

QUARTERLY INTERNATIONAL JOURNAL

ECOLOGY

ENVIRONMENT & CONSERVATION

ISSN 0971-795 X



Ecology, Environment and Conservation

PEER-REVIEWED JOURNAL

UGC-CARE JOURNAL

(List II- Globally recognized databases)

Please check by ISSN -0971-785X



Current Issue

Vol 27, Issue 3, 2021

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ISSN: 0971-785X

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Email id: rktivedy@gmail.com

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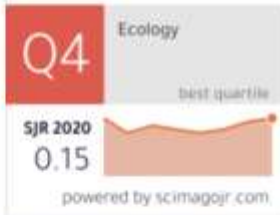
Department of Environmental sciences

University of PUNE

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Ecology, Environment and Conservation



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SCOPUS - H Index - 15

SCOPUS Indexing (coverage)

confirmed for 2021

NAAS Rating - 5.41

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[Contents](#)

[Search Articles](#)

AVIFAUNA COMPOSITION OF TWO NATURAL AND ARTIFICIAL WETLANDS IN JIJEL REGION OF NORTH-EASTERN ALGERIA (THE BENI HAROUN DAM AND REDJLA MARSH)

Chabou Sarra, Khammar Hichem, Hadjab Ramzi and Saheb Menouar

[Get Abstract](#)

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POSITIONING OF ENVIRONMENTAL EDUCATION IN LIFE SCIENCES (GRADE 12)

Sikhulile Bonginkosi Msezane

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RESOURCE-SAVING RESTORATION TECHNOLOGIES OF THE DEGRADED IRRIGATED LANDS IN SOUTHEASTERN KAZAKHSTAN

Tastanbek Atakulov, Sagynbay Kaldybaev, Kenzhe Erzhanova and Ashirali Smanov

[Get Abstract](#)

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IMPORTANCE OF ALDER FORESTS FOR BIRDS IN THE NORTH-EAST OF ALGERIA: COMPOSITION AND STRUCTURE OF BREEDING BIRDS STANDS AND THE EFFECT OF

ECOLOGY, ENVIRONMENT AND CONSERVATION

VOL. 26 (November Suppl. Issue) : 2020

CONTENTS

- S1–S5 Preliminary study of dengue virus serotype on *Aedes* mosquitoes in endemic area, Surabaya, Indonesia, January 2020
—Aulia Azzahra, Lucky Vera Oktavia, Muhammad Fariz Naviyanto, Shifa Fauziyah, Teguh Hari Sucipto, Dwi Winarni, Sri Puji Astutik Wahyuningsih, Siti Churrotin, Ilham Harlan Amarullah and Soegeng Soegianto
- S6–S11 Remote sensing and GIS based assessment of groundwater potential zones in AMU campus using AHP approach
—S. Said and M. Anees
- S12–S17 Population-level of *Nannochloropsis* sp. as an enrichment diet for marine rotifer *Brachionus rotundiformis* in mass culture tanks
—Putu Angga Wiradana, Mayadita Dwi Sani, Raden Joko Kuncoroningrat Susilo, Arif Nur Muhammad Ansori, Ni Nyoman Sri Septiani, Deny Suhernawan Yusup and Agoes Soegianto
- S18–S22 Experimental study on behaviour of fiber reinforced concrete and fly ash for rigid Pavements
—Jayant Virat and Humaib Nasir
- S23–S28 Availability of ecological resources in power plant Tanjung Tiram Village, South Konawe, Indonesia
—Ferasari Ferasari, La Sara, La Rianda and La Onu La Ola
- S29–S35 Faunal diversity of Kitchen Gardens of Sikkim
—Aranya Jha, Sangeeta Jha and Ajeya Jha
- S36–S40 *Oreochromis mossambicus* accumulates lead without showing growth inhibition
—Sumah Yulaipi, Aunurohim, Arif Luqman, Dewi Hidayati and Agoes Soegianto
- S41–S48 Heavy metal concentration of Chandigarh urban soils due to urbanization in a changing environment: An ecological assessment
—Viney Kumar, Rupinder Kaur and A. N. Singh
- S49–S54 Effect of addition of onion (*Allium cepa* L.) extract in ringer's diluent on spermatozoa quality of *Gallus domesticus* at room temperature
—Sakinato Mazidda, Suyadi and Dyah Hikmawati
- S55–S60 Fish diversity of River Bhagirathi Upstream to Tehri Dam Reservoir, Uttarakhand (India)
—M.S. Rawat, Dhyaal Singh and O.P. Gusain
- S61–S64 Size structure and gonad maturity of red snapper *Lutjanus malabaricus* in Pinrang waters, Makassar Strait, South Sulawesi, Indonesia
—Nuraeni L. Rapi, Mesalina Tri Hidayani, Murwantoko, Djumanto and Agoes Soegianto
- S65–S69 A review analysis on environmental factors influencing morphology and behaviour of estuarine Mollusc
—Arundhati Ganguly, Banani Mandal, Arunava Mukherjee and Susanta Kumar Chakraborty
- S70–S78 The influence of ozone exposure on organoleptic and chlorophyll levels of curly lettuce (*Lactuca sativa* L.)
—Suryani Dyah Astuti, Hery Purnobasuki, Miratul Khasanah, Siti Khoiriyatul, Nurul Fitriyah, Deny Arifianto and Fadli Ama
- S79–S83 Growth and nutrient uptake of indian mustard [*Brassica juncea* (L.) Czern and Coss.] genotypes as influenced by nitrogen and sulphur fertilization under irrigated condition
—Harsita Nayak, J. S. Bohra and Shiv Poojan Yadav

- S84–S90 Biofouling colonization on cubic artificial reefs in Pantai Damas, Trenggalek, Indonesia
—*Andik Isdianto, Oktiyas Muzaky Luthfi, Shafa Thasya Thaeraniza and Agoes Soegianto*
- S91–S97 A study on web asymmetry and prey capture in *Argiope pulchella* Thorell, 1881 (Araneae: Araneidae)
—*Sangeeta Das, Jatin Kalita and Nilutpal Mahanta*
- S98–S103 The effectiveness of solenoid magnetic fields to reduce precipitation levels of CaCO₃ in hard water
—*Fadli Ama, Suryani D. Astuti, Tri A. Prijo, Qod’nu Rahmawati, Yunus Susilo and Rahma A. Puspitasari*
- S104–S108 Nutrition-based benefits of Kitchen Gardens: An investigation of gender differences
—*Ananya Jha, Sangeeta Jha, Shenga Sherap, Rajlakshmi Mallik and Ajeya Jha*
- S109–S113 Potency of phosphate solubilizing mold from rhizosphere soil in Mangrove Center Tuban, Indonesia
—*Tini Surtiningsih, Arina Putri Ramadhani, Dinda Rahmi Anindi, Ni’matuzahroh, Tri Nurhariyati and Fatimah*
- S114–S122 Investigating local community’s perception on tourism development in protected areas: A study on Sunderbans Tiger Reserve, India
—*Ananya Ghosh, Pankaj Kumar Tyagi and Pawan Gupta*
- S123–S126 Phytochemical in the methanol extract of *Piper sarmentosum*
—*Junairiah, Tri Nurhariyati, and Nabilah Istighfari Zuraidassanaaz*
- S127–S134 Analysis of water quality status in Bordoibam Bilmukh wetland ecosystem of Assam, India
—*Jayanta Sonowal, Kaustubh Rakshit and Debojit Baruah*
- S135–S139 Utilization of bagasse and sawdust as bio-based insulation on the walls of the ship’s accommodation ceiling
—*Tristiandinda Permata, D. Hikmawati, Aurista Miftahatul Ilmah and Jailani*
- S140–S144 Prediction of temperature data for Ghataprabha Sub-basin using change factor method
—*Bharath A., Preethi S., Manjunatha M., Ranjitha B. Tangadagi and Shankara*
- S145–S155 Mapping of land potentially for maize plant in Madura Island-Indonesia using remote sensing data and geographic information systems (GIS)
—*Suhartono, Agoes Soegianto and Achmad Amzeri*
- S156–S161 Biototoxicity analysis of different doses of *Beauveria bassiana* (Balsamo) Vuillemin against Nymph of *Odontotermes obesus* (R.)
—*Anjana Intodia, Arti Prasad and Bharati Veerwal*
- S162–S165 The effect of cooking methods to the existence of *Bacillus* sp. spores in beef
—*Adityas Putri Pamartha, Mochammad Lazuardi, Nenny Harijani, Agnes Theresia Soelih Estoepangestie, Didik Handijatno, Martia Rani Tacharina and Dadik Raharjo*
- S166–S169 Effectiveness of planned teaching programme about ‘E-waste management’ among Jr. College going students
—*Rutuja M. Ghorpade, Nandkumar R. Kakade, Tukaram B. Zagade, Anagha V. Katti and Sneha S. Mahindrakar*
- S170–S173 Isolation and identification of fungal infections causing death in leopard gecko’s (*Eublepharis macularius*) eggs
—*Erwin Nugroho Indhi, Koesnoto Supranianondo, Sri Chusniati, Djoko Legowo, Suryanie Sarudji, Martia Rani Tacharina and Didik Handijatno*
- S174–S181 Assessment of elemental Carbon, Nitrogen, Hydrogen and Sulphur in alluvial sediments of River Yamuna in Delhi region
—*Vivek Chopra and Jai Gopal Sharma*

- S182–S187 Antifungal potency againsts *Candida albicans* (ATCC 10231) and its activity as biosurfactant of WNA 4.1.13 fermented growth of sediment from mangrove Wonorejo Surabaya Indonesia
—C. Rahayuningsih, S. Chusniati, D. Handijatno, L. Maslachah, S. Sarudji and Rahmi Sugihartuti
- S188–S196 A review on impact of coal mining on soil properties and reclamation by organic amendments
—Poonam Poonia, Ram Prasad Choudhary and Sangita Parihar
- S197–S201 Characterization of *Aeromonas hydrophila* bacteria on dumbo catfish (*Clarias gariepinus*) from Bungo Jambi Province, Indonesia
—A. Indrawati, T. Wulandari, F. H. Pasaribu and A. B. Rifai
- S202–S209 *Moringa oleifera* : A potent immune booster in the catastrophe of Covid -19
—Madhumita Bhattacharjee
- S210–S214 Identification of worms in the digestive tract of water monitor lizards through gastrointestinal surgery
—A. N. Faradis, Mufasirin, S. Mulyati, Kusnoto, I. S. Yudaniayanti and E. Suprihati
- S215–S220 GCMS analysis of Phyto Components of the musky smelling *Dendrobium moschatum*
—Dipika Rajput and L.R. Saikia
- S221–S224 Correlation between muara grouper fish weight (*Epinephelus coioides*) with *Anisakis* worm infection level in Mayangan Indonesia
—T. D. Setyaningrum, S. Koesdarto, T. R. Yustinasari, Kusnoto, M. Yunus and E. B. Aksono
- S225–S230 Effect of nitrogen and zinc levels on growth and yield of Basmati rice
—Nirmal Joshi, Shiv Prakash Singh, Tikendra Kumar Yadav and Uppu Sai Sravan
- S231–S237 Isolation of *Actinomycetes* from mangrove sediments at Ujung Pangkah, Gresik, Indonesia
—A. R. Hidayatullah, R. Sugihartuti, D. Handijatno, S. Chusniati, L. Maslachah and S. Sarudji
- S238–S244 Development, environmental impact and green growth: India
—Dheeraj Verma, Vartika Singh, Prodyut Bhattacharya and Jagdish Kishwan
- S245–S247 Description of breeding management Timor deer (*Cervus timorensis*) in Merauke, Papua Province, Indonesia
—K. R. Ismail, Ismudiono, I. N. Triana, P. Srianto, M. Hariadi and S. Utama
- S248–S251 Microgreens: Exciting new food for 21st Century
—Shashank Sharma, Priyanka Dhingra and Sameer Koranne
- S252–S254 Acanthocephala worm detection in cavity body of frog (*Fejervarva cancrivora*) in Surabaya, Indonesia
—S. L. Rahmatika, S. Koesdarto, E. P. Hestianah, E. D. Poetranto, L. T. Suwanti and Kusnoto
- S255–S260 Assessment of spatio- temporal changes in current Jhum cultivation of *Thysanolaena maxima* in Mawthai village of Umsning Tehsil in Meghalaya
—Raymond Wahlang and S. S. Chaturvedi
- S261–S264 Antibiotic resistance profile of *Escherichia coli* isolates collected from cloaca swabs on laying hens in Udanawu Sub-District, Blitar District, Indonesia
—Freshinta Jellia Wibisono, Bambang Sumiarto, Tri Untari, Mustofa Helmi Effendi, Dian Ayu Permatasari and Adriana Mutamsari Witaningrum
- S265–S266 Determination of oil and grease present in the Hussain Sagarlake, Hyderabad, Telangana, India
—Anitha and S. Kedarini
- S267–S270 Isolation and identification of *Lactobacillus* sp. bacteria in asian palm civet (*Paradoxurus hermaphroditus*) feces
—Dinda Jelita Jauharah, Sri Chusniati, Mohammad Anam Al Arif, Wiwiek Tyasningsih, Suryanie Sarudji, Agnes Theresia and Soelih Estoepangestie

- S271–S275 **Assessment of water quality index for Shivrath river in Durg, Chhattisgarh State, India**
—*Sukhpreet Kaur Bhatia and Sumita Nair*
- S276–S280 **The effectiveness of antibacterial essential oil of cinnamon (*Cinnamomum burmannii*) on *Staphylococcus aureus***
—*M. L. Hakim, S. Susilowati, M. H. Effendi, W. Tyasningsih, R. Sugihartuti, S. Chusniati and A. M. Witaningrum*
- S281–S285 **The impact of consumer's engagement in Pro-environment activities on the preference for green food products**
—*Deepika Jindoliya and Gagandeep Nagra*
- S286–S290 **Cone maturation timing and seed germination in *Pinus roxburghii* (Serg.) in the central Himalayan region of Uttarakhand, India**
—*Amit Mittal, Nandan Singh, Ashish Tewari and Shruti Shah*
- S291–S294 **Total plate count of beef meat at traditional markets in south of Surabaya, Indonesia**
—*Z. Aminullah, W. P. Lokapirnasari, N. Harijani*, M. H. Effendi, Budiarto and W. Tyasningsih*
- S295–S299 **Behaviour of concrete Brick and flyash Brick on infilled frame under cyclic loading**
—*K. Senthil, S. Rupali, Ajay Pratap, A. Thakur and A. P. Singh*
- S300–S306 **Sero-prevalence and hematological investigation of *Bovine brucellosis* under extreme ecological conditions**
—*Aamir Shehzad, Awais Masud, Tabassam Fatima, S. Bibi and Fedik Abdul Rantam*
- S307–S313 **Life forms classification and biological spectrum in natural and human impacted ecosystems of Senapati district, Manipur, India**
—*Ng Niirou and Asha Gupta*
- S314–S320 **Distribution of gastrointestinal parasite in beef cattle through feces examination at Gunung Tabur Sub-District, Berau Regency, Indonesia**
—*Rosyida Dwi Rahmawati, Nunuk Dyah Retno Lastuti, Mustofa Helmi Effendi, Setiawan Koesdarto, Soeharsono and Muhammad Yunus*
- S321–S326 **Decolorisation of Textile Dyes using Immobilised PPO from Tomato Peel and Pulp**
—*Sr. Sandra Horta, Agnel Arul John and S. Parijatham Kanchana*
- S327–S332 **The biosurfactant activity of supernatant fermentation broth isolates bacterial origin of Surabaya's Wonorejo mangrove sediment and its potential as an antifungal against *candida albicans* ATCC 10231**
—*Bima Widya Pramudianto, Suryanie Sarudji, Rahmi Sugihartuti, Didik Handijanto, Wiewiek Tyasningsih and Eduardus Bimo Aksono*
- S333–S336 **Zooplankton diversity in Amaravathi Dam Tirupur District, Tamilnadu, India**
—*A. Krishnamoorthi and K. Moorthikumar*
- S337–S342 **Implementation of fotogrametry techniques as body mass estimation of Indo-pacific bottle nose dolphin (*Tursiops aduncus*) in Bali dolphin lodge**
—*Muhammad Adifian Latif, Amar Ma'ruf, Erma Safitri, Yeni Dhamayanti, Soeharsono and Boedi Setiawan*
- S343–S344 **Iron removal of water by using different parts of *Musa paradisiaca***
—*K. S. Beenakumari*
- S345–S350 **Trace element contamination in fruits and vegetables grown in low nutrient availability soil environment by using inductively coupled plasma mass spectrometry**
—*N. Swathi, P. Padmavathi and N.V.S. Venugopal*
- S351–S356 **Biopigments and Rubisco expression under Heavy metal stress in *Spirulina platensis***
—*Ameesh Dev Singh and Gajendra Pal Singh*

- S357–S359 Systematic survey on population of *Gyps himalayensis* in Hirpora Wildlife Sanctuary, Jammu and Kashmir, India
—Hameem Mushtaq Wani, Mustahson F. Fazili, Samina A. Charoo and Riyaz Ahmad
- S360–S368 Experimental study of biomedical waste incinerator using input-output method: A case study of biomedical waste incinerator at Etmadpur, Agra, India
—Sandeep Kumar Verma, N.B. Singh, C.N. Tripathi and P.K. Sharma
- S369–S377 Quantifying and mapping sediment retention ecosystem services in a mountain landscape of Southern Western Ghats, India
—Shiju Chacko, C. Ravichandran, Jikku Kurian and S.M. Vairave
- S378–S381 Wastewater characterization of grossly polluted textile industries located at main stem of River Ganga in Uttar Pradesh, India
—Ajit Kumar Vidyarthi, Pankaj Kumar, Surindra Negi and Vipin Kumar
- S382–S386 Performance analysis of existing sewage treatment plants in Prayagraj, Uttar Pradesh
—Ajit Kumar Vidyarthi and Raj Kishore Singh
- S387–S392 Floristic cortege of the genre *Lavatera* a Malvaceae for the two species: *Lavatera maritima* and *L. flava* in the region of sabra (Tlemcen, Western of Algeria)
—Ghalem Sarra, Hassani Faiçal, Bensouna Amel, Khatir Hadj and Aouadj Sid Ahmed
- S393–S396 Surface water quality and pollution load in river Kali-east: A tributary of river Ganga, India
—Ajit Kumar Vidyarthi, Vivek Rana, Garima Dublsh, Prabhat Ranjan and Mrinal Kanti Biswas
- S397–S407 Assessment of water quality of Choyyia Nadi (River) Catchment area in Bijnor District, Uttar Pradesh, India
—Matta Gagan, Rajput Ayush, Rajput Akshay, Kumar, Pawan, Kumar, Avinash, Nayak, Anjali, Kumar Ajendra, Dhingra, Gulshan K., Chauhan Avnish, Chadha, Sanjeev Kumar and Wats, Meenu
- S408–S414 Flowering pattern and floral architecture of Wild and cultivated varieties of Jamun (*Syzygium cumini* L.) for pollination and productivity
—Eswarappa, G. and Somashekar, R.K.
- S415–S422 The rate of absorption of carbon dioxide and moisture content in Linggua (*Pterocarpus Indicus* Willd.) for climate change management
—Gun Mardiatmoko, Jacob Kailola, Radios Simanjuntak and Agustinus Kastanya
- S423–S427 Ecological significance of plant life forms of an urban green space of Purulia Region, West Bengal, India
—Rimi Roy, Manideepa Bhattacharya, Barsha Baral and Deblina Das Modak
- S428–S433 A study of the risk of ground water pollution by shallow septic tank system in Aligarh, India
—Sohail Ayub, Md. Meraj Faisal and Pushpendra Kumar Sharma
- S434–S438 Preliminary analysis of fungal macroflora in Madras Christian College vegetation and ecological aspects
—Mirfath Jahan, Jeya Rathi J., Kumar M. and Santhosh S.
- S439–S443 Biodecolorization of reactive red HE7B and reactive orange 3R through Indigenous bacterial isolate *Microbacterium oryzae* strain JC8 isolated from textile effluent
—Ravi Kant Rahi and Varsha Gupta

Isolation of *Actinomycetes* from mangrove sediments at Ujung Pangkah, Gresik, Indonesia

A. R. Hidayatullah, R. Sugihartuti*, D. Handijatno, S. Chusniati, L. Maslachah and S. Sarudji

Faculty of Veterinary Medicine, Universitas Airlangga, Surabaya, Indonesia

(Received 22 March, 2020; Accepted 16 July, 2020)

ABSTRACT

This research was conducted to get actinomycetes isolate from mangrove sediments at Ujung Pangkah in Gresik-Indonesia. Sediment samples were taken from mangrove rhizosphere in Ujung Pangkah on 3 different location sampling areas. Ten grams of soil sample was accurately weighed and transferred to add 100 mL of sterile NaCl 0.9% mixed well for 10 minutes. The resultant solution was serially diluted up to 10^{-12} (10^{-3} , 10^{-4} until 10^{-12}) with NaCl 0.9%. One millilitre of each intermediate dilution (10^{-3} , 10^{-4} until 10^{-12}) was added to 10 mL of sterile molten Starch Casein Agar medium which has been supplemented using both chloramphenicol and griseofulvin each one 0.05 ppm individually in separate flasks. The plates were incubated for the growth of actinomycetes colonies at 28°C and observed intermittently during incubation. After 7 days of incubation, the colonies showing the characteristics of actinomycetes (rough, chalky, powdery appearance radiating growth and leathery texture) were observed. Identification of actinomycetes isolates was conducted by observing macroscopic characteristic of the colony, microscopic of conidial, bacterial cell and the ability of bacteria to resist from acid alcohol. Identification was done based on Bergey's manual of determinative if bacteriology. The result of this research obtained 9 different isolates actinomycetes. The 9 isolate was identified as 3 genera from actinomycetes. The genera of isolate obtained are *Micromonospora*, *Nocardia* and *Streptomyces*.

Key words: *Actinomycetes isolate, Incubation, Bacteriology*

Introduction

Mangroves are saltwater forest ecosystems which is found in tropical and sub-tropical regions throughout the world (Huang *et al.*, 2008). Mangrove ecosystems are characterized by their surface soil being flooded by water where salinity and surface fluctuations are affected by tides. Mangrove soils have a unique character for the growth of various kinds of microorganisms that play an important role in degrading the soil. Several factors that affect mangrove ecosystems cause microorganisms to adapt by producing unique metabolites, namely primary and secondary metabolites (Thatoi *et al.*, 2013)

Some groups of microorganisms that can be

found in mangrove ecosystems include fungi, bacteria, algae and protozoa. Microbial groups in tropical mangrove forests consist of 91 % bacteria and fungi, 7 % algae and 2 % protozoa (Palla *et al.*, 2018). According to Naikpatil and Rathod (Naikpatil and Rathod, 2011), actinomycetes make up 10 – 15 % of microbial communities in the soil. In the mangrove ecosystem to date only 5 % of microbes have been chemically isolated and examined and belong to groups of bacteria, fungi and actinomycetes (Xu, 2015).

The order actinomycetes or actinomycetales are members of the Actinobacteria groups. Actinomycetes are a group of Gram-positive bacteria and have high G and C content (~ 55 %) in their DNA

(Naikpatil and Rathod, 2011), This organism has a structure that resembles a fungus. Only after DNA testing, this group is classified as bacteria.

This bacterium has many important roles in various industries because of its ability to produce a number of diverse metabolite compounds. These metabolite compounds have benefits such as, antibiotics, antifungal, antiviral, anticancer, enzymes, immunosuppressants and other compounds that are beneficial in industry (Xu *et al.*, 2014). On this basis actinomycetes are widely used as medicinal ingredients in tackling diseases. In addition, actinomycetes also have an important role in soil mineralization, nutrient immobilization, antibiosis and plant growth promoter production (Sonia *et al.*, 2011) which later can also be applied to agriculture.

Aquatic actinomycetes have been shown to have an important role in the discovery of several new bioactive compounds such as research conducted by Huang *et al.* (Huang *et al.*, 2008) on rifamycin from *Micromonospora*, Fehling *et al.* (Fehling *et al.*, 2003) found salinosporamide-A as an anticancer metabolite of the *Salinispora* strain, Marinomisin from *Marinophilus sp.* and much more. According to Anzai *et al.* (2008) out of 22,500 biologically active compounds, 45 % are derived from actinomycetes.

Most actinomycetes are organisms that live freely and are widely distributed in nature both in terrestrial and aquatic ecosystems. Among various ecosystems, very little research has been carried out to obtain actinomycetes isolates from mangrove ecosystems, one of which is the Ujung Pangkah Mangrove ecosystem in Gresik. Ujung Pangkah Mangrove Ecosystem is one of the river mouths of Bengawan Solo. Based on fields observations conducted by Rudianto (Rudianto, 2014) in this ecosystem there are many industrial waste contaminants that are channeled into watersheds that produce chemical wastes such a hydrocarbons and heavy metals.

This research was conducted to isolate and identify actinomycetes from Ujung Pangkah Mangrove ecosystem, Gresik Regency, Indonesia. Actinomycetes in this ecosystem are thought to have the potential to produce primary and secondary metabolites. Actinomycetes will produce primary and secondary metabolites in extreme conditions such as the Ujung Pangkah mangrove ecosystem, which is polluted by various types of waste. The results of isolation from this study can be used for further research such as screening for primary and secondary

metabolite that produce actinomycetes.

Materials and Methods

Research Samples

The materials used in this study were mangrove sediments, Starch-Casein Agar, NaCl 0.9%, violet crystals, safranin, acetone alcohol, lugol, aquades, 70 % alcohol, and emersion oil.

Sterilization of Tools and Materials

Sterilization of tools made of glass and materials using an autoclave with a 121°C temperature and 2 atm of pressure for 15 minutes. Oze sterilized using bunsen combustion fire, while the equipment that cannot stand the heat is sterilized using 70 % alcohol (Cappuccino and Sherman, 2013)

Sampling

Soil samples taken at several points refer to research conducted by Fatiqin (Fatiqin, 2011) which divides into three sampling areas which are far from the sea, approaching the sea, and dealing with the sea. Sampling was carried out at three locations (red dots) in the Ujung Pangkah mangrove ecosystem in accordance with the three point image. Each location was taken two samples from adjacent points each of 5 g. Samples are taken using pipes with a 5 – 20 cm depth from the surface where microbial activity can generally be found (Priyadarshini *et al.*, 2016). Samples were taken around the mangrove's roots because the microbes concentration around the roots



Fig. 1. Viewing of sampling locations in the Ujung Pangkah mangrove ecosystem. Note: 1. Location facing the sea, 2. Location close to the sea, 3. Location far from the sea.

is generally greater than in far areas from the roots (Arifiyanto, 2018). The samples obtained are stored in a sample glass and covered with aluminum foil and then put in a ice box to be brought to the laboratory.

Characterization of Soil Samples

Soil sample identification aims to get the soil samples characteristics. This identification includes pH of the soil, soil color, and soil temperature. The identification is carried out at the sampling location.

Total of Plate Numbers Calculation

Samples were taken as much as ten g and suspended using physiological NaCl into the Erlenmeyer flask until reaching 100 mL volume and shaken for 10 minutes. Samples are diluted in stages starting from 10^{-3} to 10^{-12} dilution. Samples that have been carried out dilution taken as much as 1 mL and dropped on a Petri dish then SCA media that has been added griseofulvin and chloramphenicol 0.05 ppm respectively poured on a cup. Then, incubate the petri dish at 28 °C for 7 days (Palla *et al.*, 2018). Colonies are calculated to determine ALT with formula:

Number of bacteria = Number of colonies x 1 / diluent factor

The media provisions that are used as the ALT basic calculation are petri containing 30-300 CFU / ml colonies.

Actinomycetes Isolation

The isolation of actinomycetes begins by taking the colony from culture on the calculation of the total plate number by taking it using sterile ose and applying it to the surface of the media using the quadrant swipe method. The taken colony is a separate colony and does not overlap with other colonies and choose different colors and shapes. Then the petri dish are incubated at 28 °C for 7 days. This treatment can be repeated to get a similar colony (Palla *et al.*, 2018).

Purification and Storage of Actinomycetes

Colonies that have been purely identified macroscopically are suspected actinomycetes bacteria deposited into oblique SCA media. The media is incubated at 28 °C for 7-14 days and growth was observed (Palla *et al.*, 2018). Media that have been incubated and there is growth of actinomycetes bacteria are stored in refrigerator.

Identification

The actinomycetes colony was observed morphologically and microscopically by referring to Bergey's manual of determinative bacteriology.

Macroscopic colony

Characterization of mummies culture that had been incubated for 7-14 days at 28 °C was observed morphology of actinomycetes colony based on shape, color, texture, growth time and mycelium colony as well as the ability of actinomycetes to give a characteristic soil odor. Colonie that have the same color and shape are grouped.

Microscopic colony

Observations that based on using light microscopy by Shirling and Gottlieb (1966) method, isolates were grouped based on the form of air mycelium which can be observed by Gram staining. In Gram staining can be observed types of Grams, the shape of bacteria and the presence or absence of spores. Acid-resistant staining is also done to find out some groups of actinomycetes that are resistant to acid resistant staining.

Results and Discussion

Characterization of Soil Samples Results

Mangrove soil samples were taken at three different sampling locations, which are far from the sea, approaching the sea, and dealing directly with the sea. The sample is taken at 10.00 AM until 12.00 PM. Each sample location has a pH level that not much different, which is obtained ± 6.8 average. The average temperature obtained is ± 28.5 °C. Soil samples obtained are quite clay at locations far from the sea. Samples at this location are dominated by mud. Contrast with the two samples taken from locations close to the sea and those dealing directly with the sea which has a slightly sandy consistency.

Soil samples taken from three points of the sampling location are mixed so, the average soil pH is ± 6.9 . Generally actinomycetes are intolerant of acids and the amount decreases at pH 5.0. In theory, actinomycetes are suitable to grow in the pH range of 6.5–8.0 (Subba Rao, 1994). Based on Kumar *et al.* (2014) in the mangrove ecosystem, the number of actinomycetes colonies counted the most in samples with an alkaline pH (± 7.5).

The temperature of the three sampling locations

obtained a range between 27 °C-29 °C. The optimal temperature for actinomycetes growth is between 25°C-30°C (Subba Rao., 1994). However, Barka et al. (Barka *et al.*, 2016) has slightly different results. The optimal temperature range is a temperature between 25 ° C-37 ° C. Temperature is known to have an influence on metabolism and bacterial growth. When the temperature increases, the rate of chemical reactions in the bacteria also increases. At a certain temperature point, the growth rate does not increase when the temperature rises. This is caused by protein denaturation when peptide bonds start to break from the tertiary and quaternary structures of proteins (Wheelis, 2011). Ujung Pangkah mangrove ecosystem has an optimal temperature range for actinomycetes growth, so that actinomycetes can grow well in this ecosystem.

Calculation of Total Plate Numbers

Soil samples obtained from three locations were mixed and the calculation of the total plate count on the Starch-Casein Agar medium. Bacterial colonies were counted using a colony counter. This calculation is carried out in the Bacteriology laboratory of the Pharmaceutical Research Services in Microbiology Laboratory, Faculty of Veterinary Medicine Universitas Airlangga. The results of the calculation of bacterial colonies obtained data as follows :

$10^{-3} = \sim$; $10^{-4} = \sim$; $10^{-5} = 520$; $10^{-6} = 118$; $10^{-7} = 22$; $10^{-8} = 8$; $10^{-9} = 7$; $10^{-10} = 1$; $10^{-11} = 3$; $10^{-12} \sim = 5$.

The results can be calculated the number of bacteria per milliliter in the sample as follows :

$$\begin{aligned} \text{Number of bacteria} &= \text{number of colonies} \times 1 / \text{diluent factor} \\ &= 118 \times 1/10^{-6} \\ &= 118000000 \\ &= 1,2 \times 10^8 \end{aligned}$$

In this soil sample, the number of bacteria per milliliter of sample was 120,000,000 bacteria isolated using SCA media. This study found 13 isolates sus-

pected of actinomycetes from 43 isolates and identified only 9 different isolates. Data obtained from the calculation of total plate numbers could not shown a picture of the actual number of actinomycetes because they are still mixed with other bacteria. According to Palla *et al.* (2018) actinomycetes that live in mangrove ecosystems are indeed not as many as in terrestrial areas. The abundance of actinomycetes is influenced by the moisture level of the soil sample. Soils filled with water are not suitable for growth of actinomycetes, whereas soils in arid and semi-arid regions can maintain large populations due to spore resistance to drought (Subba Rao, 1994).

Isolation and Identification of Actinomycetes Bacteria

Actinomycetes bacteria were isolated on SCA media. The isolated bacteria were taken from the calculation of the total plate count. Intake of the colony is carried out in separate colonies. Bacterial isolation is repeated until a pure colony is obtained. The isolation was obtained as many as 43 isolates and 13 isolates were thought to be an actinomycetes group and consisted of 9 different isolates. Following are the macroscopic observations of colonies which are thought to be a group of actinomycetes bacteria :

Microscopic identification is also established by observing cell shapes and Gram types. Gram staining can also be used to determine differences in the structure of actinomycetes and fungi. Actinomycetes are Gram-positive which have branched hyphae that often develop into mycelium and have a rod shape (Chakraborty, 2015). The results of these observations indicate that 13 isolates that were thought to belong to the actinomycetes group were Gram-positive and rod-shaped. Pratiwi (Chakraborty, 2015) explains, the structure of bacterial cells, the cell wall of Gram-positive bacteria contains a thick layer of peptidoglycan so that it can

Table 1. The macroscopic observations of the colonies suspected actinomycetes group

Group	Mycelium Aerial Color	Mycelium Substrate Color	Number of Isolate	Isolate Name
1	Bluish white	Yellowish white	2	C21B, C34A
2	Pale white	Pale white	2	C25A, C25B
3	Bright white	Yellowish white	2	C31C, C101B
4	Pale white	Brown	2	C36BB, C48AC
5	Grey	Grey	1	C42A
6	Pale white	White	2	C44A, C102C
7	White	Red	1	C101C
8	Reddish white	White	1	C51B

form a rigid structure, and there is theatric acid which contains alcohol and phosphate. So when there is a purple-iodine crystal complex that enters a Gram-positive bacterial cell it cannot be washed away by alcohol because of the presence of a strong peptidoglycan layer on the cell wall. In Gram staining, spores can also be observed. Gram staining actually only colors vegetative cells from bacteria. The presence of these spores can be marked by the presence of parts that are not stained with Gram staining. However, Gram staining cannot be used to detect free sepora as in dead bacteria. As the result that bacteria has died, there are no vegetative cells that can be colored by Gram staining. Acid-resistant staining in this study was used to determine several genera from the actinomycetes group such as nocardia and mycobacterium. In acid-resistant staining could be known types of bacteria that have thick wax walls which resistant to discoloration using acid alcohol. Bacteria that are resistant to acid alcohol discoloration will be painted red, while those that are not acid resistant are painted blue.

Grouping of *Actinomycetes* Isolates

The grouping of actinomycetes was done by observing color and shape of the colony. This method was carried out to facilitate identification by classifying isolates based on the color of aerial mycelium and mycelium substrate. The morphology of actinomycetes growing on the media can be used to identify the characteristics of actinomycetes, but this information cannot be placed up to the specific genus (Basavaraj, 2010). The results of this grouping obtained 9 different isolates. Microscopic observations

were made to observe the similarity of the cell structure of each group. This microscopic observation can also be used to determine the genus of bacteria. Group A was thought to be a group of actinomycetaceae family. In this family is characterized by the presence of bacterial cells that experience fragmentation into small rod shapes. After acid resistant staining, it is suspected that group one is a group of the nocardia genus. The nocardia genus belongs to the actinomycetaceae group which is resistant to acid-resistant staining as described in Bergey's Manual of Determinative Bacteriology. Morphological formation of group one also has similarities with the genus nocardia. Microscopic observations show the formation of spores in group one, so that it can be used to differentiate with the genus mycobacterium which are both acid resistant but do not produce spores and have slightly different structures.

Group B has members of groups 2, 3, 5, 6 and 8. This group is thought to be a group of the streptomycetaceae family and belongs to the genus *Streptomyces*. *Streptomyces* has a characteristic that is the location of the conidia within the mycelium forming a chain that distinguishes it from other genera in a family. Mycelium *streptomyces* has a characteristic that is not fragmented so that long spore chains are formed. These *streptomyces* species can be identified based on the color of the colony in certain media according to Bergey's Manual of Determinative Bacteriology. The colony's color identification must also be supported by observing conidial shapes.

Group C consists of two groups, 4 and 7. This

Table 2. Microscopic description analysis.

Number	Isolate	Gram	Spora	Acid resistant	Form of mycelium
1	C21B	+	+	+	Partially segmented and partially separated
2	C25A	+	+	-	Segmented
3	C25B	+	+	-	Segmented
4	C31C	+	+	-	Segmented
5	C34A	+	+	+	Partially segmented and partially separated
6	C36BB	+	+	-	Not segmented
7	C42A	+	+	-	Segmented
8	C44A	+	+	-	Segmented
9	C48AC	+	+	-	Not segmented
10	C51B	+	+	-	Segmented
11	C101B	+	+	-	Segmented
12	C101C	+	+	-	Not segmented
13	C102C	+	+	-	Segmented

group is thought to be a genus of micromonosporae. The micromonospora genus has the characteristic that there is no septa in the mycelium. Spores of the genus micromonospora are located in conidia which attach to simple conidiophores on the surface of the mycelium. This genus has similar structure with genus thermoactinomyces in one family. The difference between the two genera is that the genus thermoactinomyces can only grow at temperatures of 50 °C-65 °C. In this study the media was incubated at 28 °C, so it can be ascertained that the growing colonies are a group of the genus micromonospora.

Conclusion

Based on the isolation of actinomycetes from mangrove sediments at the Ujung Pangkah Gresik-Indonesia, 13 isolates were obtained consisting of three genera. The three isolates consisted of genera Streptomyces, nocardia and micromonospora. The genus Streptomyces and micromonospora belong to the same family, streptomycetaceae, while the genus nocardia belongs to the family actinomycetaceae. Most of the isolates observed were identified as genus Streptomyces with 7 isolates. The genus nocardia consists of 2 isolates and 4 micromonosporae.

Acknowledgement

The existence of this research requires the support or cooperation of ecosystems from Ujung Pangkah Mangrove, Gresik Regency, Indonesia, so that this study provides the expected results and is useful for further research.

References

- Arifiyanto, A. 2018. *Isolation of Actinomycetes from Rhizosfer in Sidoarjo Mud Area and the Activity of Biosurfactant Antimicrobial Production*. Departemen Biologi. Fakultas Sains dan Teknologi. UNIVERSITAS AIRLANGGA. [Text in Indonesian].
- Anzai, K., Nakashima, T., Kuwahara, N., Suzuki, R., Ohfuku, Y., Takeshita, S. and Ando, K. 2008. Actinomycete bacteria isolated from the sediments at coastal and offshore area of Nagasaki Prefecture, Japan: diversity and biological activity. *Journal of Bioscience and Bioengineering*. 106(2) : 215-217.
- Basavaraj, K.N., Chandrashekhara, S., Shamarez, A.M., Goudanavar, P.S. and Manvi, F.V., 2010. Isolation and morphological characterization of antibiotic producing actinomycetes. *Tropical Journal of Pharmaceutical Research*. 9(3).
- Barka, E.A., Vatsa, P., Sanchez, L., Gaveau-Vaillant, N., Jacquard, C., Klenk, H.P., Clément, C., Ouhdouch, Y. and van Wezel, G.P. 2016. Taxonomy, physiology, and natural products of Actinobacteria. *Microbiol. Mol. Biol. Rev.* 80(1) : 1-43.
- Cappuccino, J. and Sherman, N. 2013. *Microbiology: A Laboratory Manual* (7th ed.) Pearson Education, San Francisco, CA. pp.145-146, 151 and 271.
- Chakraborty, S., Ghosh, M., Chakraborti, S., Jana, S., Sen, K.K., Kokare, C. and Zhang, L. 2015. Biosurfactant produced from Actinomycetes nocardiosis A17: characterization and its biological evaluation. *International Journal of Biological Macromolecules*. 79 : 405-412.
- Feling, R.H., Buchanan, G.O., Mincer, T.J., Kauffman, C.A., Jensen, P.R. and Fenical, W. 2003. Salinosporamide A: a highly cytotoxic proteasome inhibitor from a novel microbial source, a marine bacterium of the new genus Salinospora. *Angewandte Chemie International Edition*. 42(3) : 355-357.
- Fatqin, A., 2011. *Actinomycetes Potential Test as an Antibiotic Products Isolated from Rhizosfer Soil in Wonorejo Mangrove, Surabaya*. (Undergraduate Thesis, UNIVERSITAS AIRLANGGA). [Text in Indonesian].
- Huang, H., Lv, J., Hu, Y., Fang, Z., Zhang, K. and Bao, S. 2008. *Micromonospora rifamycinica* sp. nov., a novel actinomycete from mangrove sediment. *International Journal of Systematic and Evolutionary Microbiology*. 58(1) : 17-20.
- Kumar, P.S., Duraipandiyar, V. and Ignacimuthu, S. 2014. Isolation, screening and partial purification of antimicrobial antibiotics from soil *Streptomyces* sp. SCA 7. *The Kaohsiung Journal of Medical Sciences*. 30(9) : 435-446.
- Li, Q., Chen, X., Jiang, Y. and Jiang, C. 2016. Morphological identification of actinobacteria. *Actinobacteria-Basics and Biotechnological Applications*. Rijeka, Croatia: InTech, pp. 59-86.
- Nurkanto, A. and Julistiono, H. 2014. Screening and study of antifungal activity of leaf litter actinomycetes isolated from Ternate Island, Indonesia. *Asian Pacific Journal of Tropical Medicine*. 7 : S238-S243.
- Naikpatil, S.V. and Rathod, J.L. 2011. Selective isolation and antimicrobial activity of rare actinomycetes from mangrove sediment of Karwar. *Journal of Ecobiotechnology*. 3 (10) : 48-53.
- Palla, M.S., Guntuku, G.S., Muthyala, M.K.K., Pingali, S. and Sahu, P.K. 2018. Isolation and molecular characterization of antifungal metabolite producing actinomycete from mangrove soil. *Beni-Suef University Journal of Basic and Applied Sciences*. 7(2) : 250-256.
- Priyadarshini, A., Singdevsachan, S.K., Tripathy, S.K., Mohanta, Y.K., Patra, J.K. and Sethi, B.K. 2016. Isolation and identification of Actinomycetes from

- Mangrove soil and extraction of secondary metabolites for antibacterial activity. *British Biotechnology Journal*. 12 (2) : 1.
- Rudianto, R. 2014. Co-management based on coastal area ecosystem restoration analysis a case study in Ujung Pangkah Subdistrict, Bungah District, Gresik, East Java. *Research Journal of Life Science*. 1(1): 54-67. [Text in Indonesian].
- Sonia, M.T., Naceur, J. and Abdennaceur, H. 2011. Studies on the ecology of actinomycetes in an agricultural soil amended with organic residues: I. identification of the dominant groups of Actinomycetales. *World Journal of Microbiology and Biotechnology*. 27(10) : 2239-2249.
- Subba Rao, N.S. 1994. Soil Microorganisms and Plants Growth. *Universitas Indonesia Press, Jakarta*. 352. [Text in Indonesian].
- Thatoi, H., Behera, B.C., Mishra, R.R. and Dutta, S.K. 2013. Biodiversity and biotechnological potential of microorganisms from mangrove ecosystems: a review. *Annals of Microbiology*. 63(1): 1-19.
- Wheelis, M. 2011. *Principles of Modern Microbiology*. Jones & Bartlett Publishers. Sudbury, MA. pp. 203-205.
- Xu, J. 2015. Bioactive natural products derived from mangrove-associated microbes. *RSC Advances*. 5(2) : 841-892.
- Xu, D.B., Ye, W.W., Han, Y., Deng, Z.X. and Hong, K. 2014. Natural products from mangrove actinomycetes. *Marine Drugs*. 12 (5) : 2590-2613.
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