The 4th International Conference on Fisheries and Marine Sciences (INCOFIMS) Surabaya Indonesia, 29 September 2021

The 4th International Conference on Fisheries and Marine Sciences (INCOFIMS) Surabaya Indonesia, 29 September 2021

International conference on fisheries and marine sciences (INCOFIMS) is an annual conference organized by Faculty of Fisheries and Marine Universitas Airlangga, Surabaya, Indonesia. The main aim is to provide a sharing platform that enables researchers, academics and practitioners from all over the world to share their most recent findings as well as to propose the best strategies to address issues and challenges which we have been currently facing in aquaculture and fisheries practices worldwide. The 1st INCOFIMS was held successfully offline in Surabaya in 2018, the second in 2019 and the third in 2020.

The 4th INCOFIMS was previously scheduled offline in Surabaya on 29th September 2021. However, due to the Covid-19 pandemic and travel restriction for foreigners come into Indonesia as well as traveling within the Indonesian islands, we had the 4th INCOFIMS in a virtual format with ZOOM on 29 September 2021, and hosted from Faculty of Fisheries and marine, Universitas Airlangga, Surabaya Indonesia. We were unable to postpone the event because INCOFIMS is our annual event and also most of the participants requested to have the conference in the virtual format (online)

The theme in the 4th INCOFIMS was “Interprofessional collaboration for enhancing the aquatic ecosystem sectors”. Technically, we had the conference divided into 2 (two) sessions in general: (1) keynote speaker session and (2) guest speaker session. In the keynote session, we had 4 (four) keynote speakers delivering a speech which were Prof Felipe Polivanov Ottoni, Ph.D.; Asst. Prof. Dr. Narongrit Muangmai and Dr. TB. Haeru Rahayu, A.Pi., M.Sc. Each keynote speaker had 1.5 hours for giving a presentation using ZOOM and 30 minutes for discussion in one virtual room. After the keynote speaker session, we proceeded to the guest speaker session in which all participants were divided into 7 (seven) rooms according to our subtopics for oral and poster presentations:

Room 1: Aquaculture technology
Room 2: Fish Nutrition
Room 3: Fish Diseases
Room 4: Marine and Aquatic Sciences
Room 5: Estuarine and Coastal Ecosystems
Room 6: Fisheries Management
Room 7: Fisheries Socio-economics

In this session, every speaker had 15 minutes for presentation and 5 minutes for discussion. Total participants joined in this conference was 225 participants from at least 6 different countries (Australia, Japan, Austria, Malaysia, Taiwan and Indonesia).

The conference was in general quite successful, acknowledging the number and enthusiasms of participants during the discussion sessions in both the keynote speaker session and guest speaker’s session. We thank all participants and organizing committee for their support to this conference and see you in the 5th INCOFIMS 2022.

Chairman
Veryl Hasan, Ph.D

Peer Review Statement

To cite this article: 2022 *IOP Conf. Ser.: Earth Environ. Sci.* **1036** 011002

Peer Review Statement

All papers published in this volume have been reviewed through processes administered by the Editors. Reviews were conducted by expert referees to the professional and scientific standards expected of a proceedings journal published by IOP Publishing.

- **Type of peer review:** Double Anonymous
- **Conference submission management system:** Morressier
- **Number of submissions received:** 195
- **Number of submissions sent for review:** 195
- **Number of submissions accepted:** 191
- **Acceptance Rate (Submissions Accepted / Submissions Received × 100):** 97.9
- **Average number of reviews per paper:** 2
- **Total number of reviewers involved:** 10
- **Contact person for queries:**
  Name: Dwi Yuli Pujiastuti, S.Pi., M.P.
  Email: dwiyp@fpk.unair.ac.id
  Affiliation: Faculty of Fisheries and Marine, Universitas Airlangga

Table of contents

Volume 1036

2022

Previous issue Next issue

The 4th International Conference on Fisheries and Marine Sciences (INCOFIMS 2021) 29/09/2021 - 29/09/2021 Online

Accepted papers received: 20 May 2022
Published online: 11 July 2022

Open all abstracts

Preface

The 4th International Conference on Fisheries and Marine Sciences (INCOFIMS 2021)

F Swastawati, P H Riyadi, M Mulyono, A Nugraheni, M Muniroh and A N Hidayati

+ Open abstract View article PDF

OPEN ACCESS 012011

Molecular phylogenetic of silver barb *barbonymus gonionotus* (bleeker, 1849) (cypriniformes: cyprinidae) in Java, Indonesia

F S Valen, M S Widodo, R A Islamy, K P Wicaksono, Soemarno, L Insani and V Hasan

+ Open abstract View article PDF

OPEN ACCESS 012012

Application of modified starch on glass bioplastic based on carrageenan from *Eucheuma cottonii* on mechanic and biodegradation properties

T A Triani, M A Alamsjah and D Y Pujiastuti

+ Open abstract View article PDF

OPEN ACCESS 012013

Analysis of Blood Glucose Levels and The Development of Ectoparasite Infestation on Pacific White Shrimp (*Litopenaeus vannamei*) Which Were Given Crude Protein *Zoothamnium penaei* at High Stocking Densities

G Mahasri, W H Satyantini and A T Mukti

+ Open abstract View article PDF

OPEN ACCESS 012014

Mitigation and adaptation to climate change through sustainable mangrove management on the coast of Rembang Regency

H A Sutanto, I Susilowati, D D Iskandar and Waridin

+ Open abstract View article PDF

OPEN ACCESS 012015

Nursery tecnique of gouramy fish *(osphronemus goramy)* at instalasi perikanan budidaya, pasuruan, east java

Jamal Asfani and Lailatul Lutfiyah

+ Open abstract View article PDF

OPEN ACCESS 012016

Acute Toxicity of domestic landfill leachate to carp fish (*Cyprinus carpio*)

H C Pratiwi, B S Rahardja and W Tjahjaningsih

+ Open abstract View article PDF

OPEN ACCESS 012017

Quality and Halal Certification of Micro and Small Enterprises Fishery Products in Sidoarjo, East Java, Indonesia

Sukoso, P T Al Huda, H Muyasyaroh, Y A D Susanti and L H Adila

+ Open abstract View article PDF

OPEN ACCESS 012018

Quality improvement formulation of salak chili paste paste based on curly red chili (*Capsicum annuum var. hailux*) and pondoh salak (*Salacca edulis reinw var. semeru*) using sensory evaluation, activity evaluation of bioactive compounds and microbiological evaluation

Joni Kusnadi, Arinta Julia Buwana Saputra, Siti Narsito Wulan and Estri Laras Arumingtyas

+ Open abstract View article PDF

OPEN ACCESS 012019

Broodstock rearing techniques brown-marbled grouper (*epinephelus fuscoguttatus*) at balai perikanan budidaya air payau (BPBAP) situbondo, east java

Dimas Khoironi Nur Septian, Darmawan Setia Budi, Suciyono and Lailatul Lutfiyah

+ Open abstract View article PDF

OPEN ACCESS 012020

The effect of barnacles powder as a source of protein feed substitution against the digestibility of crude protein and crude fiber on male rabbits rex

B Agustono, F Maulana, S H Warsito and M A Al-Arif

+ Open abstract View article PDF

OPEN ACCESS 012021

Diet and feeding strategy of the common silver-biddy, *Gerres oyena* (Forsskål 1775) in the seagrass beds of Karang Congkak Island, Kepulauan Seribu National Park

A K Putri, M F Nazal and C P H Simanjuntak

+ Open abstract View article PDF

OPEN ACCESS 012022

Sensory and Chemical Properties of Long Jawed Mackerel (*Rastrelliger kanagurta* L.) Fish Balls with Addition of Canna (*Canna edulis* Kerr.) Starch Concentration as a Filler

Novita Herdiana, A Sapta Z, Susilawati and Silaturahmi Widaputri

+ Open abstract View article PDF

OPEN ACCESS 012023

Comparison Effects Of Seaweed Concentrations On Total Bacteria And Yeast Kombucha *Gracilaria Verrucosa* During The Production Process

Wahyu Andrianto, Sudarno, Rahayu kusdarwati and dan Dwitha nirmala

+ Open abstract View article PDF

OPEN ACCESS 012024

Identification of *Escherichia coli* and *Salmonella* on Fishery Product from Juanda Airport, East Java, Indonesia

R Hidayah, M F Ulkhaq and R Wilis

+ Open abstract View article PDF

OPEN ACCESS 012025

Risk analysis of catfish cultivation (*Pangasius hypophthalmus*) business in Gondosuli Village, Gondang, Tulungagung

Supriyadi Supriyadi, Kartika Intan Abdillah and Mimit Primyastanto

+ Open abstract View article PDF

OPEN ACCESS 012026

Evaluation of the culture of *Spirulina sp.* with Walne nutrient plus vitamin B12, KCl, NPK, ZA CaO and urea

A L A Suyoso, L A Sari, P D W Sari, D D Nindarwi and dan S Arsad

+ Open abstract View article PDF

OPEN ACCESS 012027

Potential feed substitute of *Cirripedia* sp. flour on body weight gain, feed conversion ratio, feed consumption of buck

B Agustono, D A Agustin, E K Sabdoningrum, M N Yunita, R T Dewi and I R T Ivani

+ Open abstract View article PDF

OPEN ACCESS 012028

Analysis of polycyclic aromatic hydrocarbons (PAHs) bioremediation by hydrocarbonoclastic degrading bacteria (*Gordonia terrae*)

A. A. Amin, A. R. T. Wahyuni, A.W Ekawati and A. Kurniawan

+ Open abstract View article PDF

OPEN ACCESS 012029

Utilization of mangrove fruit (*Rhizophora stylosa*) as an alternative to low caffeinated drinks

S A Marctyas, D Y Pujiastuti and E Saputra

+ Open abstract View article PDF

OPEN ACCESS 012030

Trophic ecology and length-weight relationship of *Hypoatherina temminckii* (Bleeker, 1854) in the seagrass ecosystem of Semak Daun Island, Kepulauan Seribu

C P H Simanjuntak, A Zahid, P Pratiwi and A G Cahyani

+ Open abstract View article PDF

OPEN ACCESS 012031

Sublethal Toxicity Insecticide Organochlorine Endosulfan on The Value of Blood Sugar Levels and The Level of Life in Tilapia (*Oreochromis niloticus*) Seeds

S Nurhadi, E D Mashitah and dan L A Sari

+ Open abstract View article PDF

OPEN ACCESS 012032

Chitosan from snapper fish scale waste (*Lutjanus* spp.) for edible coating

A Florencia, S E D Putra and Y P Mukti

+ Open abstract View article PDF

OPEN ACCESS 012033

Application of modified starch on plastic bag bioplastic based on carrageenan from *Eucheuma cottonii* on mechanic and biodegradation properties

R A Sofianto, M A Alamsjah and D Y Pujiastuti

+ Open abstract View article PDF

OPEN ACCESS 012034

Characterization of Tilapia collagen-loaded chitosan nanofibers synthesized by electrospinning method for wound dressing

C A M Rani, A Safira, M Suryadiningrat, F Fikri, D K Wardhana and M T E Purnama

+ Open abstract View article PDF

OPEN ACCESS 012035

Determining of water bioremediation dosage in recirculating water system for Cantik Grouper (*Epinephelus fuscoguttatus* × *Epinephelus microdon*) nurseries

B Astari, S Ismi and K Mahardika

+ Open abstract View article PDF

OPEN ACCESS 012036

The Effect of Laserpuncture on Accelerate Gonadal Maturity of Female Striped Catfish (*Pangasianodon hypophthalmus*)

M Patmadevi, A T Mukti, A S Mubarak and S D Astuti

+ Open abstract View article PDF

OPEN ACCESS 012037

Nutritional Fact of Traditional Cracker (Amplang and Getas) from Bangka Belitung

Arief Rubiana Basarah, Patmawati, Muhamad Amin, Ahmad Fahrul Syarif, Mu'alimah Hudatwi and Sapto Andriyono

+ Open abstract View article PDF

OPEN ACCESS 012038

Characterizations of Milkfish Sauce on Amino Acid Content with Variations in Addition of Salt and Pineapple Extract Concentration

N. P. Listyaningrum, R. Lailatussifa and T. R. Andayani

+ Open abstract View article PDF

OPEN ACCESS 012039

Polysaccharide isolation of Brown seaweed: *Sargassum* sp and its photoprotection activity

M A P Panjaitan, R D Kasitowati, N Putri, A Yamindago, R Asmara, D Aliviyanti and D C Pratiwi

+ Open abstract View article PDF

OPEN ACCESS 012040

Proximate Analysis of Fisheries Dried Products from Mamuju Traditional Market Post-Earthquake Disaster

Eka Saputra, Wahyu Isroni, Arief Rubiana Basarah and Sapto Andriyono

+ Open abstract View article PDF

OPEN ACCESS 012041

The Intensive Program of Hatchery Aquaculture Performance of Gourami (Osphronemus gouramy) in IPB Pandaan

V B Pribawastuti and S H Samara

+ Open abstract View article PDF

OPEN ACCESS 012042

The effect of different maceration solutions towards characteristic gelatin from bone of common carp (*Cyprinus carpio*)

E Ristyanti, J Triastuti and E Saputra

+ Open abstract View article PDF

OPEN ACCESS 012043

Growth pattern and abundance of *Varuna Literatta* (Fabricius, 1798) in the new industrial area in the north coast of Gresik, East Java

F Fariedah, M S Widodo and R Yuwanita

+ Open abstract View article PDF

OPEN ACCESS 012044

The Effect of Rumen's Cattle Waste as Alternative Nutrient Sources for *Thalasiossira* sp Aquaculture

M B Santanumurti, M I Ogara, L Santoso and S Hudaidah

+ Open abstract View article PDF

OPEN ACCESS 012045

Inhibitory Rate of Chitosan Against *Vibrio harveyi* use in vitro method

Haryagita, R Kusdarwati and H Suprapto

+ Open abstract View article PDF

OPEN ACCESS 012046

Measurement of Indonesian Marine Health Index to Assess The Health of The Coastal Ecosystem of Tuban, East Java

M I Joesidawati and S Suwarsih

+ Open abstract View article PDF

OPEN ACCESS 012047

Physical characteristic and antioxidant activities of liquid bath soap with substitution of β-carotene crude extract from *Gracilaria* sp.

M M Ircham, A S Mubarak and E Saputra

+ Open abstract View article PDF

OPEN ACCESS 012048

Cultivation of Phytase-Producing Bacteria as Probiotic Candidate on Molasses and Tempe-Processing Waste

Zaid Al Gifari, Ihza Agistna, Khairil Anwar, Anwar Rosyidi, Muhamad Ali and Muhamad Amin

+ Open abstract View article PDF

OPEN ACCESS 012049

Effectiveness of striped catfish (*Pangasianodon hypophthalmus*) cultivation in aquaponic system with three different plant against ammonia (NH_3), nitrite (NO_2), and nitrate (NO_3)

S Andriani, N N Dewi and B S Rahardja

+ Open abstract View article PDF

OPEN ACCESS 012050

The effect of different feed on Ctenopoma (*Ctenopoma acutirostre*) production through survival rate and growth

M B Santanumurti, L Alfarisi, L Santoso and S Hudaidah

+ Open abstract View article PDF

OPEN ACCESS 012051

Potential of sappan wood extract as a feed additive on the feed conversion on organic matter digestibility on the buck exposed to heat stress

B Agustono, I Rahmaniyah, F Fikri and R A Prastiya

+ Open abstract View article PDF

OPEN ACCESS 012052

Reproductive ability of endemic species of Lake Ajamaru *Melanotaenia ajamaruensis* Allen & Cross, 1980 on spawning at various sex ratio

D S Said, N Mayasari and dan T Kadarini

+ Open abstract View article PDF

OPEN ACCESS 012053

Diversity of holothuroidea by spicule type in the intertidal zone of Sepanjang Beach, Gunungkidul, Yogyakarta

N Yunita, A Amalia, F A Ponkiyawati, L N Janah, L U Khasanah, M B Gavintri, Z Nurahmah, F B R Khalallia and R Epilurahman

+ Open abstract View article PDF

OPEN ACCESS 012054

Length-weight relationships and condition factors of fifteen fish species from Pabean Bay, West Java, Indonesia

L S Syafei, D Sudinno and I N Y Parawangsa

+ Open abstract View article PDF

OPEN ACCESS 012055

Study on the potential of sea urchin *Tripneustes gratilla* (Linnaeus, 1758) as a bioindicator dangerous plastic pollution in environment of gunungkidul beach Yogyakarta

S D Pratita, M B Gavintri, A N Rizkyta, L U Khasanah, F A Ponkiyawati and B Retnoaji

+ Open abstract View article PDF

OPEN ACCESS 012056

Opportunities and its problem for the development of Tilapia aquaculture business in Banjar Regency, South Kalimantan

N Nurlaili and H Hikmah

+ Open abstract View article PDF

OPEN ACCESS 012057

The dominance and proportions of plankton in Pacific white shrimp (*Litopenaeus vannamei*) ponds cultivated with the intensive system in Bulukumba Regency, South Sulawesi, Indonesia

M D Sani, P A Wiradana, A Y Maharani, R E Mawli and A T Mukti

+ Open abstract View article PDF

OPEN ACCESS 012058

The effect of bromelain enzyme on pineapple core on the relationship between platelet count and hematocrit value in carp (*Cyprinus carpio*) infested with *Argulus japonicus*

A B Fahturohman, Kismiyati and Y Rahmawati

+ Open abstract View article PDF

OPEN ACCESS 012059

Diversity of intertidal fishes in Porok Beach, Gunungkidul, Yogyakarta

E M Azizah, N Puspaningrum, D R Palupi, A A Fadlillah, A R Ismail, E T Nugroho, B M Effendi, F R Palupi and Z N Putri

+ Open abstract View article PDF

OPEN ACCESS 012060

Analysis of causes of changes in conditions of vannamei shrimp hepatopancreas indicators at PT. Lombang Sumber Rejeki Sumenep

T A Setyastuti, D Sukamto and I E Fawwaz

+ Open abstract View article PDF

OPEN ACCESS 012061

The effect addional omega-3 in feed on the gonadal maturity level and fecundity of female silver rasbora, *Rasbora argyrotaenia*

H Nabila, A S Mubarak and D S Budi

+ Open abstract View article PDF

OPEN ACCESS 012062

Climate change impact on shrimp (*Litopenaeus vannamei*) farming in Banyuwangi, East Java

M Yuniartik, E W Setyaningrum, S H Yuniari, S R Faturakhmat and H Prasetio

+ Open abstract View article PDF

OPEN ACCESS 012063

The Growth and Bacteriocin Productions of *Enterococcus Faecium* Cultured in Aerobic and Anaerobic Conditions

Muhamad Ali, Melinda Sanggu Artha, Muhamad Aidil Fadjar Suryadi, Djoko Kisworo and Muhamad Amin

+ Open abstract View article PDF

OPEN ACCESS 012064

Evaluation on feeding with sente leaves and enrichment on the conditioning of giant gourami (*Osphronemus gouramy*) broodstock

S Nashrullah, L A Sari and S Arshad

+ Open abstract View article PDF

OPEN ACCESS 012065

Molecular and phylogenetic analysis of *Sardinella lemuru* Bleeker 1835 at fishing ground Canggu-Bali inferred D-loop mutations of mtDNA

V A Winata, R D Kasitowati, F Iranawati, W S Pranowo and A Sartimbul

+ Open abstract View article PDF

OPEN ACCESS 012066

Description of Silver Barb *Barbonymus gonionotus* (Bleeker, 1849) (Cypriniformes: Cyprinidae) From Madura Island, Indonesia

F S Valen, V Hasan, F P Ottoni, A L Nafisyah, M Erwinda and A N Annisa

+ Open abstract View article PDF

OPEN ACCESS 012067

The Ecological Habitat of Spiny Lobster (*Panulirus* spp.): Case Study on Lobster Fishing Ground in Trenggalek, East Java, Indonesia

Muhamad Amin, Anis Fitria, Nur Aini Muslichah and Laila Musdalifah

+ Open abstract View article PDF

Bacterial Viability of *Edwardsiella tarda* from Silver Rasbora (*Rasbora argyrotaenia*) after Infection with Immmersion Methods

Bacterial Viability of *Edwardsiella tarda* from Silver Rasbora (*Rasbora argyrotaenia*) after Infection with Immmersion Methods

N Husna[1], R Kusdarwati[2], M F Ulkhaq[2*].

[1]Program Study of Aquaculture, Faculty of Fisheries and Marine, Airlangga University Campus Banyuwangi, 113 Wijayakusuma Street, Banyuwangi District, East Java, Indonesia
[2]Departement of Aquaculture, Faculty of Fisheries and Marine, Airlangga University, Campus C, Mulyorejo Street, Surabaya, East Java, Indonesia.

[*]*Corresponding author:* m-faizalulkhaq@fpk.unair.ac.id

Abstract. Silver rasbora (*Rasbora argyrotaenia*) is a freshwater fishery commodity that has high economic value. However, fulfilling the demand for silver rasbora still relies on catches from nature, so cultivation is needed. The problem that occurs in the cultivation process is the *Edwardsiella tarda* infection which causes Edwardsiellosis disease. The purpose of this study was to determine the bacterial viability of *E. tarda* from silver rasbora after infection with immersion methods. The Total Plate Count (TPC) from blood, liver and kidney was taken from infected fish after 14 days immersion with bacterial suspension. The results showed that *E. tarda* infection had occurred in the blood, liver and kidneys as indicated by an increasing the density of bacteria in each organ along with the increasing of the concentration of infected bacteria. The highest density of *E. tarda* bacteria infected in silver rasbora was in the blood and the least was in the kidneys.

1. Introduction

Silver rasbora (*Rasbora argyrotaenia*) is a freshwater fishery commodity that has high economic value [1]. However, the fulfillment of the demand for silver rasbora still relies on catches from nature so that it can cause a decrease in fish resources [2]. Fish farming is the alternative activity to fulfill the increasing market needs. According to [3], there are several factors that influence fish farming activities, including water quality management, feed, cultured organisms, and disease control. One of the problems that occur in the cultivation process that causes economic losses is the infection of pathogenic bacteria such as *Edwardsiella* which causes Edwardsiellosis disease [4,5]. *Edwardsiella* bacteria are opportunistic bacteria that infect freshwater and marine fish [6,7].

E. tarda infection can cause exophthalmia, red spots on the abdomen, abdominal swelling, bleeding on the fins and skin [8,9,10]. *E. tarda* can producing hemolysin, and dermatoxin [11,12]. Furthermore, *E. tarda* has a bacterial protein secretion system, including Type I to Type VI (T6SS) system which can penetrate, survive, and replicate in epithelial cells and phagocytes cells [13,14]. Previous study reported that *E. tarda* bacteria can infect freshwater fish species *Catla catla* from the family Cyprinidae [15]. This indicates that *E. tarda* also has the potential to infect silver rasbora, which are freshwater fish from similar family [16].

The increased viability of bacteria in the fish body due to the immune system being unable to fight infection causes *E. tarda* to spread and infect deeper tissues, reaching the circulatory and lymphatic

systems so that it can affect the survival of fish [17]. This can affect the work of internal organs, such as the liver, kidneys, spleen, and host muscles [18,19].

However, research on the bacterial viability of *E. tarda* infected in silver rasbora (*R. argyrotaenia*) is still limited. The purpose of this study was to determine the bacterial viability of *E. tarda* from silver rasbora after infection with immersion methods. The data obtained are used as a reference for the density of bacteria in the waters capable of infecting silver rasbora to accelerate the process of handling and preventing Edwardsiellosis disease in silver rasbora cultivation.

2.Methods

This research was carried out from October 2020 to February 2021 at the Instrument Laboratory of the Faculty of Fisheries and Marine PSDKU Airlangga University in Banyuwangi.

2.1 Experimental Fish and Bacterial Preparation

Silver rasbora (*Rasbora argyrotaenia*) as many as 200 fish with a length of 5.4-7 cm and a weight of 0.47±2.63 grams. All fishes was immersed in NaCl solution at a dose of 30 ppm for 5 minutes to remove the ectoparasites and then acclimatized for approximately 7 days. *E. tarda* bacteria was identified biochemically using Gram staining, oxidative/fermentative test, SIM test, catalase test and oxidase test [20].

2.2 Bacterial infection

Bacterial infection of *E. tarda* in silver rasbora was carried out by immersion for 14 days in twenty four aquariums (40x40x30 cm, 10 liters water) with density of 10 fish/aquarium containing *E. tarda* bacteria with a density of 10^{11} CFU/mL, 10^{12} CFU/mL, and 10^{13} CFU/mL and without bacterial immersion as a negative control.

2.3 Blood, Liver and Kidney Sampling

Blood, liver and kidney samples were taken from 10% of the total population of the test fish in order to represent all the test fish. The fish used as samples were fish that did not show clinical signs or fish that showed clinical symptoms of infection with *E. tarda*. Sampling organs was carried out before infection and 14 days after infection. Samples from blood, liver and kidneys that have been obtained will be diluted starting from 10^{-2}, 10^{-4}, 10^{-6}, and 10^{-8}, using physiological NaCl, then spread on *Xylose-Lysine Deoxycholate Agar* selective media (XLD) to be cultured and then incubated (28^{0}C) for 24 hours. The density of the growing *E. tarda* bacterial colonies was calculated using the TPC (*Total Plate Count*) method using a hand counter.

2.4 Statistical Analysis

The colony of bacterial in blood, wounds, liver and kidney were analyzed stratistically using Analysis of Variance (ANOVA) using IBM SPSS 20 software (α=0.05). If it is significantly different, Duncan's Multiple Range Test (DMRT) (95% confidence interval) were used to analyse the significance between all treatments. Clinical symptom data were analyzed descriptively.

3.Results and Discussion

3.1 Result

The total density of bacteria in the blood, liver and kidneys that have been infected with *E. tarda* bacteria can be seen in Table 1.

Table 1. Density of *E. tarda* Bacteria in Blood, Liver and Kidneys of Silver Rasbora after Immersion with *E. tarda* for 14 Days

Parameter	Bacterial Density (x10^4CFU/mL)			
	P0	P1	P2	P3
Blood	0^{a}±0	0.245^{a}±0.049	0.71^{b}±0.212	0.875^{b}±0.205
Liver	0^{a}±0	0.185^{a}±0.106	0.44^{ab} ±0.113	0.88^{b}±0.113
Kidney	0^{a}±0	0.32^{a}±0.098	0.585^{a}±0.374	0.45^{a} ±0.155

Description: P0: fish were reared without in *E. tarda* bacteria (Control), P1: fish were immersed in *E. tarda* suspension with a density of 10^{11} CFU/mL, P2: fish were immersed in *E. tarda* suspension with a density of 10^{12} CFU/mL, P3: fish were immersed in *E. tarda* suspension with a density of 10^{13} CFU/mL. Different superscripts on the same line showed significant differences ($P<0.05$).

The results of bacterial isolation from blood showed that there was a significant difference ($P<0.05$) on the density of *E. tarda* bacteria in treatment P3 (immersion of *E. tarda* 10^{13} CFU/mL), is 0.875±0.205 x10^4 CFU/mL and P2 (immersion of *E. tarda* 10^{12} CFU/mL), is 0.71±0.212 x10^4 CFU/mL compared to P1 (immersion of *E. tarda* 10^{11} CFU/mL), is 0.245±0.049 x10^4CFU/mL and P0 (negative control).

The highest bacterial density was found in the blood, which was around 0.245±0.049 x10^4 - 0.875±0.205 x10^4 CFU/mL compared to other organs (liver and kidney). The presence of *E. tarda* in the liver was less than the total density of bacteria in the blood, which was around 0.185±0.106 x10^4 - 0.88±0.113 x10^4 CFU/mL in all treatments. The results of bacterial counts from the kidney organs after *E. tarda* infection also showed that there was also no significant difference ($P>0.05$) in the density of *E. tarda* bacteria from all treatments, in the range of 0.32±0.098 x10^4CFU/mL to 0.45±1.55 x10^4CFU/ ml. The total density of bacteria in the kidneys was less than in the blood and liver, which was around 0.32±0.098 x10^4 - 0.45±0.155 x10^4 CFU/mL.

3.2 Discussion

E. tarda can infect several internal fish organs, including liver, kidney, lymph and muscle [17]. The results showed that *E. tarda* infection had occurred in the blood, liver and kidneys as indicated by an increase in the density of bacteria in each organ along with the increase in the concentration of the infected bacteria. Based on the results, the highest bacterial density was found in the blood compared to other organs (liver and kidney). The components of red blood cells contain a lot of iron which is used as a source of nutrients for the growth and proliferation of bacteria [21,22]. [18, 19] stated that *E. tarda* is a septicemic bacterium, so it is commonly found in blood.

Based on the results, the presence of *E. tarda* in the liver was less than the total density of bacteria in the blood in all treatments. These phenomenon caused by the defense system that occurs in the liver, such as macrophages in the form of Kupffer cells [8,10,23]. Resident Kupffer cells play a role in the initial response to injury or damage by releasing pro-inflammatory cytokines and chemokines, such as CCL2 CCL5 [24] (Safithri., 2018), TNF- and IL-6 which function to process cell repair [25]. According to [26], Kupffer cells are a type of hepatic sinusoidal macrophage that has a function to phagocytize pathogens that enter the liver, so that when fish are infected with *E. tarda*, Kupffer cells will phagocytize these pathogens so that cell damage does not occur in the liver.

The results of bacterial counts from the kidney organs after *E. tarda* infection also showed that there was also no significant difference (P>0.05) in the density of *E. tarda* bacteria from all treatments. Based on the results of the study also showed the presence of *E. tarda* infection in the kidneys. However, based on the results of the study, it was shown that the total density of bacteria in the kidneys was less than in the blood and liver. The density of *E. tarda* bacteria also did not cause a very significant difference between treatments (P<0.05). Differences in the number of bacteria in the kidneys can be caused by the body's defense system in the form of macrophages [8,10,23]. The decrease in the number of bacterial density in the kidney is thought to be caused by the defense system of *E. tarda* bacteria unable to fight macrophages in the kidney, so that some bacteria were successfully phagocytized by macrophages [27]. Based on the results of the study also showed that *E. tarda* bacterial infection did not cause external pathological changes such as bleeding and enlargement of the kidneys. According to [28] the density of *E. tarda* bacteria that can cause changes in external clinical symptoms in the kidney is $0.3x10^8$ CFU/mL. So the percentage of bacterial density 0.32 ± 0.098 $x10^4$ - 0.45 ± 0.155 $x10^4$ CFU/mL can already infect fish, but has not caused external clinical symptoms in the kidneys.

Silver rasbora after infection with *E. tarda* bacteria did not show external clinical symptoms, such as the absence of wounds and changes in behavior. It can be assumed that the total bacterial density still does not meet quorum sensing ($0.128\text{-}1.024x10^7$ CFU/mL) [29]. In addition, the absence of clinical symptoms, there was also no mortality or 100% survival in silvers after infection with *E. tarda*, this could be due to the level of pathogenicity of *E. tarda* bacteria determined based on the ability of these bacteria to infect non-specific immunity in fish [30] and the level of bacterial pathogenesis based on the degree of compatibility of the host (susceptable host) [31]. In an unsuitable host, even though the bacteria is pathogenic for certain types of fish, it will not produce the same effect as a suitable host [32]. Host match rates can be receptor based [33]. Based on this statement, it is suspected that bacteria that enter the body are capable of infecting, but have not affected the survival of changes in clinical symptoms and behavior.

4. Conclusion

The conclusion of this study was the highest density of *E. tarda* bacteria infected in silver rasbora was in the blood and the least was in the kidneys.

5. References

[1] Budiharjo A 2003 *BioSMART* 5(1) 55-59.
[2] Budiharjo A 2002. *Biodiversitas Journal of Biological Diversity*. 3(2): 225-230.
[3] Sharma M, Shrivastav A B, Sahni Y P, and Pandey G 2012 *International Research Journal of Pharmacy* 3 (7) 123-127.
[4] Sahoo PK, Swain P, Sahoo SK, Mukherjee SC, Sahu AK. 2000 *Asian Fish Science*. 13 357-362.
[5] Park S B, Takashi A, and Tae S J 2012 *Veterinary Research* 43(67) 1-11.
[6] Katharios P, Kokkari C, Dourala N, Smyrli M 2015 *BMC Veterinary Research*. 11 155.
[7] Wang X, Wang Q, Xiao J, Liu Q, W H 2010 *Fish and Shellfish Immunology*. 29: 1082-1091.
[8] Egusa S 1976 *Fish Pathol* 10 103–114.
[9] Sahoo PK, Mukherjee SC, Sahoo SK 1998 *Journal Aquacult*. 6 57–66.
[10] Miyazaki T, Egusa S 1976 *Fish Patho.l* 11 33–43.
[11] Wiedenmayer AA 2006 *Aquatic Animal Health*. 1-135
[12] Moustafa E M, Amira A E O, and Waleid S O 2016 *World Veteriner Journal.* 6(3) 101-109.
[13] Leung KY, Siame BA, Tenkink BJ, Noort RJ, Mok Y 2012 Review: *Microbes and Infection*. 14: 26-34.
[14] Liu Y, Gao Y, Liu X, Liu Q, Zhang Y 2017 *BMC Microbiology*. 17: 112.
[15] Devi T B, Abraham T J, and Kamiliya D 2016 *Archives of Polish Fisheries*. 24(4) 209-217.
[16] Anthony J W and Maurice K 1993 (New York: Periplus)

[17] Kerie Y, Anwar N and Takele A 2019 *Fish Aqua* 10 (2): 1-6.
[18] Plumb J A, Inglis V, Roberts R J, and Bromage N R 1993 (Oxford: Blackwell) pp 61–79
[19] Yan X H, Li Y, Chen J X, Wang Y G, and Li Q F 2006 *Ocean Univ.China* 36:649–654.
[20] Indonesian National Standard 2663 : 2011. (Jakarta: SNI). pp. 18.
[21] Xie HX, Lu JF, Zhou Y, Yi J, Yu XJ, 2015 *Infection Immunity* 83: 1650-1660.
[22] Chakraborty S, Li M, Chatterjee C, Sivaraman J, Leung KY 2010 *Journal of Biological Chemistry* 285: 38876-38888.
[23] Miyazaki T, Egusa S. 1976 *Fish Pathol*, 11:67–75.
[24] Safithri, F. 2018. *Jurnal Berkala Ilmuah Kedokteran dan Kesehatan*. 2(4) 9-26.
[25] Yoshiya S, Shirabe K, Imai D, Toshima T, Yamashita Y 2015 *Gastroenterol*. 50: 573-582.
[26] Thomson, A.W, Knolle, P.A. 2010 *Nat Rev Immunol*. 10 753–766.
[27] Abbas A dan Lichman A 2005 *DNA ill. Inc.Philadelphia*. 41 391–411.
[28] El-Yazeed, H A and Mai D I 2009.. *Veterinary Medical Journal* 19 (1) 44-50.
[29] Charway G N A, Seulki P, Daeung Y, Jae-Young J, Do-Hung K, Won-Kyo J, Young-Mog K 2018. *Indian Journal Microbiol*:1-5.
[30] Rao, Putanae S, Srinivasa, L, Tit M, Leung K Y. 2001*American Society For Microbiology*. USA.
[31] Kebede B and Habtamu T 2016 *Fish Aqua.* 7(4):1-6.
[32] Narwiyani S, and Kurniasih 2011 *J. Riset Akuakultur*, 6 (2) 291-301.
[33] Zhou Y, Liua Y L, He T T, Laghari Z A, Nie P, Gao Q, Xie H X 2016. *Vet.y Micro*190:12-18.