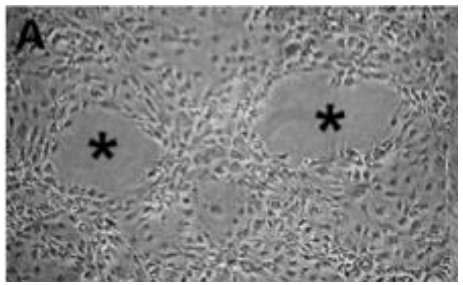






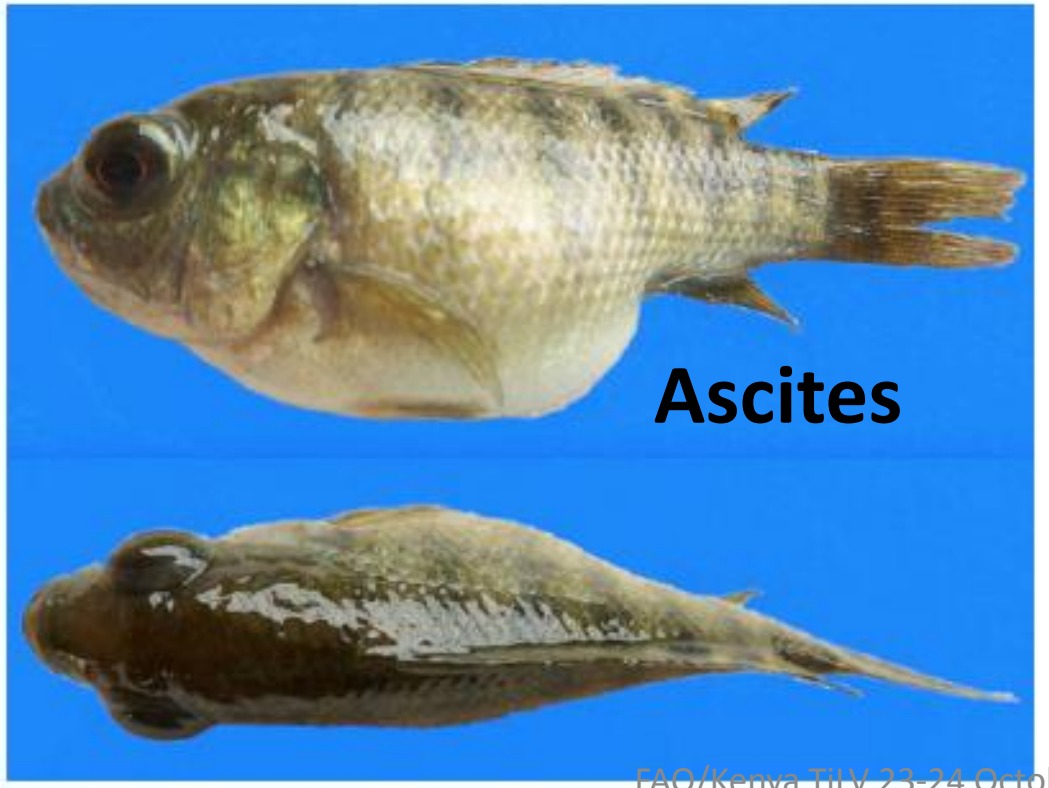


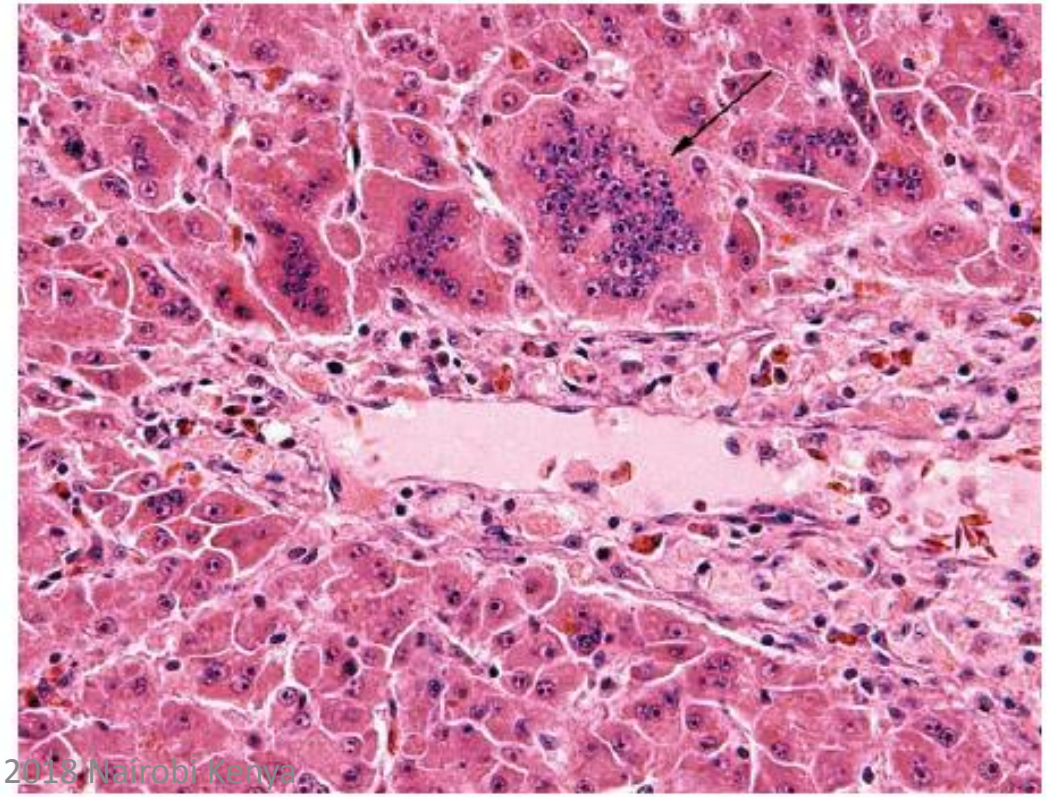
Syncytial hepatitis of farmed tilapia, *Oreochromis niloticus* (L.): a case report

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]

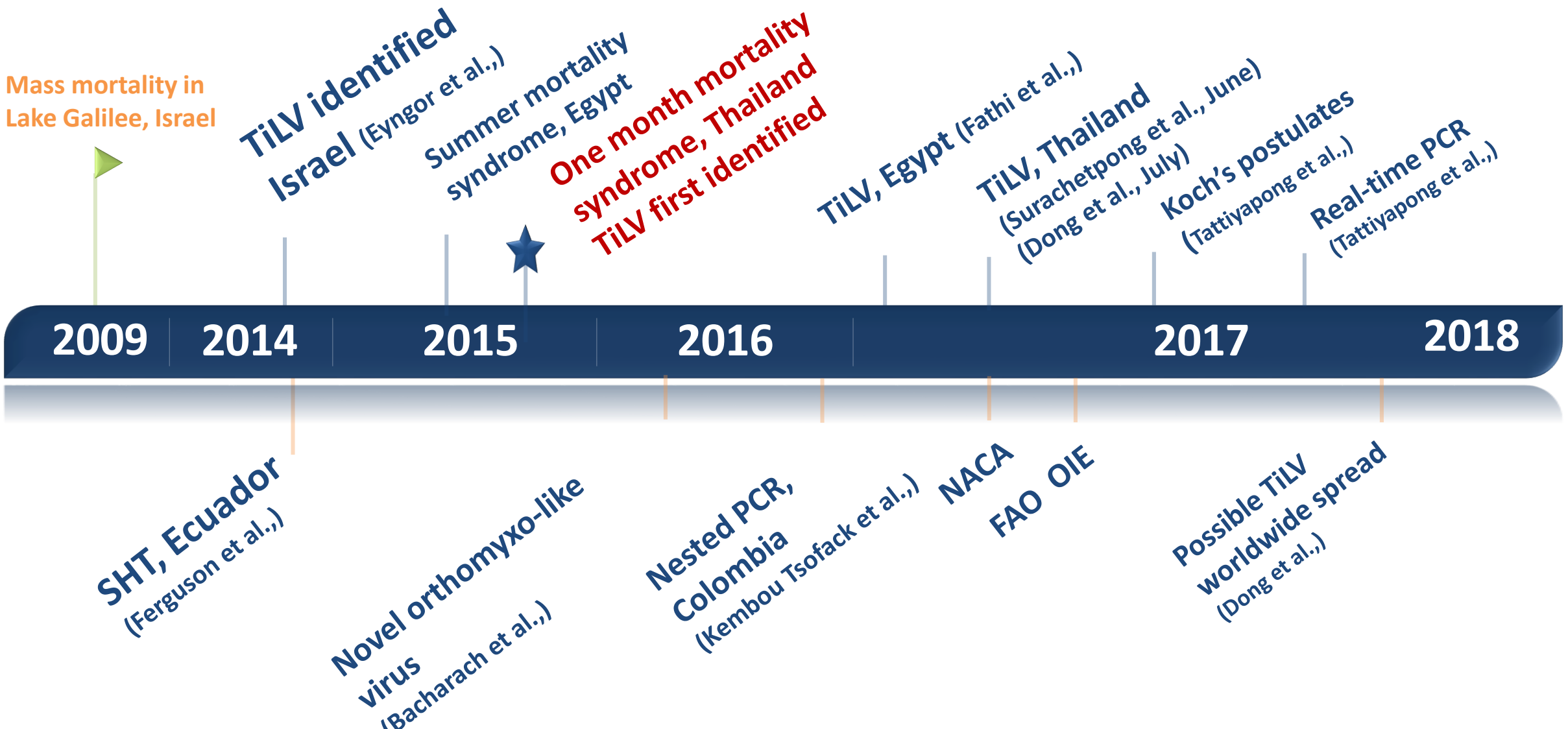
1 Marine Medicine Programme, School of Veterinary Medicine, St George's University, St George, Grenada
2 Produmar S.A., Guayaquil, Ecuador
3 Department of Pathology, Royal (Dick) School of Veterinary Medicine, University of Edinburgh, Edinburgh, Scotland, UK



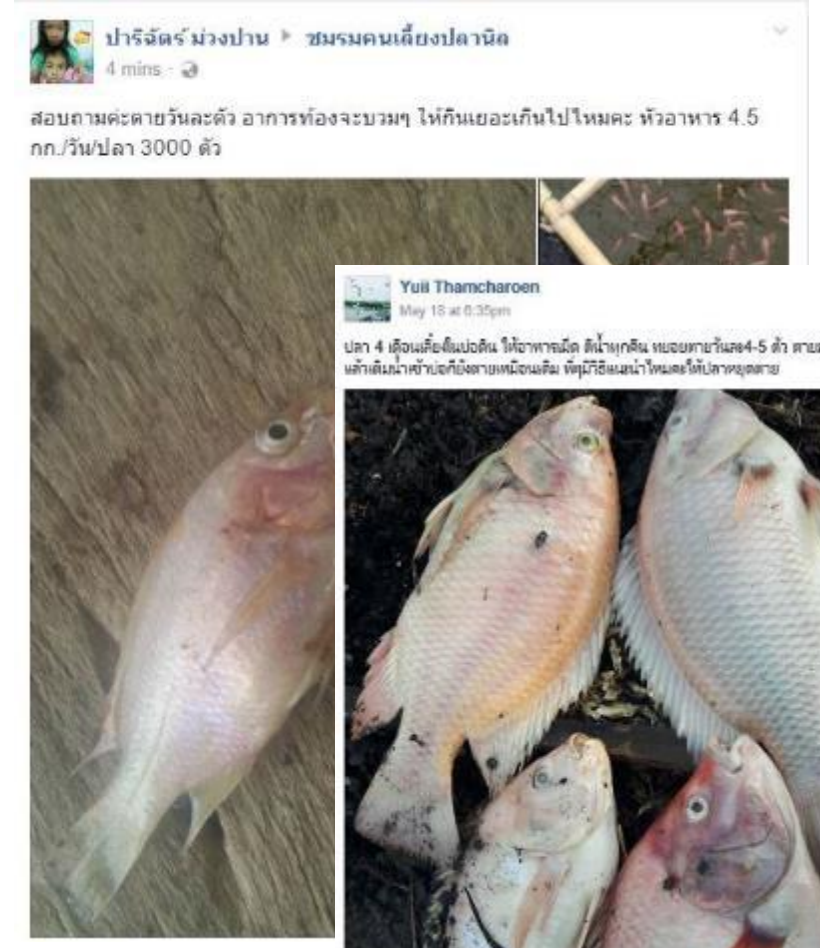


TiLV discovery

TiLV discovery



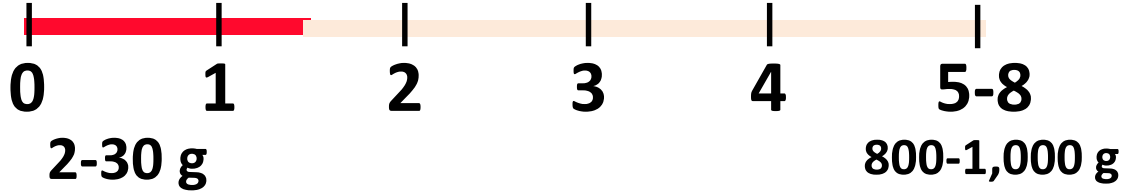
Unknown cause(s) of tilapia death



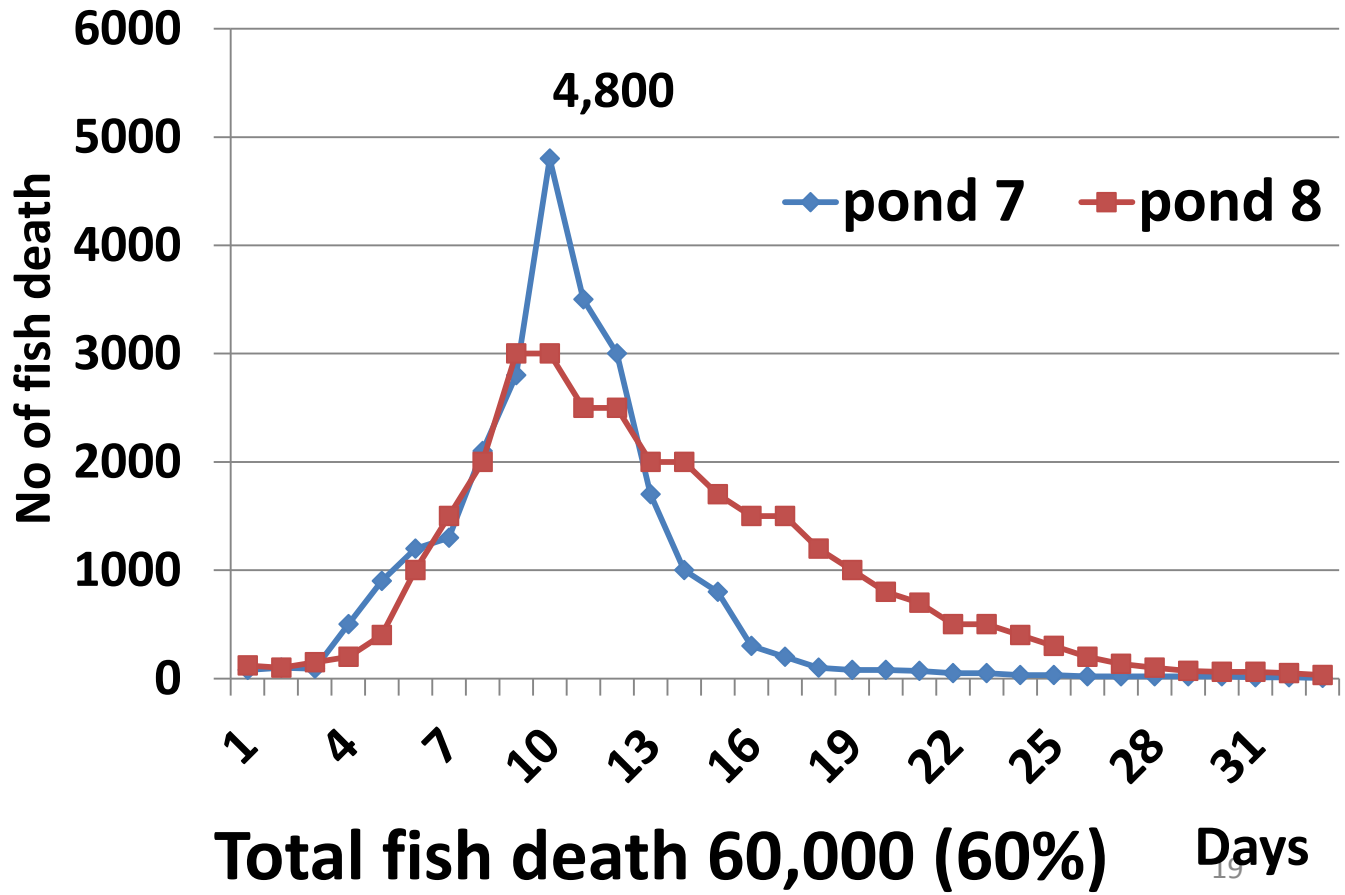
Pictures from facebook

Tilapia One Month Mortality Syndrome (TOMMS)

Months in cage/pond culture



Pattern of fish death



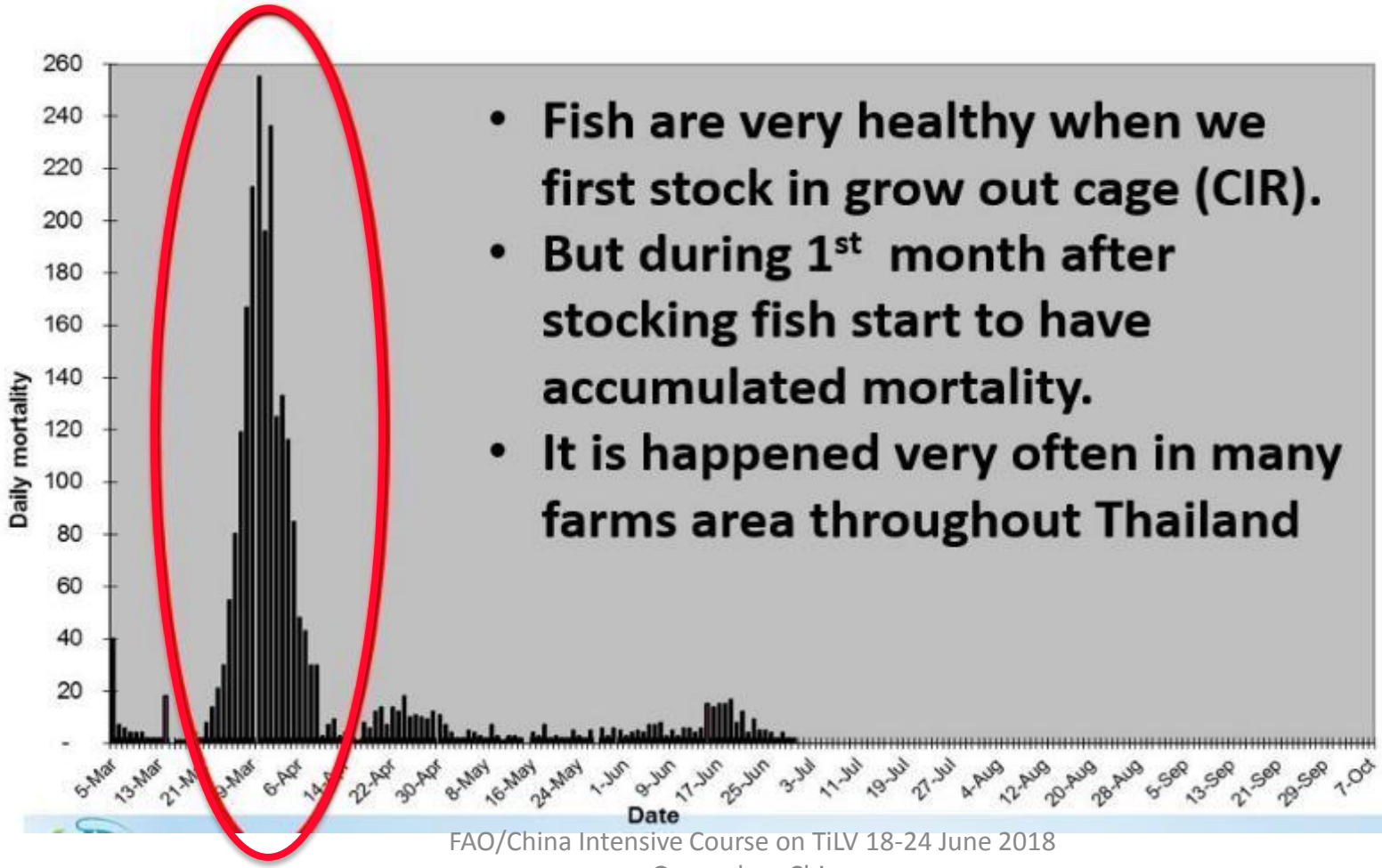
Total fish death 60,000 (60%)

Tilapia One Month Mortality Syndrome (TOMMS)

- High mortality rates often found within one month of transferring tilapia into rearing sites
    - Transportation?
    - Acclimatization to new environments?
    - Pathogens at rearing site?

***Parinda kumchum , MSD ,Bangkok Thailand 2015

1st month post stocking mortality



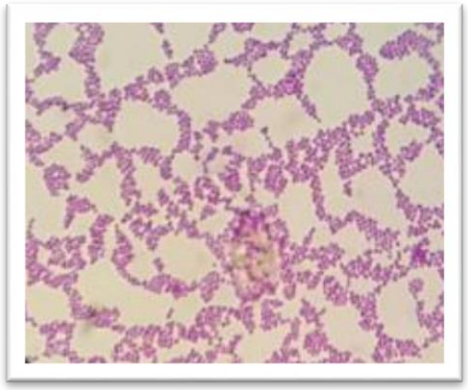
- Fish are very healthy when we first stock in grow out cage (CIR).
- But during 1st month after stocking fish start to have accumulated mortality.
- It is happened very often in many farms area throughout Thailand

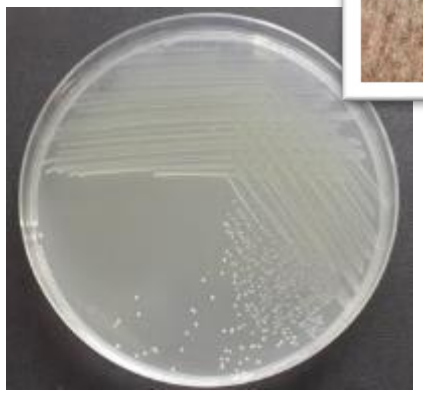
Immunized tilapia fingerings

ปลาผ่านเชื้ออนุบาลในแม่น้ำเมืองกาญครับสนใจติดต่อสอบถามได้ครับ

- **Nursery culture in the river**
- **Survived from massive die offs**
- **Healthy in growout period**

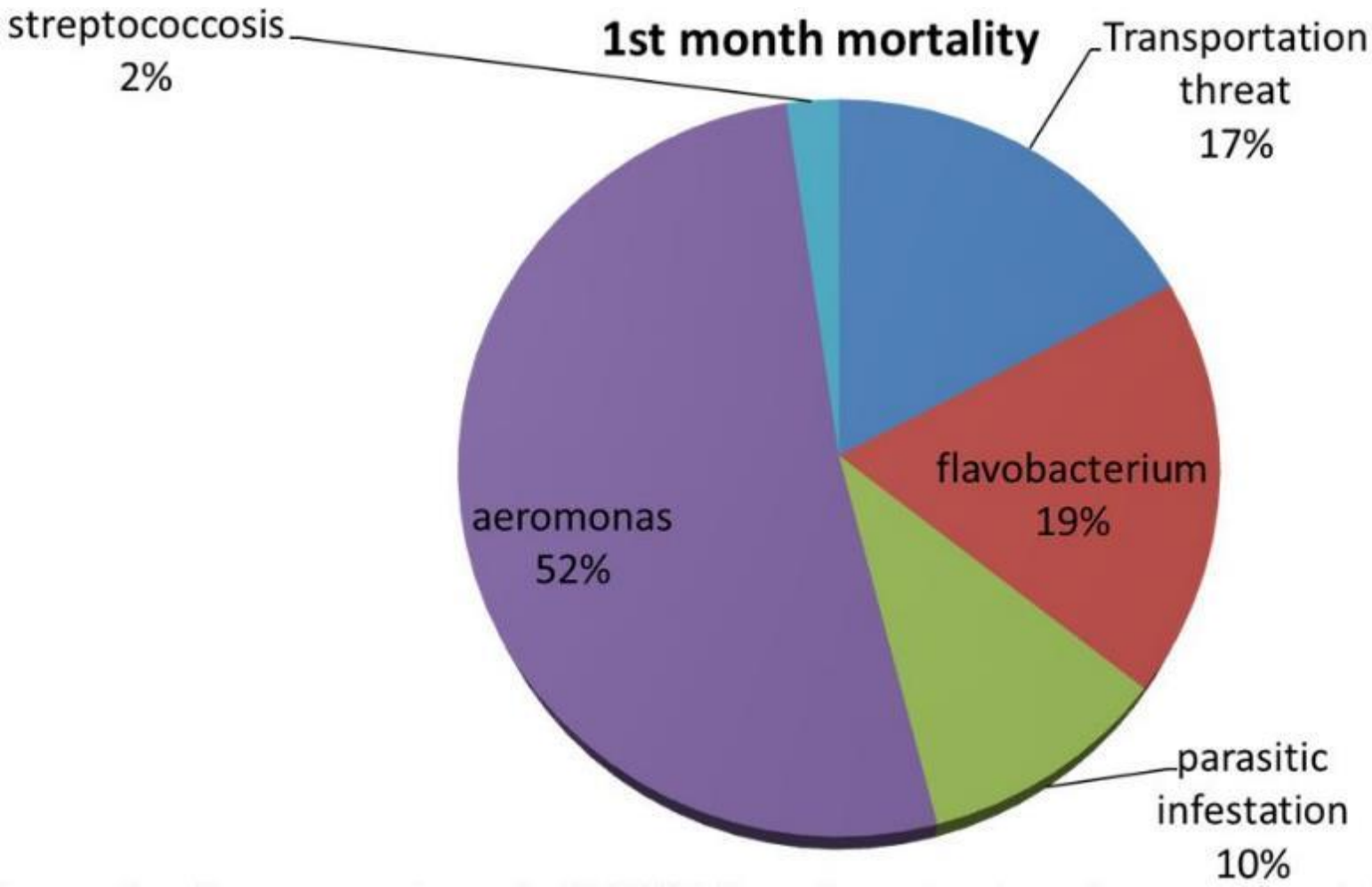
Screening for external parasites and other bacterial infection





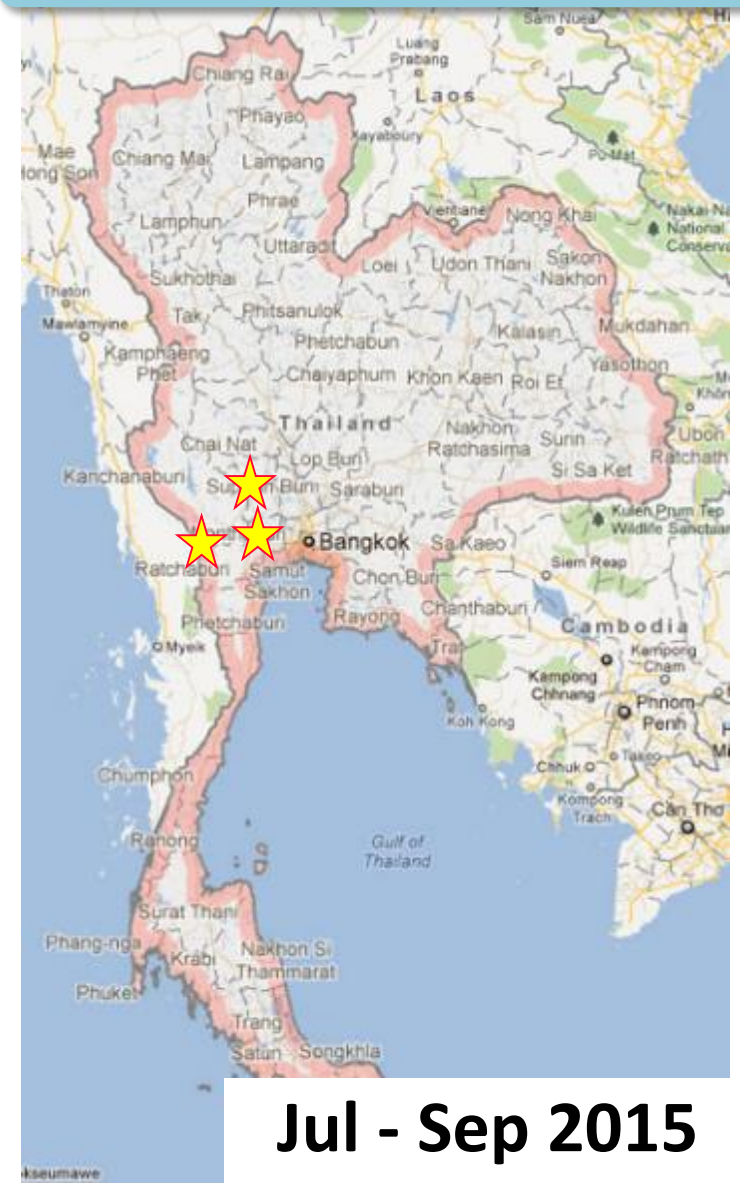
Bacterial culture results of TOMMS

Data between June and December 2015 in a hatchery farm (600,000 fingerlings)



TOMMS & Tilapia Lake Virus ??

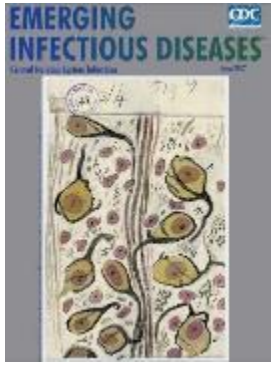
TiLV discovery from TOMMS in 2015



Jul - Sep 2015

Farm owners

Outbreaks of Tilapia Lake Virus Infection, Thailand, 2015–2016

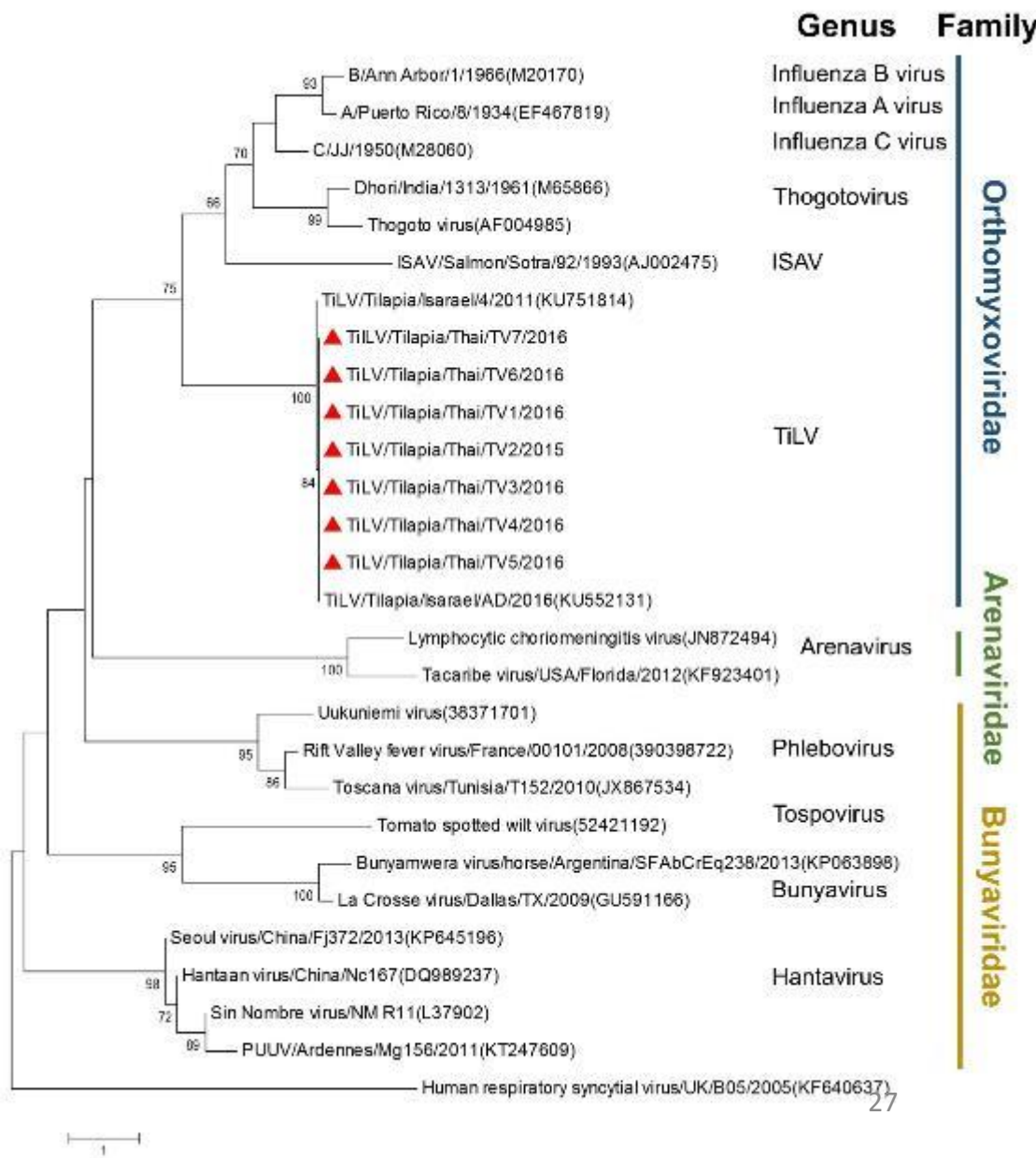


Win Surachetpong, Taveesak Janetanakit, Nutthawan Nonthabenjawan, Puntanat Tattiyapong, Kwanrawee Sirikanchana, Alongkorn Amonsin

Author affiliations:, Kasetsart University, Bangkok, Thailand (W. Surachetpong, P. Tattiyapong); Chulalongkorn University, Bangkok (T. Janetanakit, N. Nonthabenjawan, A. Amonsin); Chulabhorn Research Institute, Bangkok (K. Sirikanchana); Ministry of Education, Bangkok (K. Sirikanchana)

DOI: https://dx.doi.org/10.3201/eid2306.161278

Orthomyxo-like virus



Aquaculture 476 (2017) 111–118

Contents lists available at ScienceDirect

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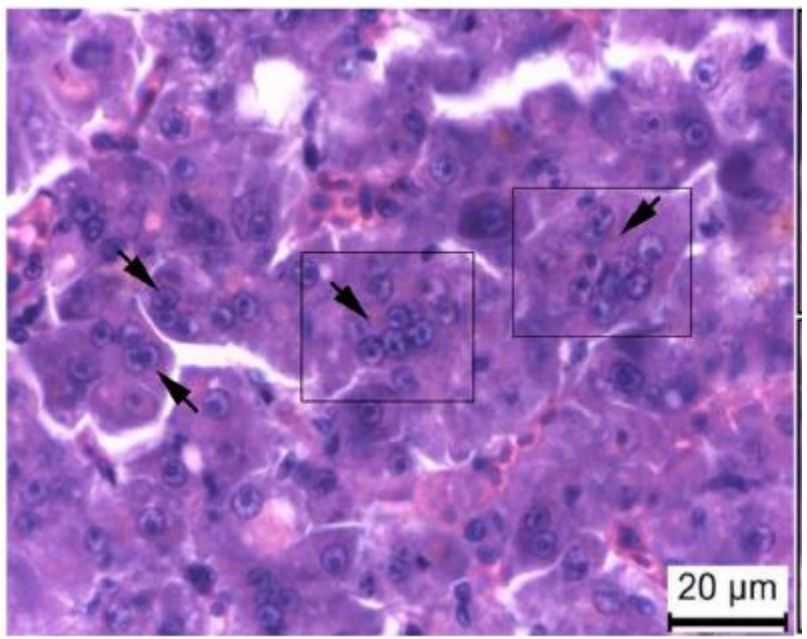
H.T. Dong[a,b,*], S. Siriroob[b], W. Meemetta[b], W. Santimanawong[b], W. Gangnonngiw[b,c], N. Pirarat[d], P. Khunrae[a], T. Rattanarojpong[a], R. Vanichviriyakit[b,e], S. Senapin[b,c,*]

Year	Farm/province	Species	Fish stage	Mortality (%)	Number of positive/ number tested
2017	A/Pathumthani	Nile tilapia *Oreochromis niloticus*	Fingerling (2.5–3 cm)	~90	14/14
2016	CL/Phetchaburi	Red and Nile tilapia	Fingerling (3.5–4 cm)	~20	7/7
	CN/Chainat	Red tilapia *Oreochromis* sp.	Fingerling (8.5–9 cm)	~90	6/6
	Control	Nile tilapia	Juvenile (~15 cm)	–	0/2



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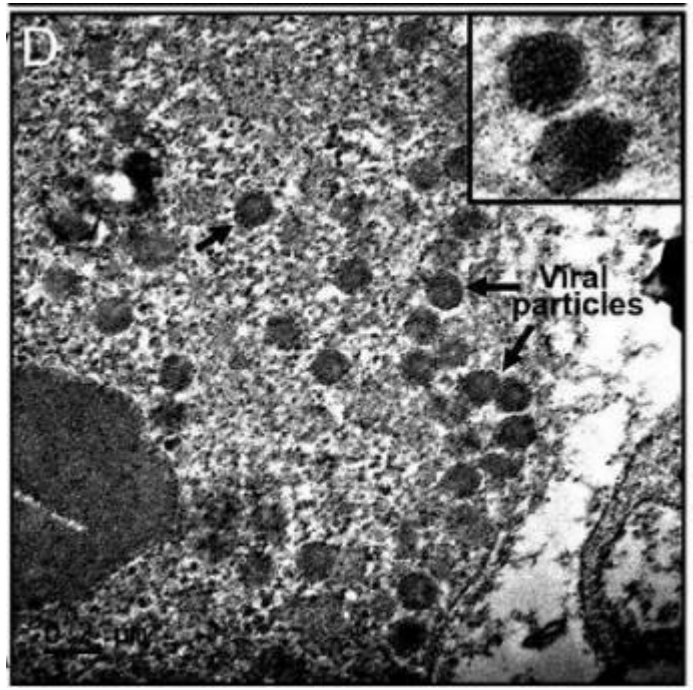
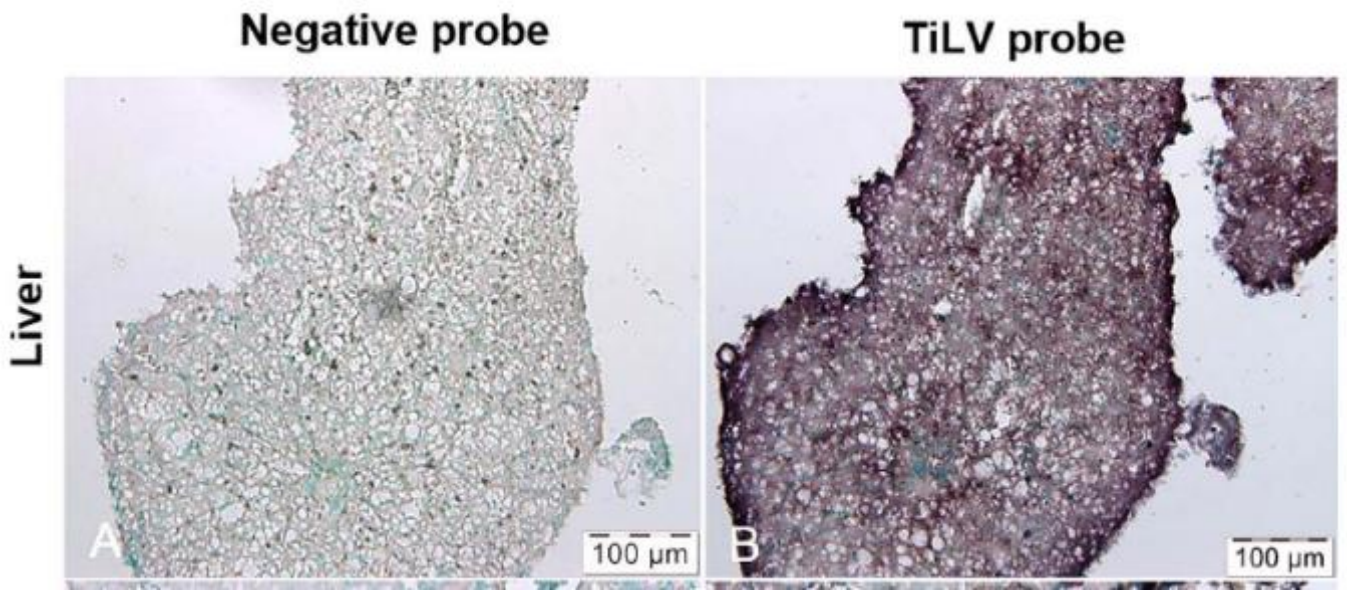


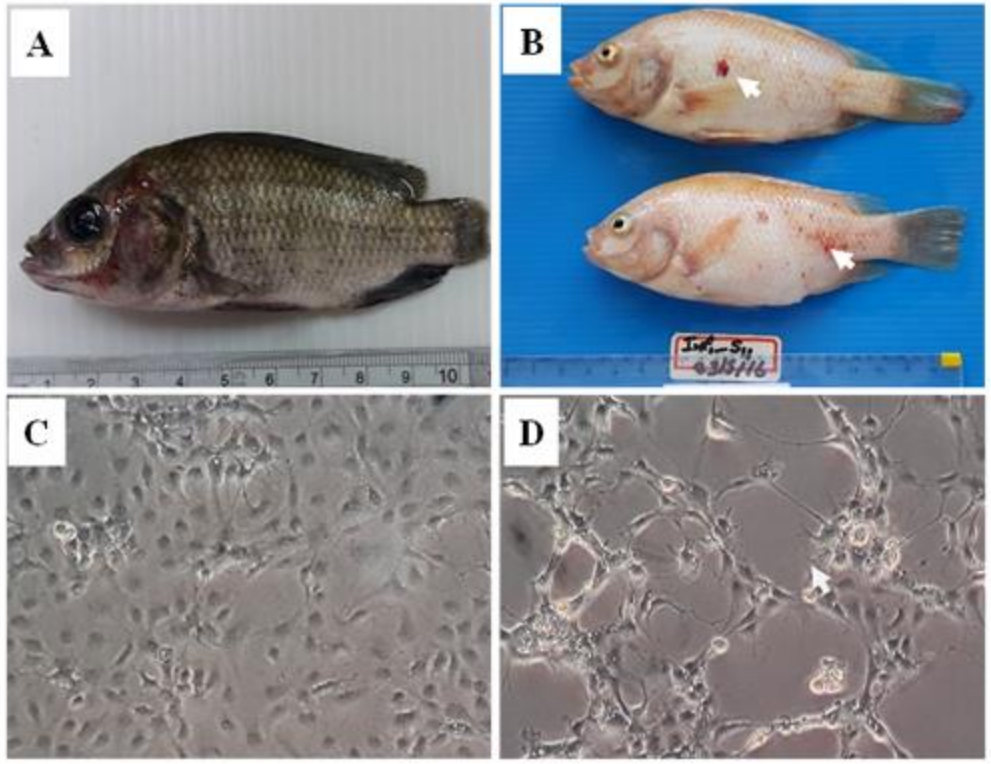


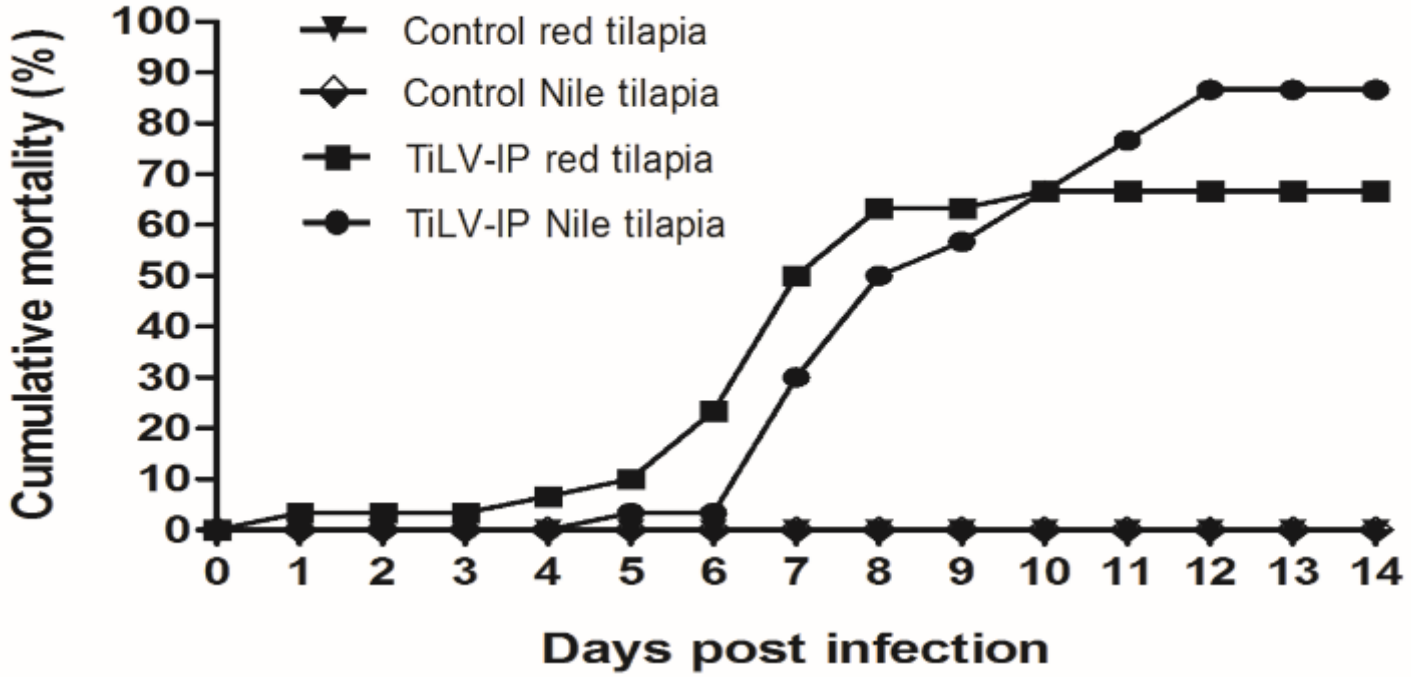
Veterinary Microbiology

Volume 207, August 2017, Pages 170–177

Experimental infection of Tilapia Lake Virus (TiLV) in Nile tilapia (*Oreochromis niloticus*) and red tilapia (*Oreochromis* spp.)

Puntanat Tattiyapong[a, b], Worawan Dachavichitlead[a, b], Win Surachetpong[a, b],





TiLV can cause disease in susceptible/normal tilapia

- **Koch's Postulates**

Veterinary Microbiology

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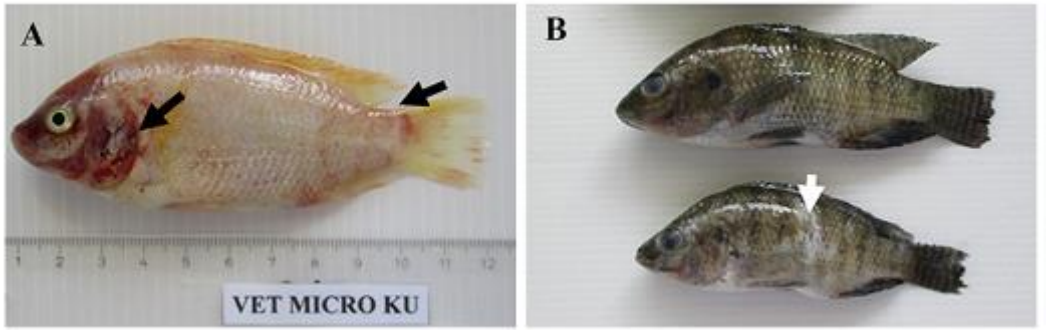
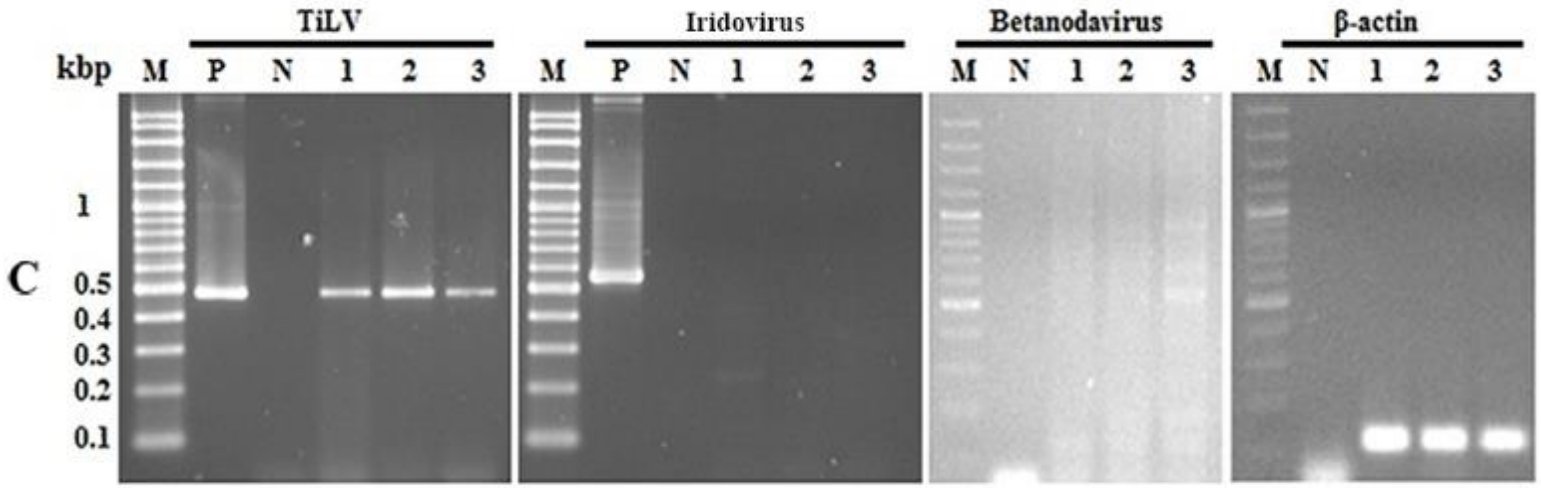
Puntanat Tattiyapong[a, b], Worawan Dachavichitlead[a, b], Win Surachetpong[a, b]

1. The pathogen is isolated from naturally exposed fish

- Skin redness, skin erosion, corneal opacity



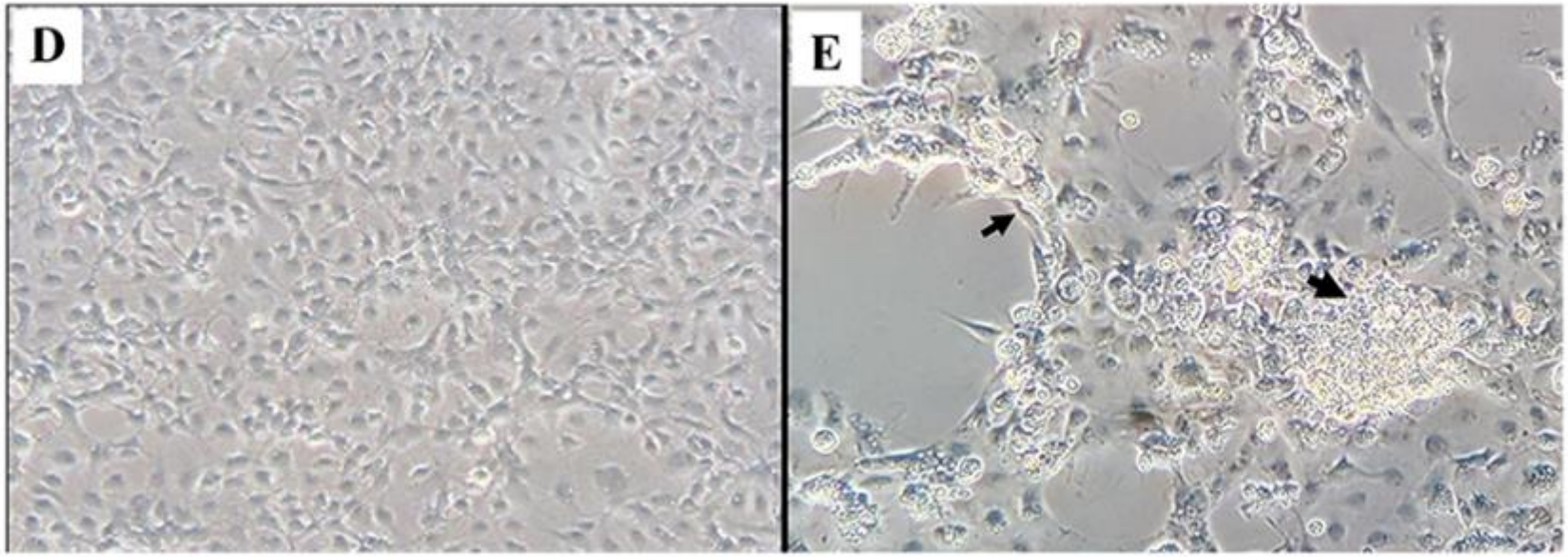
- Detection only TiLV, not other viruses



Tattiyapong, Dachavichitlead, and Surachetpong 2017. *Veterinary Microbiology. 207: 170-177*

2. The pathogen was isolated in E-11 cells

Normal brain **Infected brain**



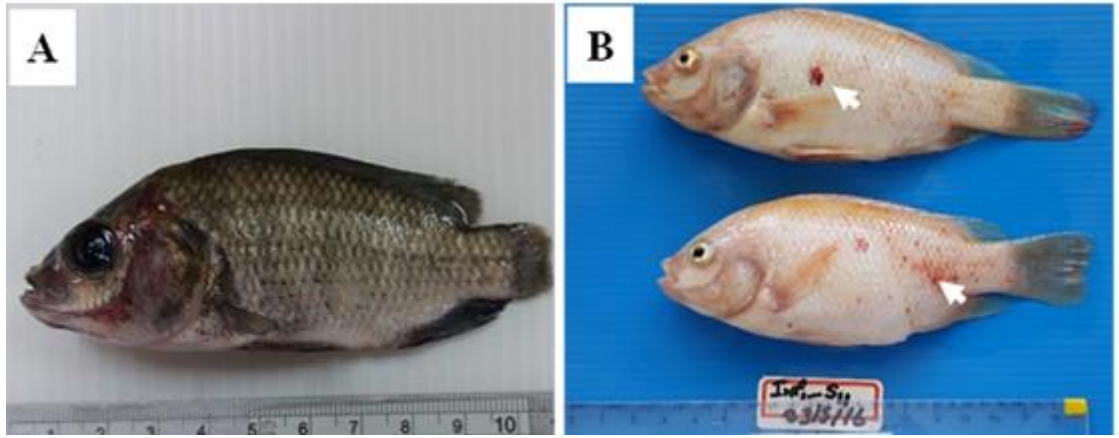
- **3-5 dpi, CPE were observed in E-11 cells inoculated with infected brain**

Tattiyapong, Dachavichitlead, and Surachetpong 2017. *Veterinary Microbiology. 207: 170-177*

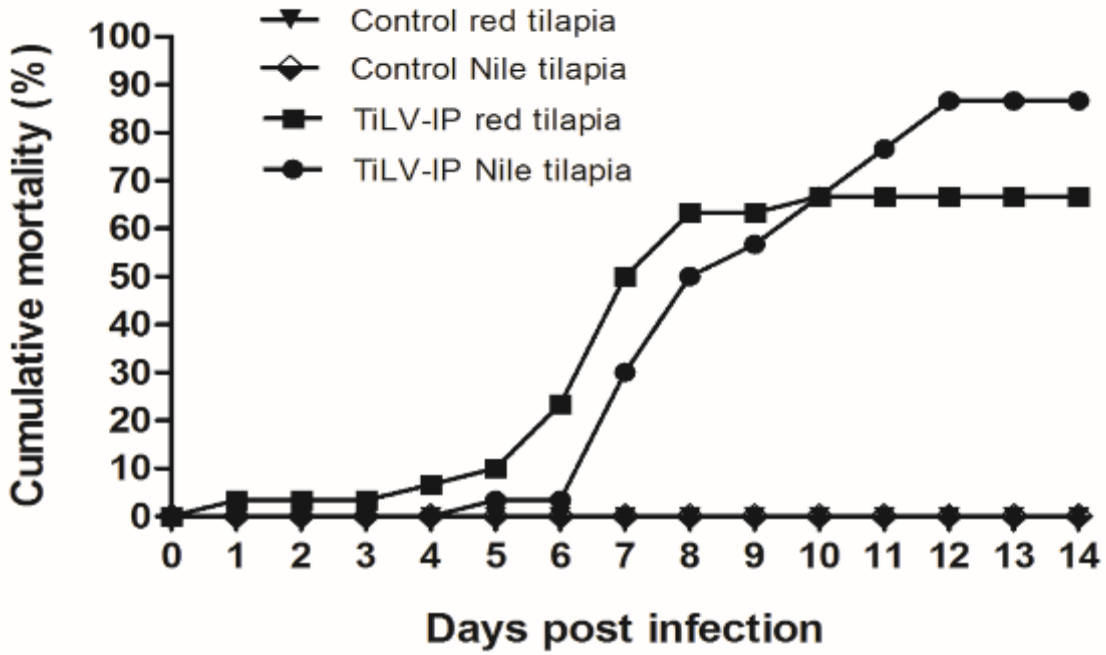
3. The pathogen caused disease in challenged fish

- **Challenged Nile and red tilapia developed clinical signs of TiLV infection with high mortality**

Clinical signs of challenged fish



Mortality rate



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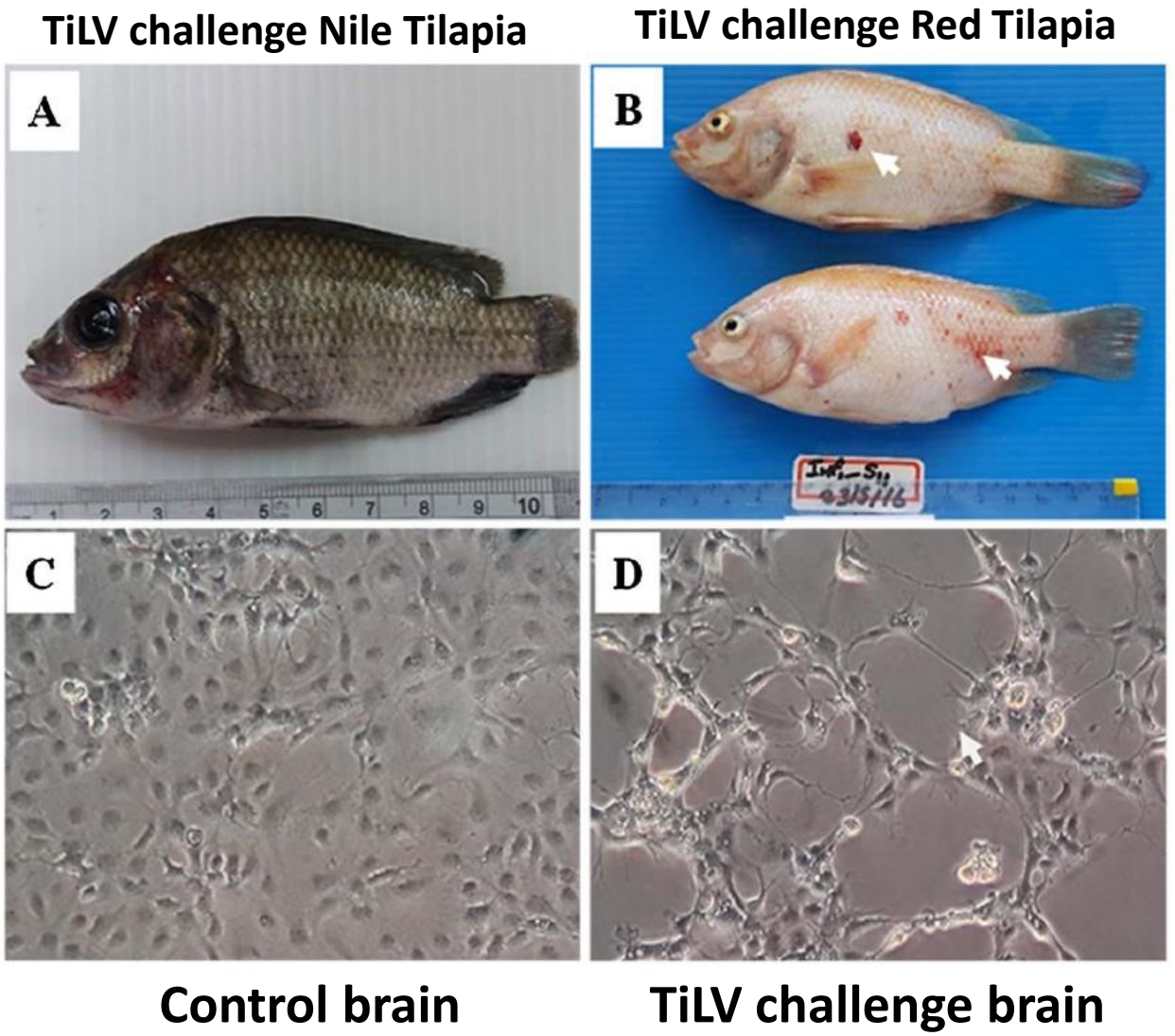
Clinical signs of challenged fish

4. The pathogen was isolated in E-11 cells

- **CPE formation in E-11 cells inoculated with brain from experimentally challenged tilapia**



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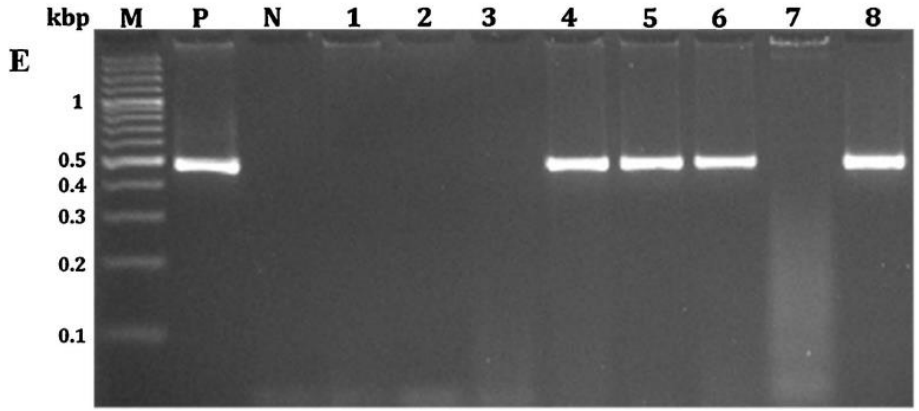
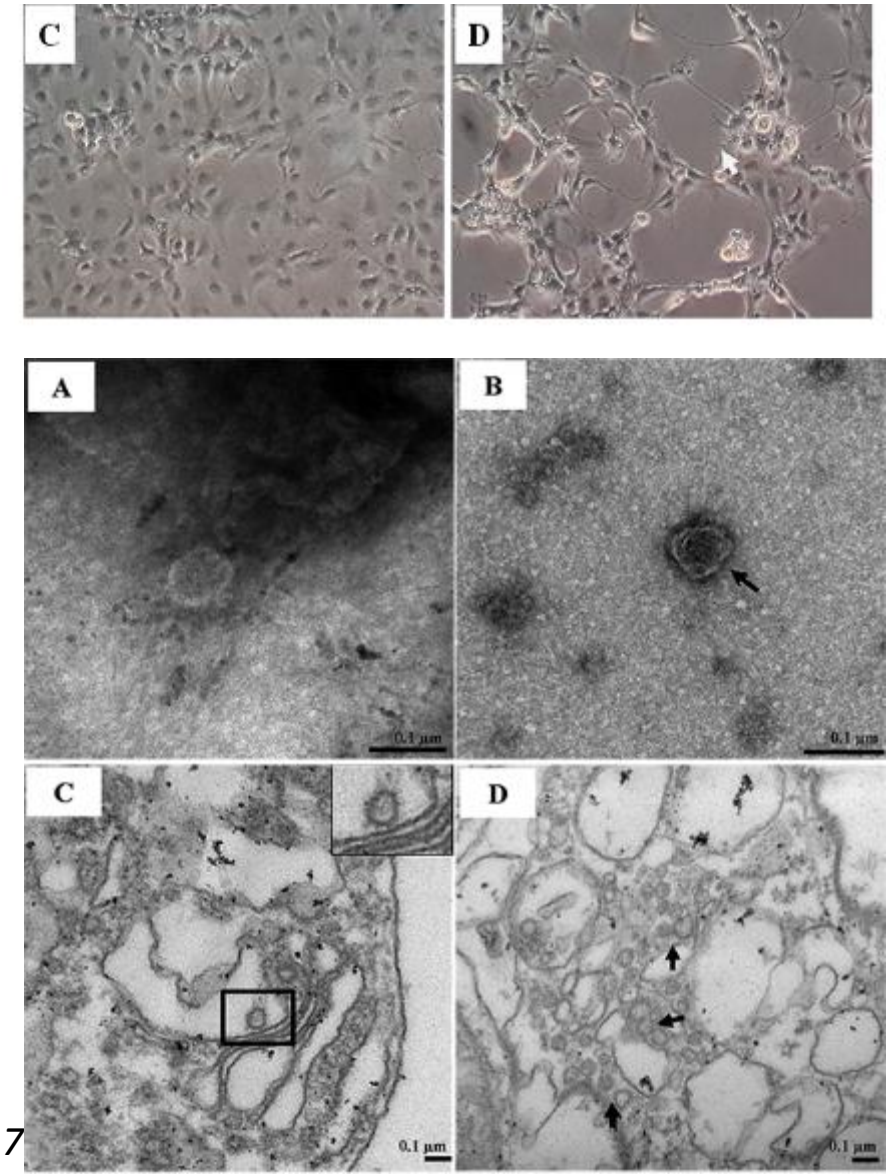
4. The pathogen was isolated from challenged fish

- **Confirmation of TiLV using RT-PCR and TEM (infected E-11, and brain tissues of challenged fish)**



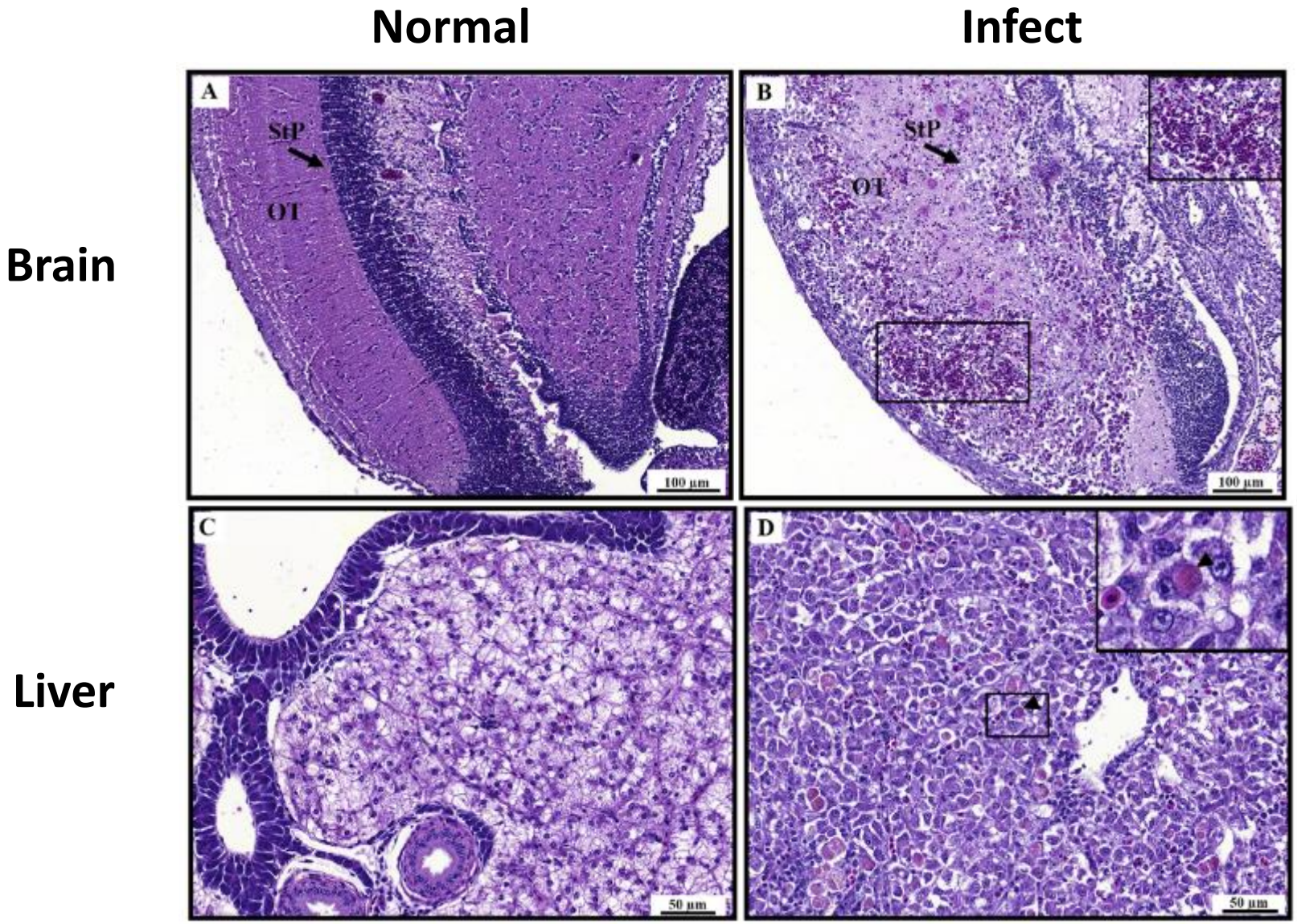
1-3: brain from PBS injected fish

4-7: brain from TiLV injected fish



Tattiyapong, et al 2017. *Veterinary Microbiology. 207: 17*

Histopathology of TiLV-challenged fish



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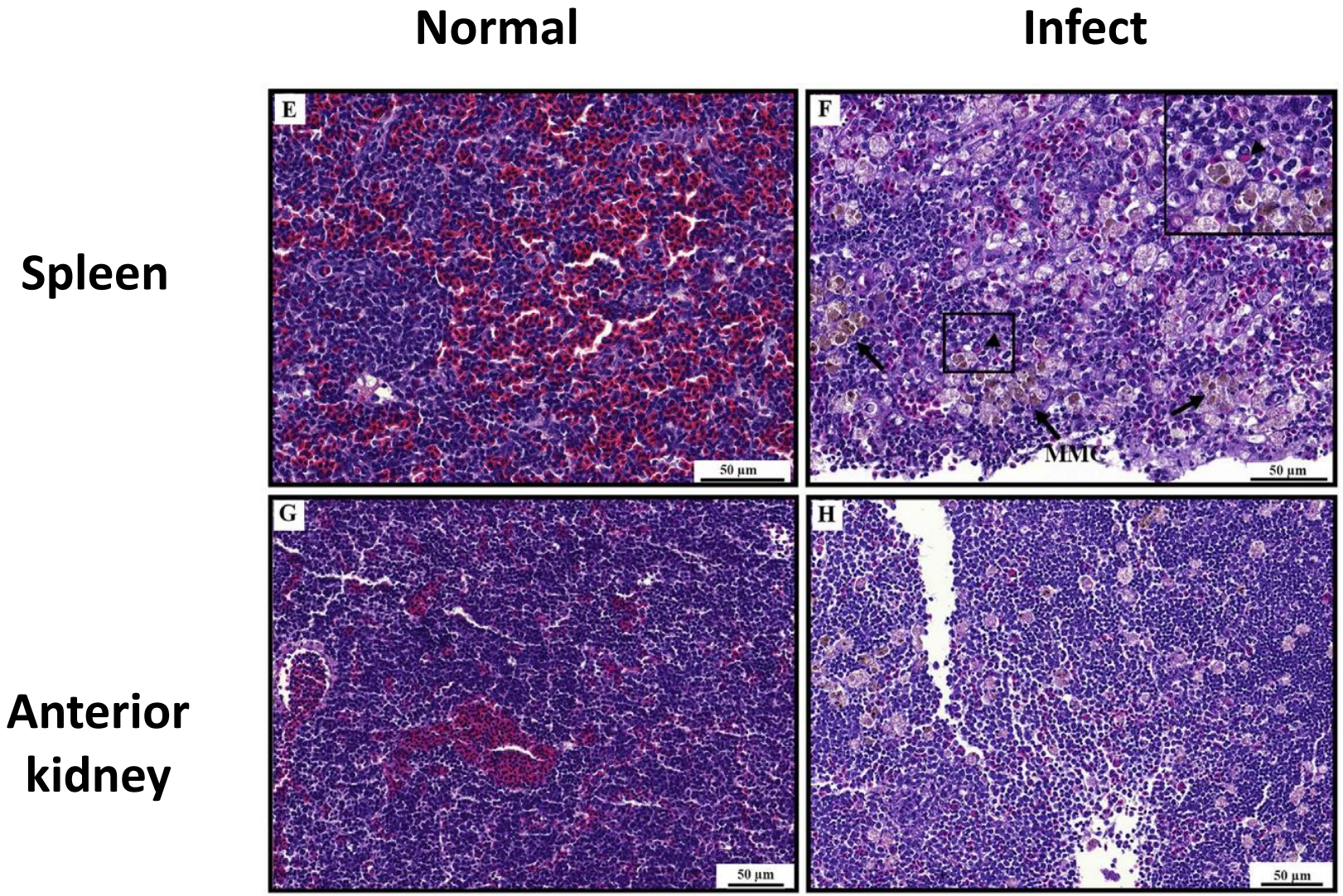
Histopathology of TiLV-challenged fish



Tattiyapong, Dachavichitlead, and Surachetpong 2017. *Veterinary Microbiology. 207: 170-177*

Aquaculture 492 (2018) 206–214

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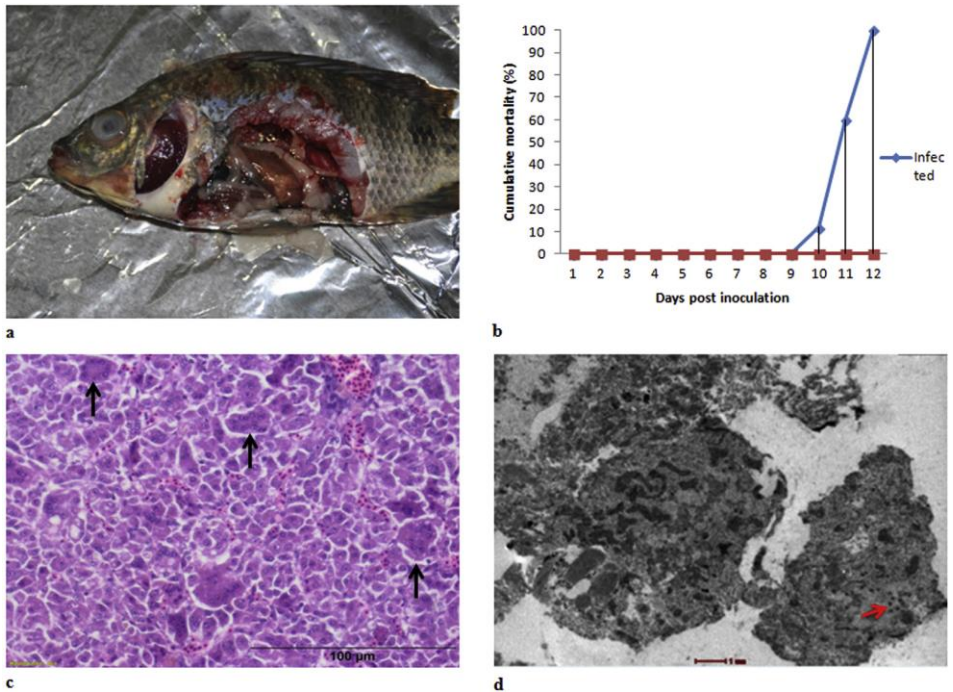
journal homepage: www.elsevier.com/locate/aquaculture

Derivation of two tilapia (*Oreochromis niloticus*) cell lines for efficient propagation of Tilapia Lake Virus (TiLV)

Raja Swaminathan Thangaraj[a,*], Charan Ravi[a], Raj Kumar[a], Arathi Dharmaratnam[a], Basheer Valaparambil Saidmuhammed[a], Pravata Kumar Pradhan[b], Neeraj Sood[b]



- **Experimental challenge of virus isolated from *O. niloticus* cell line**

SHORT COMMUNICATION

WILEY Journal of Fish Diseases

Detection of Tilapia Lake Virus in Egyptian fish farms experiencing high mortalities in 2015

P Nicholson[1*] | M A Fathi[2,3*] | A Fischer[4] | C Mohan[5] | E Schieck[4] | N Mishra[6] | A Heinimann[7] | J Frey[1] | B Wieland[8] | J Jores[1,4]

Bacteria in the genus *Aeromonas* are commonly isolated from TiLV infected fish

Farm ID	Diseased fish/total fish sampled Morbidity rate (%)[a]	TiLV detected	*Aeromonas* species detected
1	7/13 (54%)	–	*A. veronii* *A. hydrophilia*
2	14/26 (54%)	–	*A. veronii*
3	13/24 (54%)	+	*A. veronii*
4	13/30 (43%)	–	*A. veronii* *A. ichthiosmia* *A. enteropelogenes*
5	21/40 (53%)	+	*A. veronii*
6	14/20 (70%)	–	*A. veronii* *A. enteropelogenes* *A. jandaei*
7	8/24 (33%)	+	*A. veronii* *A. ichthiosmia*
8	10/10 (100%)	+	*A. enteropelogenes* *A. hydrophilia*
WF	0/20 (0%)	–	*A. veronii*

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Short communication

A case of natural co-infection of Tilapia Lake Virus and *Aeromonas veronii* in a Malaysian red hybrid tilapia (*Oreochromis niloticus* × *O. mossambicus*) farm experiencing high mortality

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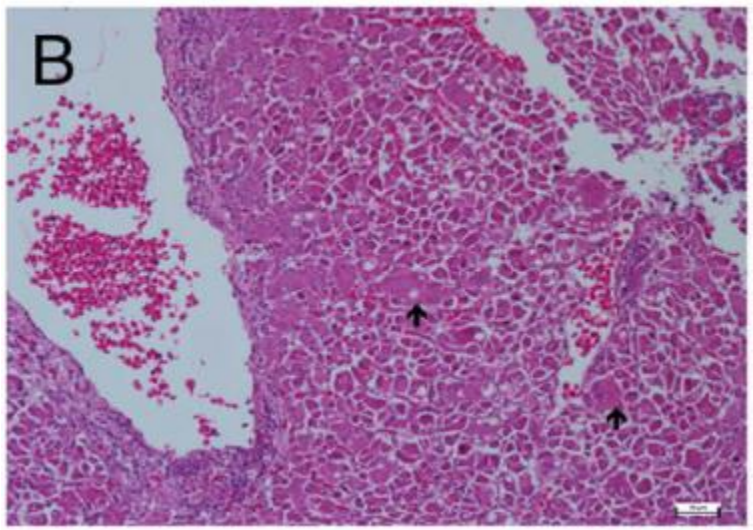

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ORIGINAL ARTICLE

WILEY Journal of Fish Diseases

Detection of tilapia lake virus (TiLV) infection by PCR in farmed and wild Nile tilapia (*Oreochromis niloticus*) from Lake Victoria

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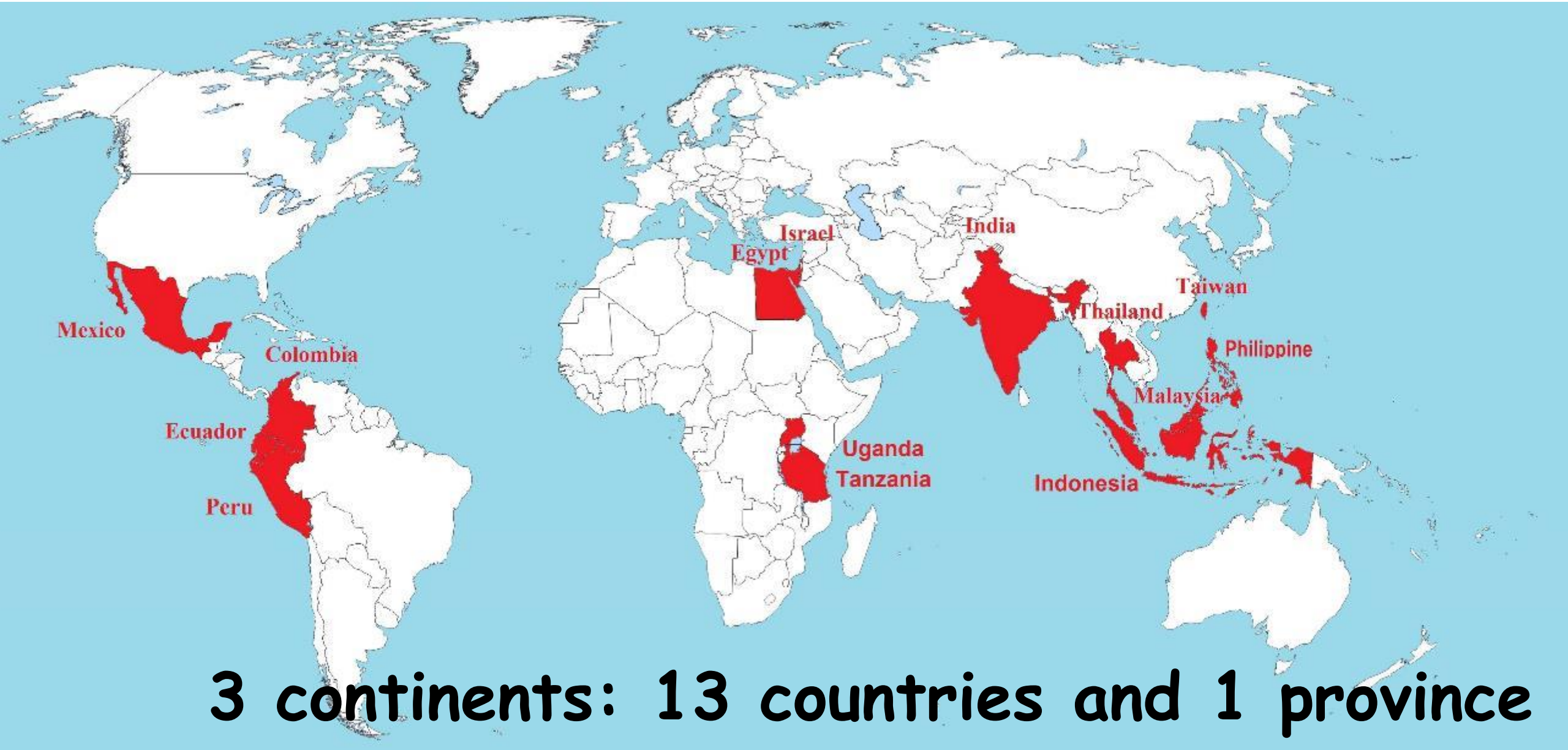
- **Examined 191 fish, 28 showed positive TiLV by PCR**
- **High prevalence in head kidney and spleen (lymphoid tissues)**

Genetic variation

Source (non-Israeli sources)	GenBank accession no.	Identity to TiLV from Israel (prototype strain)		References
		GenBank accession no. of Israeli TiLV	% nt identity	
Chinese Taipei	Not available	Segment 3 (Accession number not specified)	93%	OIE (2017b)
Ecuador	Not available	Full genome sequences KU751814–KU751823	97.2–99.0%	Bacharach *et al.* (2016a)
Ecuador	Not available	KJ605629 (ORF)	98% to 100%	del-Pozo *et al.* (2017)
Egypt	Not available	KU751816 (segment 3)	93%	Fathi *et al.* (2017)
Egypt	KY817381–KY817390	Segments 3, 4 and 9 (Accession numbers not specified)	93%	Nicholson *et al.* (2017)
India	MF502419, MF574205 and MF582636	KJ605629 (segment 3)	96.4–97.2%	Behera *et al.* (2018)
Indonesia	Not available	KU751816 and KJ605629 (segment 3)	97%	Koesharyani *et al.* (2018)
Malaysia	MF685337	KU751822 (segment 9)	97%	Amal *et al.* (2018)
Philippines	Not available	Segment 3 (Accession number not specified)	94–95%	OIE (2017f)
Tanzania (Lake Victoria)	MF526980–MF526996	KU552132 (contig 7 = segment 2) KU751815 (= NC029921, segment 2)	Not given†	Mugimba *et al.* (2018)
Thailand	KY615742	KU751814 (segment 1)	96.3–97.5%	Dong *et al.* (2017a)
Thailand	KY615743	KU751818 (segment 5)		
Thailand	KY615744 to KY615745	KU751822 (segment 9)		
Thailand	KX631921 KX631930–KX631936	Full genome sequences KU751814–KU751823	95.6–99.1%	Surachetpong *et al.* (2017)
Uganda (Lake Victoria)	MF536423–MF536432	KU552132 (contig 7 = segment 2) KU751815 (= NC029921,segment 2)	Not given†	Mugimba *et al.* (2018)

†Authors state that sequences were 'identical with' or 'closely related to' the Israeli sequences.

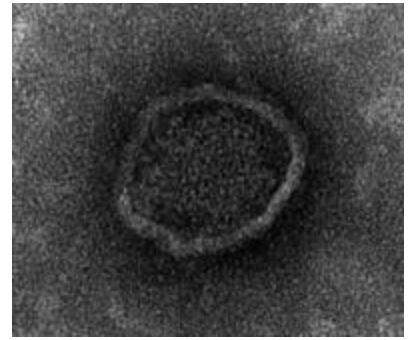
Current distribution of TiLV



3 continents: 13 countries and 1 province

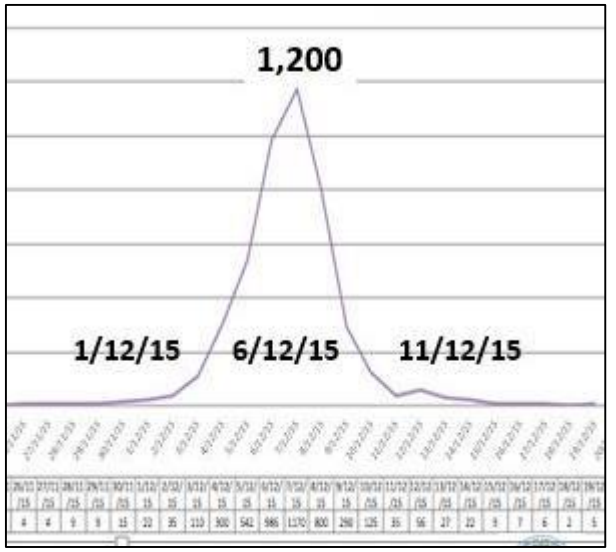
Current information on TiLV

- RNA, envelope, Orthomyxo-like virus
- Mainly affect tilapia, closely related species and giant gourami
- No report of harm to human or other animals
- **Strict biosecurity** reduces the impact of disease and spreading of the virus
- Not on the **OIE disease list**



How do we know if TiLV exists in a farm?

- High mortality **20-90%**
- Swimming at water surface
- Skin redness, erosion
- Red tilapia (pale body)
- Exophthalmos,
  scale protrusion




Daily mortality above 1-2% for 3 consecutive days
60-100 fish/cage Fish size 20 g
No history of high mortality since stocking
Win Surachetpong

Economic impact of TiLV & TOMMS

- Fry and Fingerling loss → Fry shortage
  - Mortality 50-90% (2-4 million US dollars)
- More impact on red tilapia than Nile tilapia
  - Shift from red to Nile tilapia

- Survivor → price ↑ ~30 g → 10-30 cent per fish
- Elimination of positive broodstock?
- Cost of farm management

Take home messages….

- **Tilapia Lake Virus has been detected since 2009**
- **To date, TiLV positive samples were reported in 14 countries**
- **Multiple infections of TiLV, bacteria and parasites are commonly found: Summer mortality, One month mortality syndrome**

Acknowledgements

WS lab members

Thank you all farmers