Bonorowo Wetlands

| Bonorowo Wetlands | vol. 7 | no. 2 | December 2017| ISSN 2088-110X | E-ISSN 2088-2475|

Emetha audouini♀ photo by Ahmet Öktener

Bonorowo Wetlands

| Bonorowo Wetlands | vol. 7 | no. 2 | December 2017| ISSN 2088-110X | E-ISSN 2088-2475 |

Published semiannually **PRINTED IN INDONESIA**

ISSN 2088-110X
E-ISSN 2088-2475

9 772088 110148

Bonorowo Wetlands

| Bonorowo Wetlands | vol. 7 | no. 2 | December 2017 |

ONLINE
http://smujo.id/bw

p-ISSN: 2088-110X, **e-ISSN:** 2088-2475

PUBLISHER
Society for Indonesian Biodiversity

CO-PUBLISHER
Institut Pertanian Bogor, Bogor, Indonesia
Nusantara Institut of Biodiversity, Universitas Sebelas Maret, Surakarta, Indonesia

OFFICE ADDRESS
1. Forest and Land Fire Laboratory, Department of Silviculture, Faculty of Forestry, Institut Pertanian Bogor. Jl. Lingkar Akademik Kampus IPB Dramaga, Bogor 16680, West Java, Indonesia. Tel.: +62 251 8626806, Fax.: +62 251 8626886;
2. Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Sebelas Maret. Jl. Ir. Sutami 36A Surakarta 57126, Central Java, Indonesia. Tel./Fax.: +62-271-663375, email: bw@smujo.id, jbonorowo@gmail.com

PERIOD OF ISSUANCE
June, December

Society for Indonesian Biodiversity

Universitas Sebelas Maret Surakarta, Indonesia

Institut Pertanian Bogor, Bogor, Indonesia

Published by Smujo International for The Society for Indonesian Biodiversity and Universitas Sebelas Maret & Institut Pertanian Bogor, Indonesia

GUIDANCE FOR AUTHORS

Aims and Scope *Bonorowo Wetlands* encourages submission of manuscripts dealing with all topics relevant to freshwater, brackish and marine coastal ecosystems, include zoology, botany, ecology, geology, sedimentology, hydrology, biogeochemistry; waste water disposal; as well as management, laws and regulations, policy and economics.

Article types The journal seeks original full-length research papers, reviews, and short communication. Manuscript of original research should be written in no more than 8,000 words (including tables and picture), or proportional with articles in this publication number. Review articles will be accommodated, while, short communication should be written at least 2,000 words, except for pre-study.

Submission The journal only accepts online submission through system or email to the editors at **jbonorowo@gmail.com** (until the end of 2017). Submitted manuscripts should be the original works of the author(s). The manuscript must be accompanied by a cover letter containing the article title, the first name and last name of all the authors, a paragraph describing the claimed novelty of the findings versus current knowledge. Submission of a manuscript implies that the submitted work has not been published before (except as part of a thesis or report, or abstract); and is not being considered for publication elsewhere. When a manuscript written by a group, all authors should read and approve the final version of the submitted manuscript and its revision; and agree the submission of manuscripts for this journal. All authors should have made substantial contributions to the concept and design of the research, acquisition of the data and its analysis; drafting of the manuscript and correcting of the revision. All authors must be responsible for the quality, accuracy, and ethics of the work.

Acceptance The only articles written in English (U.S. English) are accepted for publication. Manuscripts will be reviewed by editors and invited reviewers (double blind review) according to their disciplines. Authors will generally be notified of acceptance, rejection, or need for revision within 1 to 2 months of receipt. The manuscript is rejected if the content does not in line with the journal scope, does not meet the standard quality, inappropriate format, complicated grammar, dishonesty (i.e. plagiarism, duplicate publications, fabrication of data, citations manipulation, etc.), or ignoring correspondence in three months. The primary criteria for publication are scientific quality and biodiversity significance. **Uncorrected proofs** will be sent to the corresponding author as *.doc* or *.rtf* files for checking and correcting of typographical errors. To avoid delay in publication, corrected proofs should be returned in 7 days. The accepted papers will be published online in a chronological order at any time, but printed in June and December.

Ethics Author(s) must obedient to the law and/or ethics in treating the object of research and pay attention to the legality of material sources and intellectual property rights.

Copyright If and when the manuscript is accepted for publication, the author(s) still hold the copyright and retain publishing rights without restrictions. Authors or others are allowed to multiply article as long as not for commercial purposes. For the new invention, authors are suggested to manage its patent before published.

Open access The journal is committed to free-open access that does not charge readers or their institutions for access. Readers are entitled to read, download, copy, distribute, print, search, or link to the full texts of articles, as long as not for commercial purposes. The license type is CC-BY-NC-SA.

A charge The journal is committed to free of charge for submission and publication of non-institutional funded research (waiver).

Reprints The sample journal reprint is only available by special request. Additional copies may be purchased when ordering by sending back the uncorrected proofs by email.

Manuscript preparation Manuscript is typed on A4 (210x297 mm^2) paper size, **in a single column**, single space, 10-point (10 pt) Times New Roman font. The margin text is 3 cm from the top, 2 cm from the bottom, and 1.8 cm from the left and right. Smaller lettering size can be applied in presenting table and figure (9 pt). Word processing program or additional software can be used, however, it must be PC compatible and Microsoft Word based (*.doc* or *.rtf*; **not *.docx***). **Scientific names** of species (incl. subspecies, variety, etc.) should be written in italic, except for italic sentence. Scientific name (genera, species, author), and cultivar or strain should be mentioned completely for the first time mentioning it in the body text, especially for taxonomic manuscripts. Name of genera can be shortened after first mentioning, except generating confusion. Name of the author can be eliminated after first mentioning. For example, *Rhizopus oryzae* L. UICC 524, hereinafter can be written as *R. oryzae* UICC 524. Using trivial name should be avoided, otherwise generating confusion. **Biochemical and chemical nomenclature** should follow the order of the IUPAC - IUB. For DNA sequence, it is better used Courier New font. Symbols of standard chemical and abbreviation of chemistry name can be applied for common and clear used, for example, completely written butilic hydroxyl toluene (BHT) to be BHT herein after. **Metric measurement** use IS denomination, usage other system should follow the value of equivalent with the denomination of IS first mentioning. Abbreviations set of, like g, mg, mL, etc. do not follow by dot. Minus index (m^{-2}, L^{-1}, h^{-1}) suggested to be used, except in things like "per-plant" or "per-plot". **Equation of mathematics** does not always can be written down in one column with text, in that case can be written separately. **Number** one to ten are expressed with words, except if it relates to measurement, while values above them written in number, except in early sentence. The fraction should be expressed in decimal. In the text, it should be used "%" rather than "percent". Avoid expressing ideas with complicated sentence and verbiage, and used efficient and effective sentence.

Title of the article should be written in compact, clear, and informative sentence, preferably not more than 20 words. Name of author(s) should be completely written. **Name and institution** address should also be completely written with street name and number (location), postal code, telephone number (O), facsimile number (O), and personal email address. For Indonesian universities, use local name. Manuscript written by a group, author for correspondence along with address is required. First page of the manuscript is used for writing above information.

Abstract should not be more than 200 words. **Keywords** is about five words, covering scientific and local name (if any), research theme, and special methods which used; and sorted from A to Z. All important **abbreviations** must be defined at their first mention. **Running title** is about five words. **Introduction** is about 400-600 words, covering the background and aims of the research. **Materials and Methods** should emphasize on the procedures and data analysis. **Results and Discussion** should be written as a series of connecting sentences, however, for manuscript with long discussion should be divided into subtitles. Thorough discussion represents the causal effect mainly explains for why and how the results of the research were taken place, and do not only re-express the mentioned results in the form of sentences. **Concluding** sentence should be given at the end of the discussion. **Acknowledgments** are expressed in a brief; all sources of institutional, private and corporate financial support for the work must be fully acknowledged, and any potential conflicts of interest are noted.

Figures and Tables of maximum of three pages should be clearly presented. Title of a picture is written down below the picture, while title of a table is written above the table. Colored figures can only be accepted if the information in the manuscript can lose without those images; chart is preferred to use black and white images. Author could consign any picture or photo for the front cover, although it does not print in the manuscript. All images property of others should be mentioned source. **There is no appendix**, all data or data analysis are incorporated into Results and Discussions. For broad data, it can be displayed on the website as a supplement.

References Author-year citations are required. In the text give the authors name followed by the year of publication and arrange from oldest to newest and from A to Z. In citing an article written by two authors, both of them should be mentioned, however, for three and more authors only the first author is mentioned followed by et al., for example: Saharjo and Nurhayati (2006) or (Boonkerd 2003a, b, c; Sugiyarto 2004; El-Bana and Nijs 2005; Balagadde et al. 2008; Webb et al. 2008). Extent citation as shown with word "*cit*" should be avoided. Reference to unpublished data and personal communication should not appear in the list but should be cited in the text only (e.g., Rifai MA 2007, pers. com. (personal communication); Setyawan AD 2007, unpublished data). In the reference list, the references should be listed in an alphabetical order (better, if only 20 for research papers). Names of journals should be abbreviated. Always use the standard abbreviation of a journal's name according to the **ISSN List of Title Word Abbreviations** (www.issn.org/2-22661-LTWA-online.php). The following examples are for guidance.

Journal:

Saharjo BH, Nurhayati AD. 2006. Domination and composition structure change at hemic peat natural regeneration following burning; a case study in Pelalawan, Riau Province. Biodiversitas 7: 154-158.

Book:

Rai MK, Carpinella C. 2006. Naturally Occurring Bioactive Compounds. Elsevier, Amsterdam.

Chapter in book:

Webb CO, Cannon CH, Davies SJ. 2008. Ecological organization, biogeography, and the phylogenetic structure of rainforest tree communities. In: Carson W, Schnitzer S (eds) Tropical Forest Community Ecology. Wiley-Blackwell, New York.

Abstract:

Assaeed AM. 2007. Seed production and dispersal of *Rhazya stricta*. 50th Annual Symposium of the International Association for Vegetation Science, Swansea, UK, 23-27 July 2007.

Proceeding:

Alikodra HS. 2000. Biodiversity for development of local autonomous government. In: Setyawan AD, Sutarno (eds.) Toward Mount Lawu National Park; Proceeding of National Seminary and Workshop on Biodiversity Conservation to Protect and Save Germplasm in Java Island. Universitas Sebelas Maret, Surakarta, 17-20 July 2000. [Indonesian]

Thesis, Dissertation:

Sugiyarto. 2004. Soil Macro-invertebrates Diversity and Inter-Cropping Plants Productivity in Agroforestry System based on Sengon. [Dissertation]. Universitas Brawijaya, Malang. [Indonesian]

Information from internet:

Balagadde FK, Song H, Ozaki J, Collins CH, Barnet M, Arnold FH, Quake SR, You L. 2008. A synthetic *Escherichia coli* predator-prey ecosystem. Mol Syst Biol 4: 187. www.molecularsystemsbiology.com. DOI:10.1038/msb.2008.24

THIS PAGE INTENTIONALLY LEFT BLANK

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 55-64

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070201

The morphological characters of *Mothocya taurica* (Czerniavsky, 1868) and *Emetha audouini* (H. Milne Edwards, 1840) from Turkey

AHMET ÖKTENER[1,♥], ALI ALAŞ[2], DILEK TÜRKER[3]

[1]Deparment of Fisheries, Sheep Research Station. Çanakkele Street 7km.,10200, Bandırma, Balıkesir, Turkey. Tel.: +90-266-7380080, ♥email: ahmetoktener@yahoo.com

[2] Department of Biology, Faculty of Education, Necmettin Erbakan University. B Block, 42090, Meram, Konya, Turkey

[3]Department of Biology, Faculty of Science, Balikesir University. Cagıs Campus, 10300, Balikesir, Turkey

Manuscript received: 30 May 2017. Revision accepted: 27 June 2017.

Abstract. *Öktener A, Alaş A, Türker D. 2017. The morphological characters of* Mothocya taurica *(Czerniavsky, 1868) and* Emetha audouini *(H. Milne Edwards, 1840) from Turkey. Bonorowo Wetlands 7: 55-64.* This paper aims to present morphological characters of two species, *Mothocya taurica* (Czerniavsky, 1868) and *Emetha audouini* (H. Milne Edwards, 1840) from Turkey. Although *Mothocya taurica* with different synonymies were described by several researchers, Bruce (1986) indicated the necessity of redescription of *Mothocya taurica*. Pleopods 1 to 5 having medial peduncle margin with 4 hooks of *Emetha audouini* is found for the first time in this study as distinct from other studies. Also, the host preferences of these parasites are given.

Keywords: Cymothoidae, *Emetha*, Isopoda, morphology, *Mothocya*, Turkey

INTRODUCTION

Cymothoids are ectoparasitic isopods on the body, fins, or inside the buccal or the branchial cavities of numerous freshwater and marine fishes. They are protandrous hermaphrodite (Bariche and Trilles 2005). Although the Cymothoidae family is famously known, there are some deficiencies from the taxonomic point of view. According to some researchers, studies concerned with molecular and morphological are needed on this family (Poore and Bruce 2012; Martin et al. 2013; Hadfield et al. 2016).

Thirty-one species in the *Mothocya* genus and two in the *Emetha* genus were listed by The World Register of Marine Species (Hadfield et al., 2017; Schotte 2007). Three species (*Mothocya epimerica*, *Mothocya taurica*, *Mothocya belonae*) of *Mothocya* and one species (*Emetha audouini*) of *Emetha* are reported from Turkish waters, but these studies include limited information about the morphology of mouth-parts (Öktener and Trilles 2004; Kırkım 1998). *Mothocya taurica* was reported from the Black Sea, the Mediterranean Sea (Trilles 1994; Bruce 1986; Kononenko 1988; Ramdane et al. 2006). Bruce (1986) pointed out a needed evaluation of the status of this species.

The morphological characters are given in the study obtain a possibility to compare the other countries' findings next time. Although it could not use electron microscopy, the DNA barcoding and molecular identification methods are reliable and expensive in this study. It aims to present the morphological characters, especially including mouthparts of *Mothocya taurica* and *Emetha audouini* from Turkey.

MATERIALS AND METHODS

Eighty-three *Alosa* sp. (Pisces; Clupeidae) and 38 picarel *Spicara maena* (Linnaeus, 1758) (Pisces; Centracanthidae), 56 *Sardinella aurita*Valenciennes, 1847 (Clupeidae), 170 *Engraulis encrasicolus* (Linnaeus, 1758) (Engraulidae) were collected with the local fishing gears in the Sea of Marmara in 2014. The collected parasites were fixed in 70% ethanol. Mouthparts and pleopods were dissected using a Wild M5 stereo microscope. The dissected parts were mounted on slides in a glycerin-gelatine mounting medium. The pleopods of isopods were stained with methylene blue. The appendages were drawn with the aid of a camera lucida (Olympus BH-DA). The photos were taken with Canon EOS 1100D camera attached to the microscope. The measurements were taken in millimeters (mm) with a micrometric program (Pro-way). The scientific names synonyms of parasite and host were checked with the WoRMS Editorial Board (2017). According to Froese and Pauly (2017), the host's feeding habits and habitat characteristics were prepared. *Mothocya taurica* (MNHN-IU-2013-18751) and *Emetha audouini* (MNHN-IU-2017-16) was deposited in the collections of the Musée National d'Histoire Naturelle (MNHN), Paris, France.

RESULTS AND DISCUSSION

***Mothocya taurica* (Czerniavsky, 1868) (Figure 1-6)**
Synonyms:
Cymothoa oestrum Rathke, 1837: 394
Cymothoa punctata Uljanin, 1872: 113-114.—Popov, 1933: 193,196-198.—Markewitsch, 1934: 224,225, pl.XLV (fig.10-11).—Nikolaeva, 1963: 1-46

Lironeca pontica Borcea, 1933a: 128.— Borcea, 1933b: 481-502, figs 1-9, pls 2-4.

Livoneca punctata Vasiliu and Carausu, 1948: 180-184, pl.II (fig.1-12),pl.III (fig.13-38d).—Carausu, 1959: 349-351,pl.I (fig.A-B)

Lironeca punctata Trilles, 1976a: 782-783, pl.I. fig.6.—Dollfus and Trilles, 1976: 828

Lironeca taurica Kussakin, 1979: 295, figs. 160, 161.

Livoneca taurica Uljanin, 1871: 113.—Uljanin, 1872: 113

Host: *Alosa* sp. (the shad); **Locality:** Bandırma Bay; **Total parasite:** 5; **Dissected parasite:** 3.

All parasites were firmly attached to the gill cavity of the host. The prevalence means the intensity of parasites were 6%, 1 respectively.

Description-female: Body length varies from 15 to 19 mm. Body slightly twisted to one side, about 2 times as long as wide. Pereon widest at pereonite 4, most narrow at pereonite 1. Coxal plates are visible in dorsal view, with round posterior margins. Length of coxae greater than width. Pereonite 1 longest, length of pereonites decreasing step by step from 3 to 7; pereonite 7 shortest. The width of the head is about 2 times the head length. Pleotelson wider than length, posterior margin evenly rounded, about 1.6 wider than long. All pleonites visible, the first pleonite distinctly narrow, 2-5. pleonites slightly wider. Pleon 1 largely and pleon 2 partially concealed by pereonite 7. Antennula and antenna are composed of eight articles. Mandible palp third article distinctly shorter than others. Maxillula with four terminal spines, one long and three short spines. Maxilla medial lobe with 2 spines, lateral lobe with 2 spines. Maxilliped article 3 with five hooked spines. Pereopods 5-7 are slightly larger than pereopod 1-4, all without spines. Pleopods 1 to 5 has a medial peduncle margin with 4 hooks. Pleopods 3 to 5 endopods with large proximomedial lobes. Uropod rami not extending beyond pleotelson; endopod slightly longer than exopod.

Distribution: Black Sea, Mediterranean Sea, Aegean Sea (Bruce 1986; Kononenko 1988; Trilles 1994; Ramdane et al. 2006; Schotte 2008a; Ramdane et al. 2009;).

Hosts: *Alosa immaculata* (Borcea 1933a; Borcea 1933b; Vasiliu and Carausu 1948; Muradian 1972; Kussakin 1979; Trilles 1976; Bruce 1986; Trilles 1994; Öktener and Trilles 2004; Olguner 2008; Öktener et al. 2010); *Alosa fallax* (Dollfus and Trilles 1976); *Atherina hepsetus* and *Gobius* sp (Markewitsch 1934); *Alosa tanaica*, *Pomatomus saltatrix* (Borcea 1933a); *Engraulis encrasicolus*, *Sprattus sprattus*, *Trachurus mediterraneus* (Nikolaeva 1963); *Helicolenus dactylopterus*, *Trisopterus minutus* (Öktener et al. 2009); *Sardina pilchardus* (Borcea 1933a; Markewitsch 1934; Vasiliu and Carausu 1948); *Scorpaena porcus* (Markewitsch 1934; Josipa et al. 2007).

The host's parasitism with *Mothocya taurica* has been examined according to family characteristics 37% of 13 hosts belong to Clupeidae, and 63% to Carangidae, Gadidae, Scorpaenidae, Pomatomidae, Sebastidae, Gobiidae, Engraulidae, and Atherinidae. The host's parasitism with *Mothocya taurica* was examined according to habitat selections; 54% of 13 host fish species are pelagic-neritic; 15% pelagic-oceanic; 15% demersal; 8% benthopelagic, and 8% bathydemersal. According to feeding habits, the host's parasitism with Mothocya taurica; all hosts are carnivorous.

Clupeidae fishes are hosts of *Mothocya taurica*. This parasite selects carnivorous and pelagic fishes as hosts for habitat and feeding habits. This study examined *Alosa* sp. is a carnivorous and pelagic fish. It is fit as preferring host for *Mothocya taurica*. Although it should indicate references to this statement that *Mothocya taurica* is also reported from *Engraulis encrasicolus* and *Sardinella aurita*, it was not found in these fishes.

Remarks: The antennula and the antenna with 8 articles found in this study agree with the descriptions of Borcea (1933b); Vasiliu and Carausu (1948); Kussakin (1979); Bruce (1986); Öktener et al. (2010, re-examined). The maxillula with four terminal spines found in this study is compatible with Kussakin (1979), Vasiliu and Carausu (1948), and Öktener et al. (2010). The medial lobe and lateral lobe with 2 spines of maxilla found in this study are compatible with the findings indicated by Kussakin (1979) and Öktener et al. (2010), while the medial lobe with 1 spine and the lateral lobe with 2 spines mentioned by Vasiliu and Carausu (1948).

The third article without setae on the lateral margin of the mandible palp found in this study is compatible with the descriptions of Kussakin (1979) and Öktener et al. (2010), excepting for Vasiliu and Carausu (1948) findings. Five spines on article 3 of the maxilliped of ovigerous female were observed in this study, while 4 spines on an article of ovigerous female maxilliped were described by Kussakin (1979); 4 spines on an article of female maxilliped by Vasiliu and Carausu (1948), and 5 spines on an article of ovigerous female maxilliped by Öktener et al. (2010).

There are limited studies about the morphology of *Mothocya taurica* (Borcea 1933b; Vasiliu and Carausu 1948; Kussakin 1979; Bruce 1986; Öktener et al. 2010). The findings of the structures of mouthparts, pereopod, and pleopod in this study agree with the previous literature. In appearance, *Mothocya taurica* is very similar to *M. belonae*. It can be distinguished by having slightly wider coxae, a rounder pleotelson, and pleopods 3 to 5 endopods with much larger proximomedial lobes as well as a distolateral extension, according to Bruce (1986).

***Emetha audouini* (H. Milne Edwards, 1840) (Figure 9-14)**

Synonyms:

Cymothoa audouini Milne-Edwards, 1840: 274-275.—Heller, 1866: 738-739.—Stalio, 1877: 237.—Stossich, 1880: 45.—Gerstaecker, 1901: 255-257

Cymothoa nigropunctata Hope, 1851: 33

Cymothoa audouinii: Hope, 1851: 33

Emetha audouinii Schioedte and Meinert, 1883: 317-321, tab.XI, fig.14-18.—Dudich, 1931: 18.—Montalenti, 1941: 337-394.—Montalenti, 1948: 27-36,tav.I (fig.1-8).—Amar, 1951: 530.—Euzet and Trilles, 1961: 190-191.—Trilles, 1962: 103-106.—Trilles, 1964: 107-108.—Trilles, 1968: 20-36, plI-IV,phot.2-5.—Thampy and John, 1974: 580, 582.—Quignard and Zaouali, 1980: 357.—Brusca, 1981: 127.—Sartor, 1986: 1-12.—Sartor, 1987: 49.—Wagele, 1987: 1-398.

Figure 1. *Mothocya taurica*♀

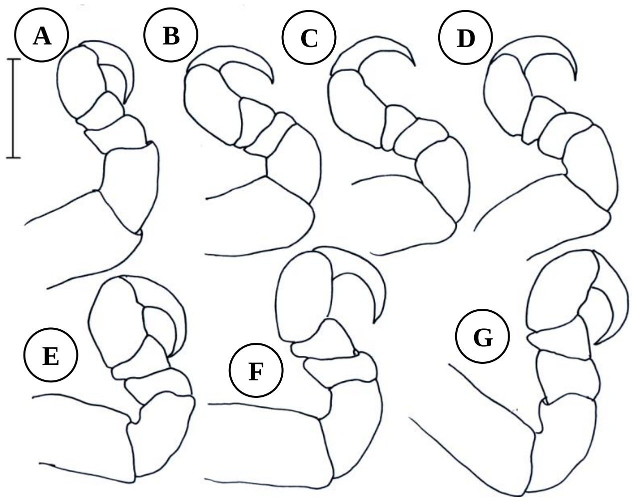


Figure 4. *Mothocya taurica* ♀, A-G. pereopods I-VII (1.2 mm)

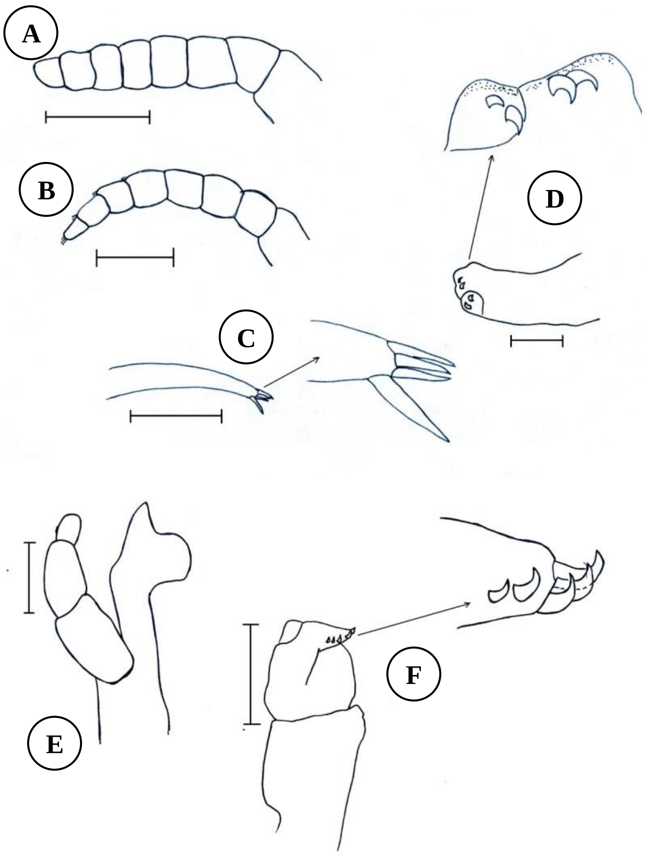


Figure 3. *Mothocya taurica* ♀, A. antennula (0.25 mm), B. antenna (0.27 mm), C. maxillula (0.40 mm), D. maxilla (0.26 mm), E. mandible (0.35 mm), F. maxilliped (0.47 mm)

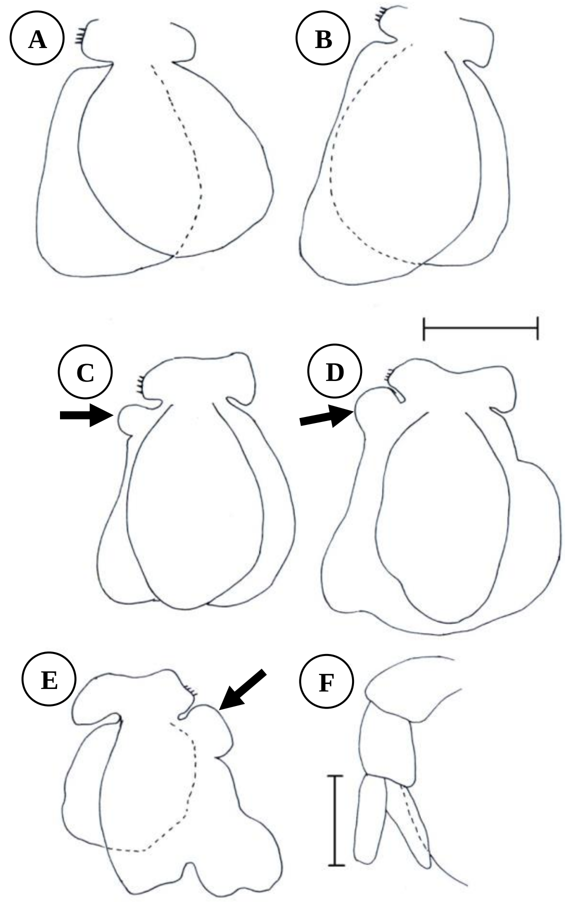


Figure 5. *Mothocya taurica* ♀, A-E. pleopods I-V (1.43 mm), F. uropod (1.33 mm).

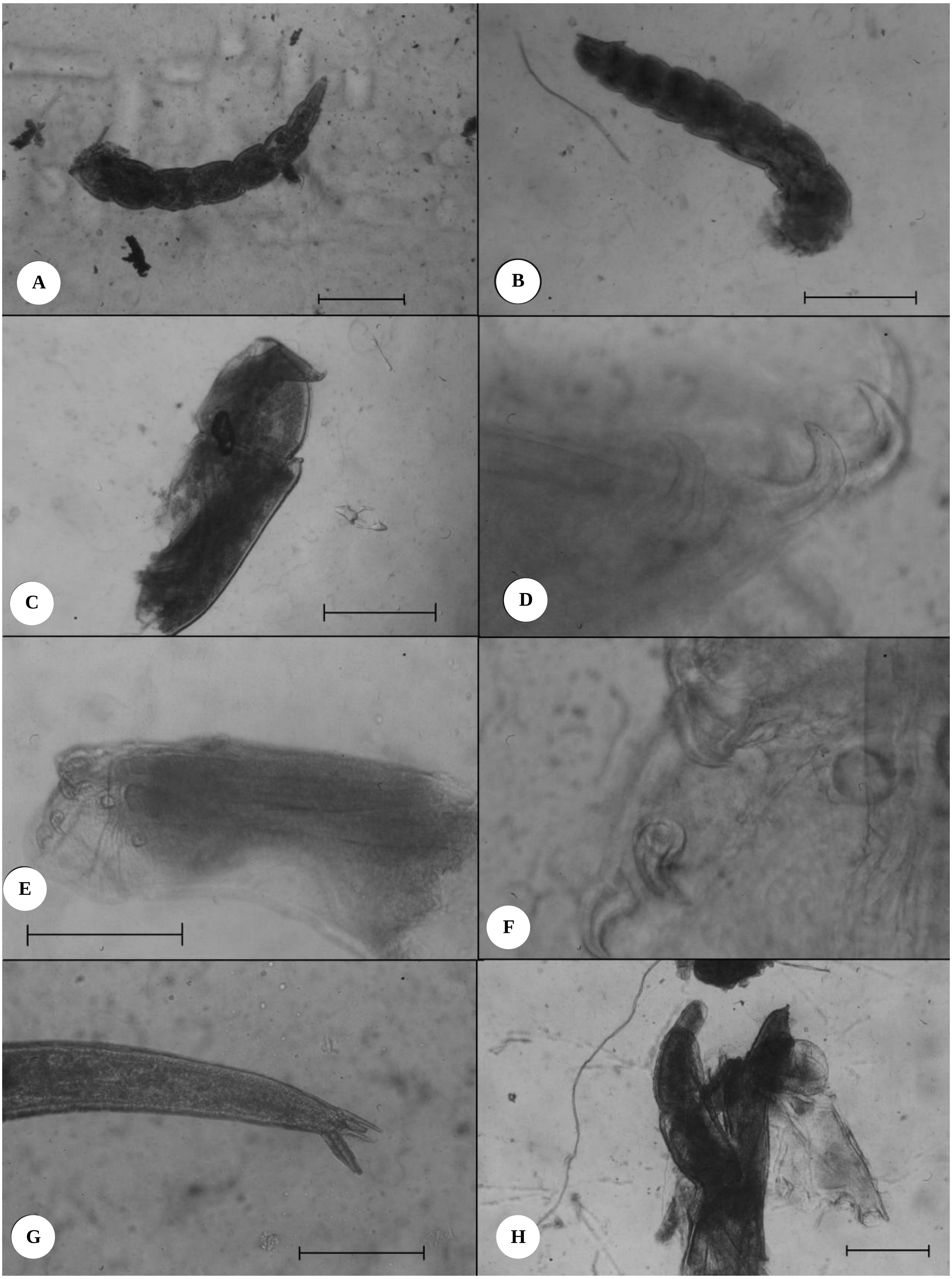


Figure 2. *Mothocya taurica* ♀, A. antenna (0.27 mm), B. antennula (0.25 mm), C. maxilliped (0.47 mm), D. spines on maxilliped (2 mm), E. maxilla (0.26 mm), F. spines on maxilla, G. maxillula (0.10 mm), H. mandible (0.14 mm)

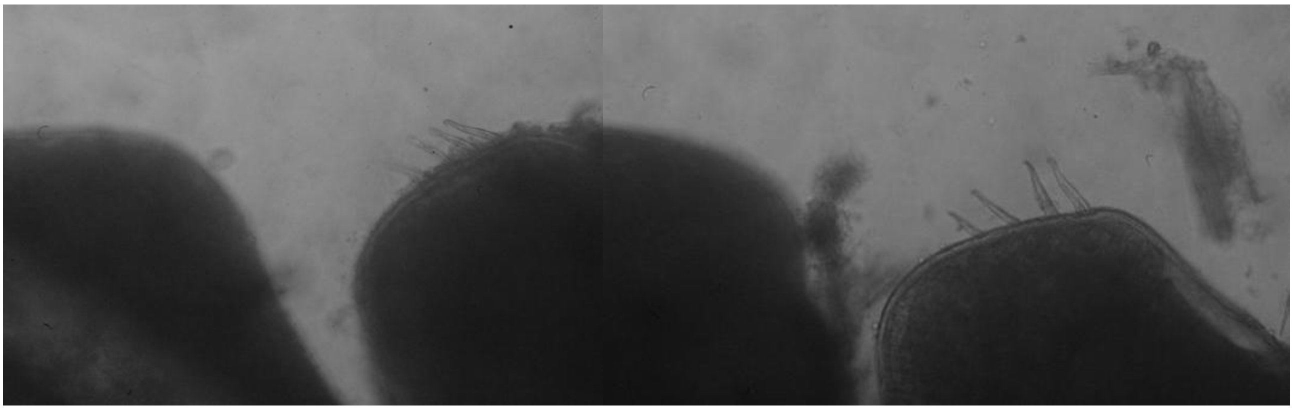


Figure 6. Four hooks on medial peduncle margin of pleopods (*Mothocya taurica*♀)

Figure 7. *Mothocya taurica* on the right and left gill chamber of fish

Figure 8. Atrophy on the gill filaments of fish parasitized by *Mothocya taurica*

Emetha audouini Carus, 1885: 442.—Quintard-Dorques, 1966: 10.—Berner, 1969: 93-95.—Trilles, 1972a: 1192-1196, fig.1-45, pl (1-3).—Trilles, 1972b: 1232-1233.—Trilles, 1972c: 1269-1277, fig.1-13.—Trilles, 1977: 8.—Romestand and Trilles, 1977: 92-95, fig.1-2.—Radujkovic, 1982: 155-161.—Radujkovic et al, 1984: 161-181.—Trilles et al. 1989: 279-306, fig.7

Emetha adriatica Bovallius, 1885: 17-20, pl.IV (fig. 34-40)

Ceratothoa salparum Gourret, 1891: 18-19, tav.I (fig.19), tav. XI (figs.7-13)

Host: *Spicara maena* (Linnaeus, 1758) (Blotched picarel); **Locality:** Bandırma Bay; **Total parasite:** 8; **Dissected parasite:** 6.

All parasites were firmly attached to the mouth cavity of the host. The prevalence means the intensity of parasites was 21%, 1 respectively.

Description-female: Body length varies from 18 to 22 mm. The body expands from anterior to posterior, later narrow. The body is about 2.5-3 times as long as wide. The width of pereonites increases from 1 to 5, after that decreasing. Pereonite 5 widest, pereonite 7 narrowest. 3-7 coxal plates visible in dorsal view. 3-5 pereonites are approximate of equal length, pereonite 7 shortest. The eyes are small, concealed by antennula and antenna. Pleotelson wider than large, posterior margin rounded. All pleonites visible, first pleonite narrowest. 2-5 pleonites wider than the first one. All pleonites are of equal length. Antennula is extending to behind the eye, composed of seven articles. Antenna extending to the middle of 1 pereon. Pereon, with eight articles. Mandible palp third article distinctly shorter than others, without setae. Maxillula with four terminal spines, one long and three short spines. Maxilla medial lobe with 2 spines, lateral lobe with 3-4 spines. Maxilliped article 6 with five hooked spines in ovigerous and non-ovigerous females. Pereopods 1-3 are slightly smaller than 4-7, all without spines. The expansion on the upper and lower parts of 5-7 pereopod is distinct from 1-4 pereopods. Pleopods gradually decrease in length. Pleopods 1 to 5 has a medial peduncle margin with 4 hooks. Uropod rami extend to the posterior margin of the pleotelson. Exopod is slightly longer than endopod.

Distribution: The Mediterranean Sea, Adriatic (Trilles 1994; Schotte 2008b).

Hosts: *Boops boops* (Montalenti 1948; Kırkım 1998); *Spicara smaris* (Montalenti 1948; Berner 1969; Papoutsoglou 1976; Trilles 1977; Trilles et al. 1989; Ramdane et al. 2009; Radujkovic et al. 1984; Kırkım 1998); *Spicara maena* (Montalenti 1948; Berner 1969; Romestand et al. 1976; Öktener and Trilles 2004); *Sarpa salpa* (Montalenti 1948); *Centracanthus cirrus* (Schioedte and Meinert 1883); *Pagellus acarne*, *Raja clavata* (Trilles et al. 1989); *Scomber scombrus* (Balcells 1954); *Dicentrarchus labrax* (Papapanagiotou et al. 1999); *Clupea* sp (Trilles 1977).

Figure 9. 1*Emetha audouini* ♀

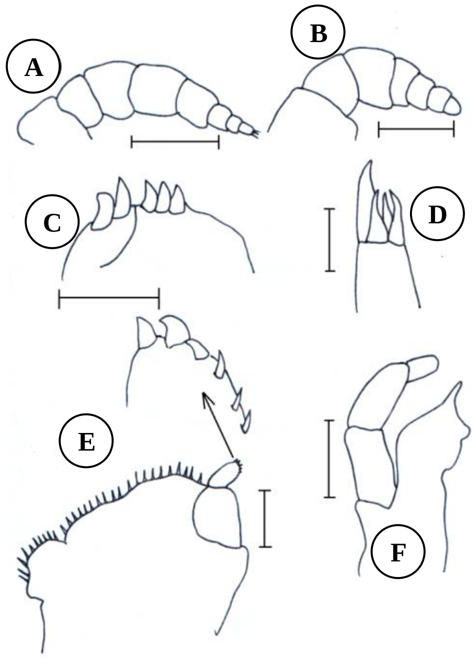


Figure 11. *Emetha audouini* ♀, A. antenna (0.68 mm), B. antennula (0.74 mm), C. maxilla (0.21 mm), D. maxillula (0.11 mm), E. maxilliped (0.24 mm), F. mandible (0.34 mm)

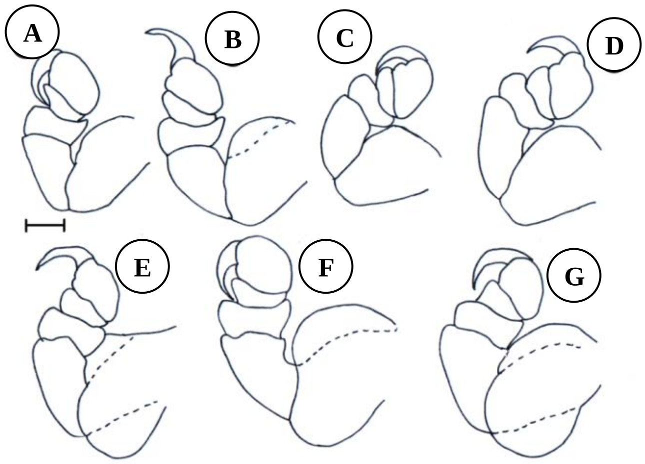


Figure 12. *Emetha audouini* ♀, A-G. pereopods I-VII (0.56 mm)

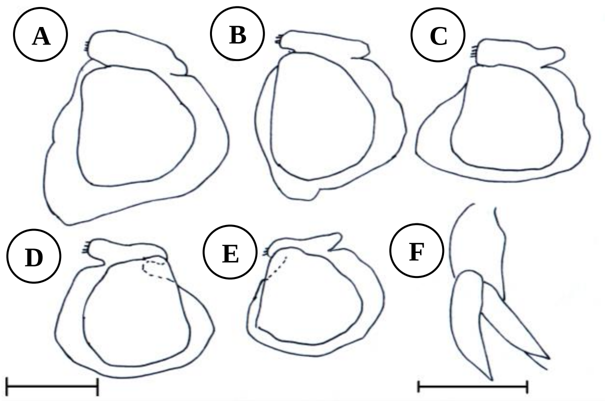


Figure 13. *Emetha audouini* ♀, A-E. pleopods I-V (1.40 mm), F. Uropod (0.96 mm)

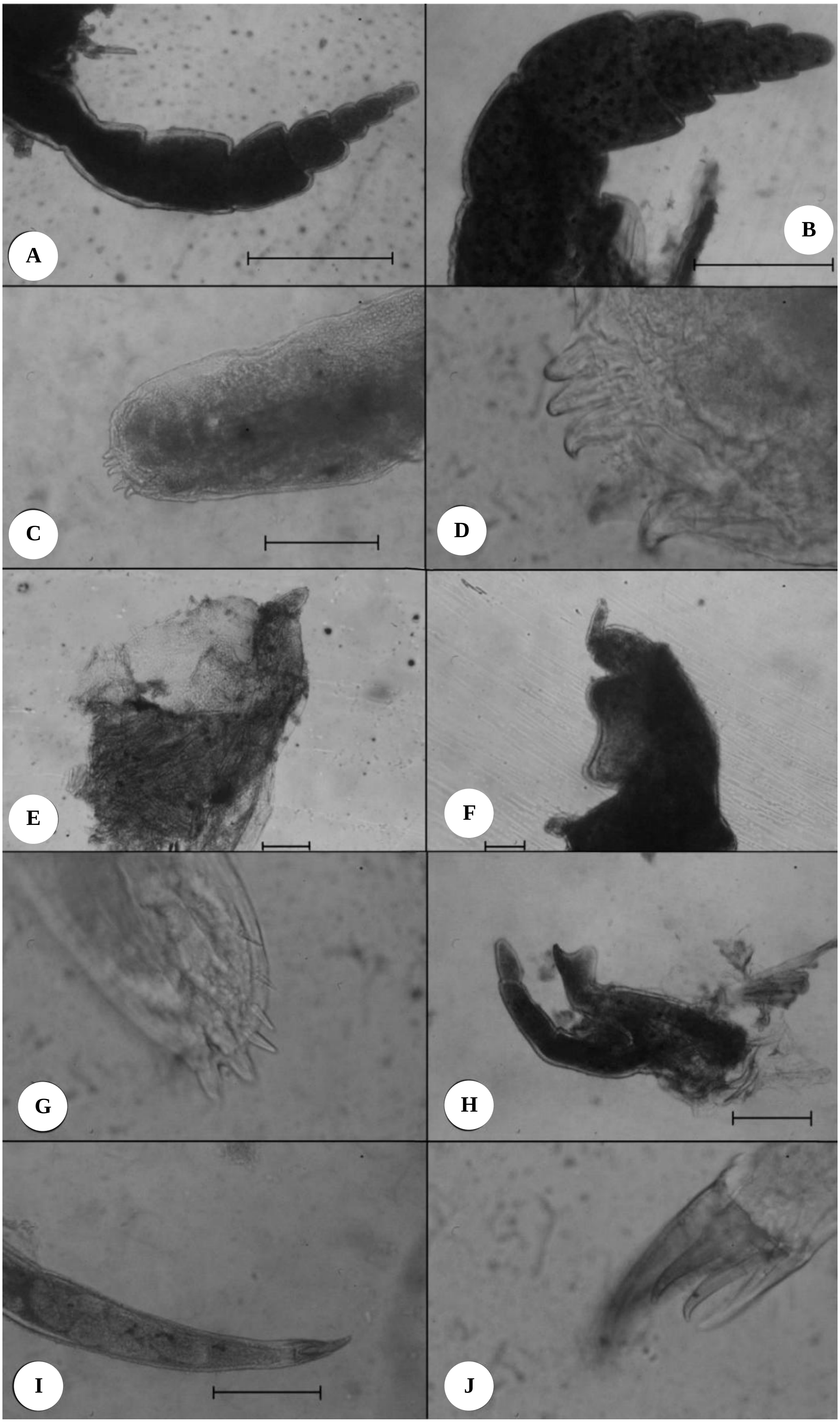


Figure 10. *Emetha audouini* ♀, A. antenna (0.68 mm), B. antennula (0.37 mm), C. maxilla (0.24 mm), D. distal of maxilla, E. maxilliped of non-ovigerous female (0.17 mm), F. maxilliped of ovigerous female (0.21 mm), G. distal of maxilliped, H. mandible (0.35 mm), I. maxillula (0.30 mm), J. distal of maxillula.

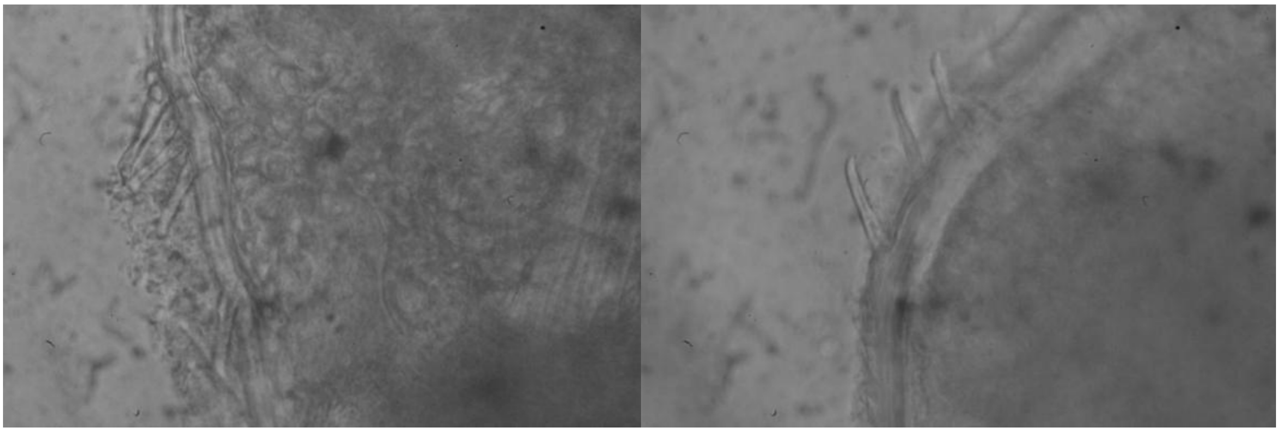


Figure 14. Four hooks on medial peduncle margin of pleopods (*Emetha audouini* ♀)

The host's parasitism with *Emetha audouini* was examined according to the family characteristics; 30% of 10 host species belong to Sparidae; 30% to Centracanthidae; 40% to Rajidae, Scombridae, Moronidae, and Clupeidae. According to habitat selections, the host's parasitism with Emetha audouini was examined; 30% of 10 host fish species are benthopelagic; 30% demersal; 30% pelagic-neritic; 10% pelagic-oceanic. The host's parasitism with *Mothocya taurica* was examined according to feeding habits; 60% of the 10 host fish species are omnivorous; 40% carnivorous.

Fish of the Centracanthidae family are the preferred hosts for *Emetha audouini,* which mainly select omnivorous and benthopelagic fishes.

Remarks: The antennula with 7 articles and antenna with 8 articles are were observed in this study, while antennula and antenna with 7 articles are indicated by Montalenti (1948); antennula with 7 articles and antenna with 9 articles by Trilles (1972) and Kırkım (1988); antennula and antenna with 8 articles by Schioedte ve Meinert (1883). The maxillula with four terminal spines found in this study is compatible with Trilles (1968, 1972) and Montalenti (1948)'s findings. The medial lobe with 4-7 spines and lateral lobe with 6-12 spines of the maxilla is found in this study, while the medial lobe without spines and the lateral lobe with 4 spines were found by Montalenti (1948); medial lobe with 1 spine and lateral lobe with 3 spines by Trilles (1968, 1972). The third article with setae on the lateral margin of the mandible palp found in this study is compatible with Trilles's (1968, 1972) and Montalenti's (1948) are finding. This study found six spines on article 3 of maxilliped of ovigerous and non-ovigerous females, while 6 spines on an article of only ovigerous female maxilliped were described by Trilles (1968). The observed expansions based on pereopod 7 distinct from pereopods 1-4 are compatible with Trilles (1968, 1972) and Montalenti (1948) findings. The pleopods 1 to 5 having peduncle medial margin with 4 hooks are found for the first time as distinct from the previous studies.

REFERENCES

Amar R. 1951. Isopodes marins de Banyuls. Vie et Milieu 2 (4): 529-530.

Balcells E. 1953. Sur des isopodes, parasites de poissons. Vie et Milieu 4 (3): 547-552.

Bariche M, Trilles JP. 2005. Preliminary check-list of Cymothoids (Crustacea, Isopoda) from Lebanon, parasitingon marine fishes. Zool Middle East 34 (1): 5-12. DOI: 10.1080/09397140.2005.10638082.

Berner L. 1969. Les principaux Cymothoides (Crustaces Isopodes) du Golfe de Marseille. Bulletin du Museum d'Histoire Naturelle de Marseille 29: 93-95.

Borcea I. 1933a. *Livoneca pontica* nov. sp., copepode parasite des Aloses et Sardines de la Mer Noire. Bulletin du Muséum National d' Histoire naturelle de Paris 2: 128-129.

Borcea I. 1933b. *Livoneca pontica* nov. sp., Cymothoide parasite des Aloses et Sardines de la Mer Noire. Annales Scientifiques de l' Université de Jassy 17: 481-502.

Bovallius C. 1885. New or imperfectly known Isopoda. Part I.Bihang till Kongl. Svenska vetenskaps-akademiens handlingar 10 (2): 1-32.

Bruce NL. 1986. Revision of the Isopod crustacean genus *Mothocya* Costa, in Hope, 1851 (Cymothoidae: Flabellifera), parasitic on marine fishes. J Nat Hist 20 (5): 1089-1112. DOI: 10.1080/00222938600770781.

Brusca RC. 1981. A monograph on the Isopoda Cymothoidae (Crustacea) of the eastern Pacific. Zool J Linn Soc 73 (2): 117-199. DOI: 10.1111/j.1096-3642.1981.tb01592.x.

Carausu A. 1959. Contribution à l'étude des Cymothoinae (Isopodes Parasites) de la mer Noire. 2. Un cas d'infestation massive avec *Livoneca punctata* (Ulj.) chez *Caspialosa pontica* (Eichw.). Trav. Lucrarile Sesiunii Stiintifice a Statiunii Soologice Marine Agigea 1959: 349-351.

Carus JY. 1885. Prodromus faunae mediterranae sive Descriptio Animalium maris Mediterranei incolarum quam comparata silva rarum quatenus innotuit adiectis locis et nominibus vulgaribus eorumque auctoribus in commodum zoologorum. Schweizerbart, Stuttgart. DOI: 10.5962/bhl.title.11523.

Dollfus RP, Trilles JP. 1976. A propos de la collection R. Ph. Dollfus, mise au point sur les Cymothoadiens jusqu' à présent récoltés sur des Téléostéens du Maroc et de l' Algérie. Bulletin du Muséum National d' Histoire naturelle de Paris 272: 821-830.

Dudich E. 1931. Systematische und biologische Untersuchungen uber die Kalkeinlagerungen des Crustaceenpanzers in polarisiertem lichte. Zoologica Stuttgart 30 (80): 1-54.

Euzet L, Trilles JP. 1961. Sur l'anatomie et la biologie de Cyclocotyla bellones (Otto, 1821) (Monogenea-Polyopisthocotylea). Revue Suisse de Zoologie 68 (2): 182-193. DOI: 10.5962/bhl.part.117723.

Froese R, Pauly D. Editors. 2017. FishBase. World Wide Web electronic publication. www.fishbase.org, version (06/2017).

Gerstaecker A. 1901. Isopoda In: Bronn HG (ed) Die Klassen und Ordnungen der Arthropoden wissenschaftlich dargestellt in Wort und

Bild.Crustacea (Zweite Hälfte: Malacostraca), Fünfter Band. II., Abtheilung.

Gourret P. 1891. Les Lemodipodes et les Isopodes de Golfe de Marseille. Annales du Musée d'Histoire Naturelle de Marseille 4 (1): 1-44.

Hadfield KA, Horton T, Bruce NL, Schotte M. 2017. Mothocya Costa in Hope, 1851. In: Boyko CB, Bruce NL, Hadfield KA, Merrin KL, Ota Y, Poore GCB, Taiti S, Schotte M, Wilson GDF (eds) (2008 onwards). World Marine, Freshwater, and Terrestrial Isopod Crustaceans database. http://www.marinespecies.org/aphia.php?p=taxdetails&id=118416.

Heller C. 1866. Carcinologische Beiträge zur Fauna des adriatischen Meeres. Verhandlungen der Zoologisch-botanischen Gessellschaft in Wien 16: 723-760.

Hope FG. 1851. Catalogo dei crostacei Italiani e di molti altri del Mediterraneo. Stabilemento Tipografico di Fr. Azzolino, Napoli. DOI: 10.5962/bhl.title.3924.

Josipa F, Mirela P, Sanja M, Jakov D. 2007. An occurrence of isopode parasitism on black scorpion fish, *Scorpaena porcus* (Perciformes, Scorpaenidae) in the eastern part of Adriatic Sea. *12th European Congress of Ichthyology* (ECI-12) Book of abstracts / Buj, Ivana; Zanella, Linda; Mrakovčić, Milorad (ed).-Zagreb: Tipomat 91.

Kırkım F. 1998. Ege Denizi Isopoda (Crustacea) Faunasının Sistematiği ve Ekolojisi Üzerine Araştırmalar. Doktora Tezi, Ege Üniversitesi, Fen Bilimleri Enstitüsü, İzmir, 233syf. [Turkish]

Kononenko AF. 1988. Parasitic Isopoda of the Black Sea fishes. Wiadomosci Parazytologiczne 34 (1), 1p.

Kussakin OG. 1979. Marine and Brackish Water lsopod Crustacea. Suborder Flabellifera. Academy of Science, USSR, Leningrad. [Russian]

Markewitsch AP. 1934. Skorupiaki pasorzytnicze ryb. Ukrainy. Die Schmarozerkrebse der Fische der Ukraine. Annales Museum Zoology Polonia 12: 223-249.

Martin MB, Bruce NL, Nowak BF. 2013. Redescription of *Ceratothoa carinata* (Bianconi, 1869) and *Ceratothoa oxyrrhynchaena* Koelbel, 1878 (Crustacea: Isopoda: Cymothoidae), buccal-attaching fish parasites new to Australia. Zootaxa 3683 (4): 395-410. DOI: 10.11646/zootaxa.3683.4.4.

Milne-Edwards H. 1840. Histoire Naturelle des Crustacés Comprenent l'anatomie, la physiologie et la classification de ces animaux. Roret, Paris.

Montalenti G. 1941. Studi sull'ermafroditismo dei Cimotoidi. 1. *Emetha audouinii* (M. Edw.) e *Anilocra physoides* (L.). Pubblicazioni della Stazione Zoologica di Napoli 18 (3): 337-394.

Montalenti G. 1948. Note sulla sistematica e la biologia di alcuni Cimotoidi del Golfo di Napoli (gen. Emetha, Mothocya, Anilocra, Nerocila). Archivio di Oceanografia e Limnologia, Venezia 5: 25-81.

Muradian Z. 1972. Contribution a La Connaissance de la Parasitofaune des Clupeides (Clupeidae, Pisces) de Roumanie. Travaux du Museum D'Histoire Naturelle Grigore Antipa 12: 11-25.

Nikolaeva VM. 1963. La faune parasite de quelques bancs locaux de poissons de la Mer Noire. Trav Station Biology of Sabastopol (traduction CNRS du texte Russe) 16: 1-46.

Öktener A, Trilles JP, Alaş A, Solak K. 2009. New hosts for species belonging to the genera *Nerocila, Anilocra, Ceratothoa, Mothocya,* and *Livoneca* (Crustacea, Isopoda, Cymothoidae). Bull Eur Assoc Fish Pathol 29 (2): 49-54.

Öktener A, Trilles JP, Koç HT, Erdoğan Z. 2010. *Mothocya taurica* (Czerniavsky, 1868) female redescription on *Alosa fallax* from the Black Sea Coasts of Turkey. Egyptian J Aquat Res 36 (4): 623-629.

Öktener A, Trilles JP. 2004. Report on the Cymothoids (Crustacea, Isopoda) collected from marine fishes in Turkey. Acta Adriatiaca 45 (2): 145-154.

Olguner A. 2008. Bazı Deniz Balıklarında Görülen Metazoan Parazitler. Yüksek Lisans Tezi, Sinop Üniversitesi, Sinop, 50syf [Turkish]

Papapanagiotou EP, Trilles JP, Photis G. 1999. First record of *Emetha audouini*, a cymothoid isopod parasite, from cultured sea bass *Dicentrarchus labrax* in Greece. Dis Aquat Org 38 (3): 235-237. DOI: 10.3354/dao038235.

Papoutsoglou SE. 1976. Metazoan parasites of fishes from Saronicos Gulf, Athens, Greece. Thalassographica, Institute of Oceanography Fishery Research Greece 1 (1): 69-91.

Poore GCB, Bruce NL.2012. Global diversity of marine Isopods (Except Asellota and Crustacean Symbionts). PLoS One 7: 1-15. DOI: 10.1371/annotation/3260cd00-89cf-4e08-ab25-07e0be598ab4.

Popov AM. 1933. Über parasitische Isopoden von Fischen aus dem Schwarzen Meer. Zoologischer Anzeiger Akademische Verlagsgesellschaft, MBH Leipzig 101: 193-198.

Quignard JP, Zaouali J. 1980. Les lagunes pé-riméditerranéennes. Bibliographie ichtyologique annotée. Premiere partie: les étangs français de Canet à Thau. Bulletin de l'Office National des Pêches de Tunisie 4: 293-360.

Quintard-Dorques B. 1966. Contribution à l'étude des poissons de la famille des Centracanthidae, Genra *Spicara* de la région de Sète. Annales de l'Université et de l'Association Régionale pour l'étude et al Recherche Scientifiques 4: 79-88.

Radujkovic BM, Romestand B, Trilles JP. 1984. Les isopodes parasites de la faune yougoslave. I. Cymothoidae parasites de poissons marins de la région de l'Adriatique Méridionale. Acta Adriatica 25: 161-181.

Radujković BM. 1982. Isopoda-parasites of the south Adriatic economically important fish species. Acta Adriatica 23 (1/2): 153-161.

Ramdane R, Bensouilah MA, Trilles JP. 2006. Comparison entre les Cymothoidae (Crustacea, Isopoda) recoltes sur les poissons teleosteens des cotes Tunisiennes et Algeriennes. Bull l'INSTM N (spécial 11: Actes des 8èmes Journées des Sciences de la Mer): 29-33.

Ramdane Z, Bensouilah MA, Trilles JP. 2009. Étude comparative des crustacés isopodes et copépodes ectoparasites de poissons marins algériens et marocains. Cybium 33 (2): 123-131.

Rathke H. 1837. Zur fauna der Krym. Memoires des Savaus Etrangers III: 291-454.

Romestand B, Trilles JP. 1977. Influence des Cymothoadiens (Crustacea, Isopoda, Flabellifera) sur certaines constantes hématologiques des poissons hôtes. Zeitschrift für Parasitenkunde 52 (1): 91-95. DOI: 10.1007/BF00380562.

Romestand B, Voss-Foucart MF, Jeuniaux Ch, Trillès JP. 1976. Les Acides AminésLibres Du Sérum Des Cimothoidae (Crustacés, Isopodes, Parasites De Poissons) Et De Quelques Téléostéens'. Arch Physiol Biochem 84 (5): 981-988. DOI: 10.3109/13813457609069459.

Sartor SM. 1986. Incidencia de isopodos parasitas (Cymothoidae) em peixes da plataforma continental Brasileira. Bolletin Institute Oceanography Saint Paulo 34: 1-12. DOI: 10.1590/S0373-55241986000100001.

Sartor SM. 1987. Desenvolvimento marsupial e ciclo de vida de Cymothoa liannae Srtor et Pires (Isopoda, Cymothoidae), parasita de peixes. Bolletin Institute Oceanography Saint Paulo 35 (1): 43-51. DOI: 10.1590/S0373-55241987000100006.

Schioedte JC, Meinert F. 1883. Symbolae and monographium *Cymothoarum crustaceorum* familiae. III. Saophridae. IV.Ceratothoinae. Naturhistorisk Tidsskrift, Kjobenhavn 13: 281-378.

Schotte M. 2007. Emetha Schioedte & Meinert, 1883. In: Boyko CB, Bruce NL, Hadfield KA, Merrin KL, Ota Y, Poore GCB, Taiti S, Schotte M, Wilson GDF (eds) (2008 onwards). World Marine, Freshwater, and Terrestrial Isopod Crustaceans database. http://marinespecies.org/aphia.php/aphia.php?p=taxdetails&id=118411.

Schotte M. 2008a. Mothocya taurica (Czerniavsky, 1868). In: Boyko CB, Bruce NL, Hadfield KA, Merrin KL, Ota Y, Poore GCB, Taiti S, Schotte M, Wilson GDF (eds) (2008 onwards). World Marine, Freshwater, and Terrestrial Isopod Crustaceans database. http://www.marinespecies.org/aphia.php?p=taxdetails&id=118904.

Schotte M. 2008b. *Emetha audouini* (H. Milne Edwards, 1840). In: Boyko CB, Bruce NL, Hadfield KA, Merrin KL, Ota Y, Poore GCB, Taiti S, Schotte M, Wilson GDF (eds) (2008 onwards). World Marine, Freshwater, and Terrestrial Isopod Crustaceans database. http://www.marinespecies.org/traits/aphia.php?p=taxdetails&id=118894.

Stalio L. 1877. Catalogo metodico e descrittivo dei Crostacei podottalmi ed Edriottalmi dell' Adriatico. Atti de Reale Instituto Veneto di Scienze, Lettre ed Arti 3: 1-274. DOI: 10.5962/bhl.title.10387.

Stossich M. 1880. Prospetto della fauna de mare Adriatico. Parte III. Bollettino della Societa Adriatica di Scienze Naturale in Trieste 6: 1-95.

Thampy DM, John PA. 1974. Sex-reversal and androgenic gland in the fish parasite Irona far (Cymothoidae: Isopoda: Crustacea). Intl Parasitol 4 (6): 575-583. DOI: 10.1016/0020-7519(74)90021-6.

Trilles JP, Radujkovic BM, Romestand B. 1989. Parasites des Poissons Marins du Montenegro: Isopodes. Acta Adriatica 30 (1/2): 279-306.

Trilles JP. 1962. Remarques morphologiques et biologiques sur les 'Isopodes Cymothoida, parasites des poissons, de l'Etang de Thau. Naturalia Monspelogia 3: 101-124.
Trilles JP. 1964. Specificite parasitaire chez les isopodes Cymothoidae Mediterraneens. Note preliminaire. Vie et Milieu 15: 105-116.
Trilles JP. 1968. Recherches sur les Isopodes. Cymothoidae de cotes Françaises. I. Systematique et faunistique. II. Bionomie et parasitism. [These de Doctorat sciences], CNRS, AO 2305, Montpellier, France.
Trilles JP. 1972a. Les Cymothoidae (Isopoda, Flabellifera) des côtes françaises (systématique, faunistique, écologie et répartition géographique). I. Les Ceratothoinae Schiodte and Meinert, 1883. Bulletin du Muséum national d'Histoire naturelle, Paris, 3e série, Zoologie 91: 1191-1230.
Trilles JP. 1972b. Les Cymothoidae (Isopoda, Flabellifera) du Muséum National d'Histoire Naturelle de Paris. Etude critique accompagnée de précisions en particulier sur la répartition géographique et l'écologie des différentes espèces représentées. I. Les Ceratothoinae Schioedte et Meinert, 1883. Bulletin du Muséum d'Histoire Naturelle, Paris, 3e série, Zoologie 91: 1231-1268. [French]
Trilles JP. 1972c. Sur la structure des plkopodes et des oosttgites chez les Cymothoidae (Isopoda, Flabellifera) et ses rapports avec les caractbistiques kologiques des espPces. Bulletin du Muséum National d'Histoire Naturelle, Paris, 3ème série 91 (3): 1269-1277.
Trilles JP. 1976. Les Cymothoidae (Isopoda, Flabellifera) des collections du Museum National d'Histoire Naturelle de Paris. IV. Les Lironecinae Schioedte et Meinert, 1884. Bulletin du Museum National d'Histoire NatureIle, Paris, 3 serie 390 (272): 773-800.
Trilles JP. 1977. Les Cymothoidae (Isopoda, Flabellifera; parasites de poissons) du Rijksmuseum van Natuurlijke Historie de Leiden. Méditerranée et Atlantique nord-oriental. Zoologische Mededelingen (Leiden) 52: 7-17.
Trilles JP. 1994. Les Cymothoidae (Crustacea, Isopoda) du Monde (Prodrome pour une Faune). Studa Marina 21/22: 1-288.
Uljanin VN. 1871. Beitrage zur fauna des Schwarzen Meeres. Protok. Sitzungsber Naturf. Antrop. Und Ethnogr. Gesellsch. Moscou, 113-114.
Uljanin VN. 1872. Data on the fauna of the Black Sea. lzvestiya Moskovskogo Obshchestva Lyubitelei Estestvoznaniya, Antropoligii i Etnografii 9: 79 132.
Vasiliu G, Carausu MA. 1948. Contribution *A* l'etude des *Cymothoinae* (Isopodes parasites) de la Mer Noire. Annales Scientifiques de l' Université de Jassy 31: 175-188.
Wägele JW. 1987. Evolution und phylogenetisches System der Isopoda: Stand der Forschung und neue Erkenntnisse. Universität Oldenburg.
WoRMS Editorial Board. 2017. World Register of Marine Species. Available from http://www.marinespecies.org at VLIZ. DOI: 10.14284/170.

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 65-73

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070202

An assessment of a tropical urban stream using benthic macroinvertebrates as a bio-indicator in Muara Angke, Jakarta, Indonesia

CHRISTOPHER KELLY[1,♥], TATANG MITRASETIA[2,♥♥], JITO SUGARDJITO[3,♥♥♥]
[1]Kings College. Strand, London WC2R 2LS, London, United Kingdom. ♥email: chris.kellyy@gmail.com
[2]Faculty of Biology, Universitas Nasional. Ps. Minggu, Jakarta Selatan 12520, Jakarta, Indonesia. ♥♥email: tatang248@gmail.com
[3]Centre for Sustainable Energy and Resources Management, Universitas Nasional. Ps. Minggu, Jakarta Selatan 12520, Jakarta, Indonesia. ♥email: sugar@unas.ac.id

Manuscript received: 31 August 2016. Revision accepted: 28 June 2017.

Abstract. *Kelly C, Mitrasetia T, Sugardjito J. 2017. An assessment of a tropical urban stream using benthic macroinvertebrates as a bio-indicator in Muara Angke, Jakarta, Indonesia. Bonorowo Wetlands 7: 65-73.* A preliminary study was conducted on the populations, density, and distributions of benthic macroinvertebrate species along the course of the Ciliwung/Angke River, Jakarta. The selected river section is highly urbanized and heavily polluted, and five study sites were selected along its course to assess the health of the river ecosystem. Benthic macroinvertebrates are used as a bio-indicator for the evaluation of ecosystem health. Their different pollution sensitivities, wide distribution, and relatively sedentary lives make them valuable candidates for this role. The focal point of the study is the Muara Angke mangrove forest and wetland, a small remnant wetland at the mouth of the river in Jakarta Metropolitan. A comparison is drawn between the different study sites to assess this unique habitat's health and potential utility. Despite several intense pressures resulting from the highly urbanized surroundings, and the mangrove's position at the mouth of the river where pollution accumulation would be highest, the data collected from this site showed a macroinvertebrate population dominated by pollution -sensitive taxa, suggesting the mangrove itself continues to provide significant ecosystem services in water -purification.

Keywords: Biological filter, urban wetland, ecosystems services, benthic macroinvertebrates

INTRODUCTION

At the margins of the land, mangrove habitat has long been recognized as a critical component of nutrient transfer and species movement across large eco-regions. They have been identified as cornerstone ecosystems, supporting healthy functioning in other habitats through the food web (Mastaller 1997). It has been demonstrated in numerous simulation studies that coastal wetland plants are capable of absorbing excess phosphates, nitrates, and other organic materials, helping to prevent eutrophication of watercourses and the asphyxiation of downstream biotic communities (Wong et al. 1997; Chu et al. 1998; Tilley et al. 2002). This is crucial for rivers receiving high levels of organic waste, particularly downstream from large-scale agriculture operations that use excessive nitrate and phosphate loading to increase yields. However, the nutrient load entering mangroves often exceeds wetland plant growth requirements, and other biological filtering mechanisms are also essential factors (Ye et al. 2001).

It is not only sedentary mangrove species that depend on coastal wetlands. Studies have demonstrated that many fish species rely on mangroves to provide a nursery habitat for their juveniles (Sasekumar et al. 1992). Many migratory birds visit these rich environments for food and shelter (Giesen et al., 2007). This makes coastal wetlands vital ecosystems in the regional context, yet applying this knowledge to coastal development in the Jakarta Metropolitan area is worryingly under-developed. In recent years, extreme degradations in Jakarta-bay's marine and coastal habitats have been observed, an ecologically and commercially significant area. Reductions in size and health of coastal wetlands have already begun to undermine several key processes outlined above and precipitated the decline of numerous species which rely on the mangrove. Perhaps most critical for this study is the ongoing reduction in commercial marine stocks. Not only do several key fish, shrimp, and mollusk species depend on mangrove habitat for at least one part of their life cycle (Burhanuddin 1993), they also depend on nutrients carried downstream from Java's fertile volcanic highlands passing through coastal wetlands (Ligtvoet et al. 1996). A robust coastal wetland has the potential to support several commercial operations for Indonesia's citizens, but this potential remains unrealized (Supartono et al., 2016).

This preliminary study investigates the influence of a unique urban wetland on regulating the water quality of a highly degraded urban stream and terrestrial-marine interactions more generally in one of the world's largest coastal metropolitan areas. The diversity, abundance, and community composition of benthic macroinvertebrates relative to wetland proximity were used to indicate overall ecosystem health and vigor in the Ciliwung-/Angke river. The ultimate purpose of this study is to appraise the immense value of one of Jakarta's remaining natural habitats in the hope of preserving its critical function in the face of otherwise unmitigated developmental pressures. The focus of this study will be the Muara Angke wetland, 170 hectares of remnant mangrove and wetlands which once dominated the Jakarta Bay, now serving as the city's last line of defense against coastal flooding and providing a home for several rare species (Palupi 2007). The role of coastal wetlands in mitigating Jakarta's Perennial flood crises has been well documented (Aerts et al. 2009; Firdaus 2013), particularly as a vital hydrological sink for the city's many rivers. Despite this, the size of the protected wetland area has continually been reduced from more than 1300 hectares due to population and developmental pressures (Nur et al. 2001).

MATERIALS AND METHODS

The study took place across five main sites along the course of the Ciliwung river, with varying degrees of proximity to the coastal wetland (Figure1). Since we are concerned with the impact of the Muara Angke Wetland on water quality being discharged into the Jakarta Bay and in the lower reaches of the river, and not to the effects of urbanized spaces on less contaminated upstream flows, the first samples will be collected from sites in densely urbanized areas representing the most significant anthropogenic influences on faunal community abundance and composition. From here, our five study sites will be divided into three groups based on their proximity to the wetland and surrounding land -use types to allow for both inter and intra-group comparisons where relevant.

This study uses sampled measurements of benthic macroinvertebrate community composition and density to indicate water quality and establish the significance of the remaining wetland area's bio-filter effect. This method has been employed in several previous studies worldwide (Rosenberg and Resh 1993;, Adakole and Annune 2003;, Diaz et al. 2013).

Benthic macroinvertebrates make excellent study specimens, being numerically abundant, readily collected and surveyed, comparatively easy to identify, and taxonomically rich (Dodson 2001). Most benthic macroinvertebrates, particularly in their larval stages, are also relatively sedentary, unable to quickly relocate, and displaying any effects of localized environmental conditions (Cairns and Prall 1993). In addition, bio-indication using benthic macro-invertebrates gives a clearer picture of water quality over time, mainly but limited to the life-cycles of identified species, which is reflected in the distribution and diversity of pollution sensitive versus pollution tolerant communities (Warwick 1993).

A total of 435 samples were taken across all study sites over four weeks period June – July 2015. Initially, it had been planned to take three samples per day at each site, but this was increased to six to reduce overall study length when slight daily variation in results was observed during the first weeks of collecting data. Instead, the six daily samples would be taken at several different specific locations surrounding each study site in an attempt to capture any spatial variation in benthic species distribution within a given area.

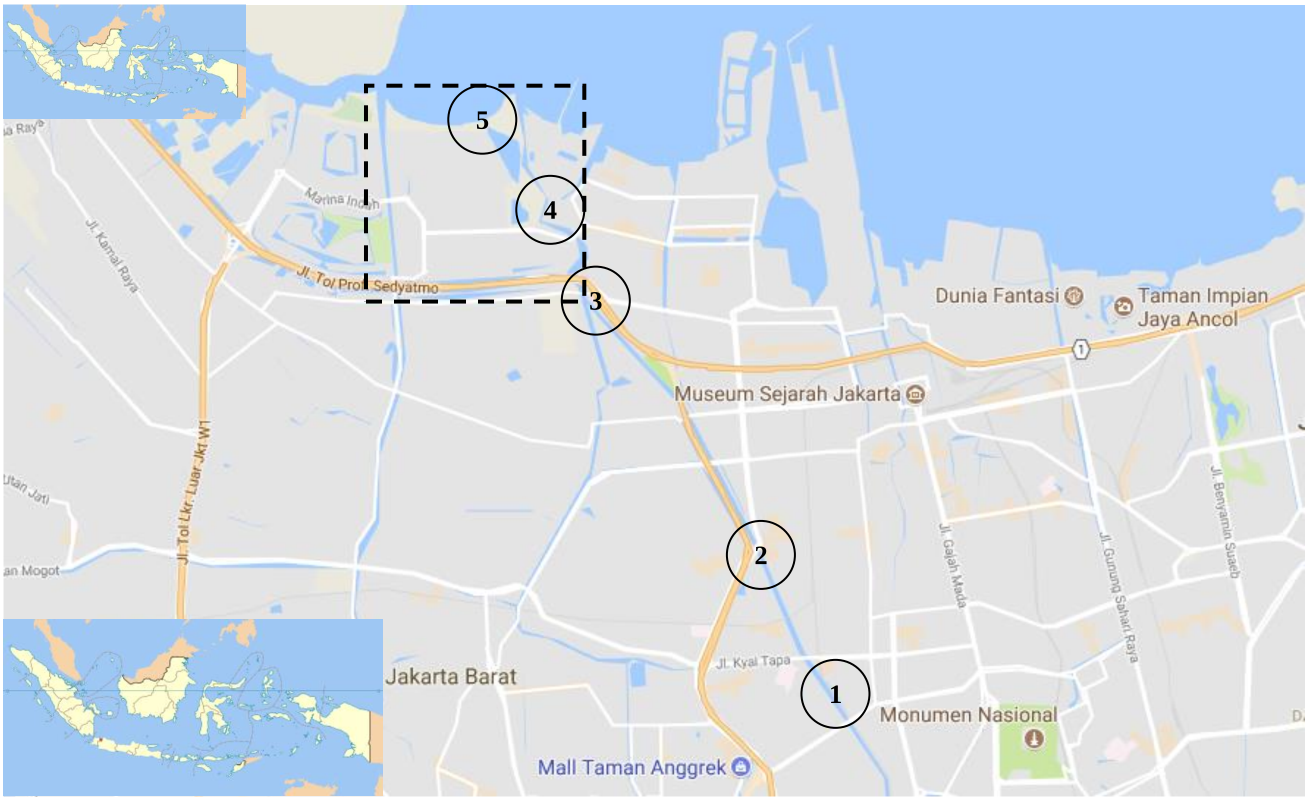


Figure 1. Situation map of Muara Angke, Jakarta, Indonesia. Box = Location of protected wetland (Muara Angke Wildlife Sanctuary)

Sampling methods involved basic disturbance sampling of river bed substrate and collection using a standard kick net. It is important to note that, due to river banks' unstable or otherwise hazardous conditions, including the build-up of highly soft and deep sediment at the waters' edge, samples were almost universally collected within two meters of the bank with deeper water samples collected where possible. Given the spatial distribution of different species throughout the benthic habitat, it is recognized that this may affect the validity of the results. Therefore, future studies may wish to employ boats or floating platforms to gather samples over a wider area.

The substrate was disturbed for thirty seconds at various points within each study site, with net contents emptied into a sample tray, spread evenly, and analyzed on-site in the field using a magnifying glass. In order to account for human error in species identification and counting, scrutiny of any given sample was limited to five minutes, or until satisfied that there were no further specimens to identify in that sample. It is also important to note that counts for a single species were capped at 50 in a given sample due to issues with counting large numbers of individuals accurately. Species were identified as precisely as possible, including considering the specific life-stage (i.e., larvae, pupae, adult). For analysis, the taxonomic group of collected specimens was deemed sufficient to conclude bio-indication given many different groups monitored and the practical difficulties of precise species identification in the field (Hodkinson and Jackson 2005).

Study areas

The main factor dictating the selection of specific sample sites was the ease of access. Large stretches of the river have artificial concrete banks that make access to the river bed extremely difficult. In the interests of safety, these areas were avoided in favor of sites where water access was possible on foot. To include a range of different circumstances influencing water quality and species community composition, sites were also selected to incorporate various nearby human activities and land uses. These sites were then grouped to reflect similarities in habitat conditions as follows: (i) Group 1 – 'Highly Urbanized' (sites 1 and 2) ; (ii) Group 2 – 'Transition Areas' (sites 3 and 4) ; (iii) Group 3 – 'Wetland Influenced' (site 5).

A brief description of each group and specific study sites is given below.

Group 1 – Highly Urbanized, representing habitats significantly affected by surrounding urban activities. This group experiences little or no influence from natural ecosystems.

Site 1 is called Roxy. It is located near the center of Jakarta's most densely populated area, is named after the adjacent Roxy Mall, and demonstrates a variety of surrounding urban land use types, including major roadways, a large mall parking area, a ground-level train route, a relatively small, informal food market and a mixture of established residential neighborhoods and *kampung* (Informal, low-income housing). High volumes of plastic waste are evident on the river banks and the water itself. This site represents the most extensive influence of various urban activities on the local benthic ecosystem. It is clearly a highly degraded habitat.

Site 2 is called 'Season City,' located near a major roadway that passes directly over the watercourse and is surrounded by many different settlement types. The urban density in the immediate vicinity is significantly less than further upstream, and the road is also considerably smaller. There is reduced land-use pressure on the river banks at this point, as evidenced by more extensive embankments, and several clusters of trees are growing adjacent to the watercourse.

Group 2 – Transition Areas, representing habitats with an increased natural component. While these sites cannot be labeled wetlands in the true sense, they are influenced by significantly more natural areas and may begin to experience a mild bio-filter effect.

Site 3 was named after Jakarta's airport toll road, which passes over the study site, responsible for converting previous wetlands and the inundation of numerous surrounding areas. At ground level, there is almost a total lack of paved or otherwise wholly impermeable surfaces; instead, the thoroughfares are largely dirt tracks connecting small pockets of informal settlement. The river banks themselves are less crowded with refuse than the sites in group 1. We can expect the numerous pockets of largely inundated green spaces surrounding the river at this stage to absorb some surface contamination, initiating a mild bio-filter effect.

Site 4, named 'Pluit,' lies at the upstream edge of the Muara Angke protected wildlife area. Despite its proximity to the largest intact area of near- pristine wetland, its position on the upstream fringe of this area limits the potential for wetland-derived improvements to local water quality. Furthermore, Site 4 is under greater pressure from local development projects and associated land-use changes than Site 3, which is surrounded by a network of fully paved roads and far more extensive human settlements, including the neighborhood of Muara Angke.

Group 3 – Wetland Influenced. These sites are sufficiently proximate to the remnant or naturalized wetland areas that we expect to see a significant influence of the aforementioned bio-filter effect on the local benthic community. This group is comprised only of site 5.

Site 5 is positioned just beyond the downstream edge of the Muara Angke Wildlife Protection Area, from which it derives its name. While this site is not within the conservation area itself, where the naturalized state of the wetland would doubtless deliver higher biodiversity, it was selected to provide an insight into the downstream bio-filter effect impact on the water having passed through the mangrove area. While this group contains only one site, it was also selected because it includes both a naturalized and artificially planted wetland area close to where samples can be collected.

We have divided the individuals collected into 3 groups representing those species capable of tolerating high levels of pollution (group 3), somewhat pollution tolerant species (group 2), and those species susceptible to pollution and degradation of aquatic habitats (group 1), according to several field guides and previous studies (Swari et al. 2014; Emere and Nasiru 2008; Azrina et al. 2006). This highlights the importance of exactly which species comprise a given ecological community when using bio-indication, in addition to basic abundance and biodiversity analyses. The purpose is to demonstrate a difference in community composition and abundance visually. We use the same primary data for the calculations and multiply the number of individuals by their pollution weighting to give our weighted abundances.

The Shannon Diversity Index (BDI) was calculated for the samples taken each day at all study sites, following Ogbeibu and Oribhabor's study. 2002). Several statistical tests such as Kruskal-Wallis and Mann-Whitney U test were performed on the data collected from the survey to determine the significance of differences between study sites and prove or disprove the stated hypotheses.

RESULTS AND DISCUSSION

Macroinvertebrate abundance and community composition

The overall macroinvertebrate abundance and community composition are summarized in table 1, with different taxa organized by their sensitivity to pollution. A total of 19 taxa were identified from a total sample of 7,168 individuals collected. The number of taxa identified at each site ranges from just 4 at site 1 (Roxy) to 11 at site 5 (Muara Angke), with a steady increase from each site further downstream, as anticipated from the increasing influence of more natural environments at each location. From group 3, we can assume a higher local water quality, perhaps attributed in part to the bio-filter effect, even if the overall biodiversity of species is equal or lower than other sites dominated by pollution tolerant species.

Site 1 displayed significantly lower species abundance and taxonomic diversity than the others (54 individuals, 0.75% of total). In contrast, recorded species were dominated by segmented worms of the orders *Oligochaetae* and *Hirudinae*; both species are known to be highly tolerant to pollution; abundance remained low throughout this study. The presence of fly larva (*Muscidae*), usually less tolerant of pollution, may be explained by the presence of discarded rotting meat and other organic material from nearby human settlements.

Site 2 saw a marked increase in the abundance and diversity of recorded taxa (239 individuals, 3.33% of total). *Oligochaetae* remained the most abundant taxa; however, several similar pollution tolerant species not present at site 1, including *Physidae*and *Chironomidae (bloodworm)*, were recorded in significant numbers, increasing the recorded diversity at this site.

The increase in species abundance and recorded taxa between sites 1 and 2 is perhaps a result of the larger surrounding earthen embankments, which may absorb some volume of contaminated run-off and provide a more suitable habitat for colonization. Furthermore, an increase in surrounding vegetation provides a greater organic nutrient input, particularly for *Physidae*, which are known to graze on decaying leaves (Magee 1993). The lower number of urban pollution sources identified in the immediate area is also likely to result in less degraded habitat. It is worth noting that the presence of a single larva from the family *Tipulidae* recorded at this site would appear to be anomalous, perhaps indicating that this individual had been dislodged from further upstream. Nevertheless, its impact on the overall trend in this study is minimal, and as such, it has been included in the final data set.

Site 3, the first of the identified 'transition sites' demonstrated a significant increase in invertebrate abundance (1,624 individuals, 22.66% of total) and a less important but notable increase in the identified taxa from a total of 7 for Sites 1 and 2 (Group 1) to 9 for Site 3 alone. A key factor that may explain this incremental increase in diversity is a similar increase in the variety of available micro-habitats along this stretch of the river, including variations in substrate, light and temperature conditions, and flow velocity. This may allow colonization of different points by new species combinations, and significant spatial heterogeneity was recorded for this study site.

There was an increase in abundance for all taxa recorded at upstream study sites except Hirudinae at site 3. A dramatic increase in the abundance of *Oligochaetae* by a factor of more than 7, clearly dominating this habitat. *Physidae* numbers also increased, and for the first time, *Ciroxidae* and *Noteridae* were recorded, indicating a significant increase in the digestible organic material present. This composition is clearly related to the presence of extensive riparian vegetation as compared to sites further upstream, given that each of these taxa is known to consume decaying plant matter (Anderson and Sedell 1979), as well as a build-up of organic and chemical pollutants tolerable to segmented worms in particular.

Unlike Site 3, the recorded abundances for many groups dropped at study site 4, perhaps attributed to a combination of heavy siltation generating warm, shallow, slow-moving water and the accumulation of upstream pollution sources allowing the remarkably tolerant *Oligochaetae* to out-compete other species for deposited organic nutrients. The two exceptions include *Notonectidae,* for which the slow-moving water likely provides a more suitable hunting ground than upstream river stretches, and the earthworms of *Lumbricidae,* for which the marshy banks and variable watermark permit the easy transition between riparian and newly flooded habitat.

Site 4 also saw a further increase in abundance (2,429 individuals, 33.89% of total), with the *Oligochaetae* worms cementing their near-total domination of the invertebrate faunal community (over 96% of recorded individuals). The worm population was very dense at this site. Since many samples contained individuals too numerous to count by hand accurately, they were allocated a count of 50+, and total population numbers are certainly under-represented.

As expected, site 5 contributed the largest number of individuals (2,822; 39.37% of total) and the greatest taxa (57.89% of total). Crucially, the taxonomic composition recorded at this location was dominated, overwhelmingly, by species identified as being sensitive to pollution and was unique amongst the 5 study sites in this regard. As identified below, all other sites were dominated by taxa from the 'pollution tolerant' group. Significant is the almost total absence of *Oligochaetae* worms, the dominant taxa at all other study sites, and their replacement as the dominant taxa by *Pachychilidae*, gilled snails known to be highly sensitive to pollution (Allen et al. 2012). *Atydae* comprises, the other significant pollution-sensitive taxa at this site is almost absent from upstream locations. Finally, the presence of predatory larvae, both dragonfly and damselfly from the order *Odanata*, at site 5 indicates a more complex food web than was identified at Site 1-4, where the majority of species documented were simply detritus feeders.

Species richness was calculated for each study site using the Shannon-Wiener index, and a significant increase was recorded for each site (27, 90.3, 541.3, 768.1, 850.9, respectively). The recorded individuals are listed by species in table 1, visualized further in figure 2.

Benthic macroinvertebrate abundance

An initial Kruskal-Wallis H test demonstrated a clear statistically significant difference in the abundances of macroinvertebrates recorded at the different study sites (x^2 (4) = 114.138, p<0.001). The mean rank scores for abundance for study sites 1-5 were 15.19; 44.41; 93.71; 115.66, and 96.03, respectively. To detect the differences in abundances recorded between pairs of study sites, we applied the Mann-Whitney U test. The results show a significant difference for all comparisons (p<0.001) except 3: 5 and 4: 5, where high counts of segmented worms at sites 3 and 4 approached the total individual count at site 5.

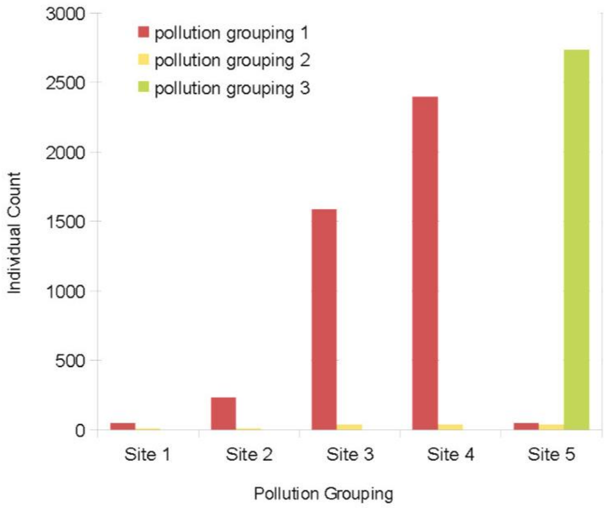


Figure 2. Distribution of individuals recorded taxa based on sensitivity to pollution (n=7,168)

Table 1. List of individuals' family taxa recorded at the study site (in Muara Angke, Jakarta, Indonesia) arranged in accordance to pollution sensitivity

Taxa	Site 1	Site 2	Site 3	Site 4	Site 5
Pollution Grouping 1					
Physidae (pouch snails)	0	6	20	8	0
Chironomidae (midges; larvae)	2	12	16	15	0
Chironomidae (bloodworms)	0	2	2	0	37
Oligochaetae (Aquatic Worms)	29	207	1517	2340	1
hirudinae (Leeches)	14	4	1	3	0
Ciroxidae (Water Boatman)	0	0	30	15	10
Notonectidae (Water Boatmen)	0	0	0	13	0
Pollution Grouping 2					
Muscidae (True Flies; larvae)	9	7	22	1	0
Simuliidae (Black Flies; larvae)	0	0	0	0	0
Noteridae (Beetles; adults; larvae)	0	0	15	6	36
Tipulidae (Crane Flies; larvae)	0	1	0	0	0
Hydropsychidae (Web-spinning Caddisflies; larvae)	0	0	0	1	0
Lumbricidae (Earthworms)	0	0	0	27	0
Nepidae (Water Scorpion; Ranatra)	0	0	0	0	2
Pollution Grouping 3					
Pachychilidae (Spiral Gilled Snails)	0	0	0	0	2416
Ampullariidae (Apple Gilled Snails)	0	0	0	0	70
Atydae (Shrimp)	0	0	1	0	212
Parathelphusidae (crabs)	0	0	0	0	2
Odanata (Damselfly; larvae)	0	0	0	0	12
Odanata (Dragonfly; larvae)	0	0	0	0	24
Total	54	239	1624	2429	2822

Although these results clearly demonstrate a significant difference between the abundances recorded at upstream sites with the least influence by the wetland and those sites further downstream, the mean rank abundances recorded at sites 3 and 4 were higher than at site 5. Although these results were not statistically significant, the similarities in higher abundances recorded at the 3 furthest downstream sites compared to those upstream suggest that wetland proximity and the presence of river- adjacent green spaces positively influence benthic biotic activity.

Weighted abundances by pollution grouping

As previously stated, to contextualize subsequent biodiversity analyses, it is important to consider the different pollution sensitivities of recorded species. A dramatic change in community composition favoring species sensitive to water-borne pollution was recorded at site 5, mainly influenced by the surrounding Muara Angke wetland. A subsequent analysis will now be conducted to determine the statistical significance of this change. The weighted abundances show an exaggerated increase in species count at site 5 compared with the abundances recorded in figure 3, which was expected given the dominance of pollution-sensitive species in that area. These test results display a greater, more significant difference between the study sites ($p<0.001$ for all comparisons) and, crucially, exaggerate the differences between sites 3, 4, and 5 because of the different weightings assigned to the species found there. This is important for this study because pollution sensitive species are more significant indicators of ecosystem health, and the distortion of our initial findings when pollution weightings are applied has increased our weighted abundances by a greater factor at each successive downstream study site when compared to the results before pollution sensitivity is accounted for.

Species diversity

As outlined in our preliminary analysis, the recorded species richness increased with each site close to the Muara Angke wetland. Still, due to the domination of specific taxonomic groups, a subsequent biodiversity analysis will be conducted to give a more accurate picture of the relationship between wetland proximity and biotic diversity at each site. Initial Kruskal-Wallis H test analysis demonstrated a clearly significant difference in Shannon Biodiversity Index rating between the 5 study sites: $x_2 (4) = 31.04$, $p<0.001$, with mean ranks of 58.03; 87.67; 61.02; 54.50 and 103.78 for sites 1-5, respectively.

To determine the significant differences between individual study sites and to enable a direct comparison, a Mann-Whitney U test was performed on the Shannon BDI results for paired site results. The results indicate the most significant differences in biodiversity can be found when comparing sites 3 and 4 to study site 5, with lower counts of fewer species accountable for the statistical insignificance recorded between upstream sites.

The results of the BDI comparison support several points. First of all, the near -total dominance of aquatic worms from the *Oligochaetae* taxa amongst the collected individuals, particularly at sites 3 and 4, has resulted in a lower BDI rating at these sites despite the higher number of total species recorded there when compared with site 2, for example. Similarly, the domination of recorded individuals at site 5 by the *Pachychilidae* gilled snails may explain the lack of significant difference in biodiversity between site 5 and site 2, where the domination by a single species, although present, was far less dramatic.

Biodiversity by site group

The five study sites from which samples were collected were placed in 3 groups to reflect similarities in local conditions, wetland proximity, and other factors that affected benthic faunal composition. No significant difference was calculated for the BDI between study site group 1 and study site group 2 ($U=1477$, $p=0.254$), consistent with the observation of low taxonomic diversity across all sites in the watercourse areas upstream of the wetland, where slight variation in species was also recorded. The differences in diversity between study site group 1 and the heavy wetland-influenced site in group 3 were more significant (mean ranks: 4.32, 51.36 respectively; $U=627.5$, $p=0.05$). Between-group 2 and group 3, the divergence was even more important (mean ranks: 31.36, 65.48 respectively;, $U=160$, $p<0.001$).

Discussion

Species and pollution in the Ciliwung River

Although the biodiversity analysis results do not fully support the hypothesis that proximity to the wetland correlates with a significantly higher biodiversity rating, they help draw several important conclusions. Namely, it is important to note that the biodiversity ratings at site 5 were consistently higher than at other sites and significantly higher than at sites 3 and 4, which rank second and third in our study for species richness and specimen abundance and share a similar dominance in recorded specimens by a single taxon. Secondly, this significant difference in BDI rating between sites 3 and 4 (group 2) and site 5 (group 3) was recorded despite only a single additional taxon being recorded at site 5, indicating that the benthic macroinvertebrate community itself is more diverse and thus in better ecological health (Schläpfer et al. 1999).

What, then, does this specific composition of invertebrate species and their abundances reveal the water quality of the Ciliwung river system throughout its lower, urbanized reaches, and, importantly, about the role of the Muara Angke Wetland therein? Firstly, it is clear that dividing the study sites into groups as stated in the methodology accurately reflects a degree of physical, chemical, and faunal variation between these groups. The low abundance and diversity recorded at highly urbanized sites clearly indicate the negative impact of numerous urban pollution sources and upstream contributors on overall ecosystem health and functioning. It is important to note that dramatic differences in the size of individuals were also noted, with upstream study sites in group 1 displaying a tendency towards far smaller individuals of the same species than in group 2. This is likely related to the significant increase in riparian vegetation surrounding these sites, providing edible material for detrital feeders and potentially compensating somewhat for the accumulation of pollutants from upstream in supporting a food web based on periphytic algae (Sharma and Rawat 2009). Although this data was not recorded, adequate facilities and time being unavailable, it was nonetheless significant and should be considered when planning subsequent studies.

While differences between group 1 and group 2 study sites were significant, the radical changes in both composition and abundance at study site 5 compared to the other four study sites emphasize the strong influence exerted by the MuaraAngke Wetland system on polluted water, as expected from the results of previous studies included in the literature. Despite variations in a specific composition, invertebrate communities at each of sites 1-4 were dominated by *Oligochaetae*, a taxon widely known to be remarkably tolerant of high levels of pollution, both organic and anthropogenic, and low oxygen or anoxic stream conditions (Lin and Yo 2008; Azrina et al. 2006). Other groups common at these sites were also found in the 'pollution tolerant' grouping, including *Chironomidae*, *Physidae*and both *Ciroxidae* and *Notonecidae* families of water boatman. Of the other taxa recorded at these sites, the vast majority fell into this grouping, with a small number of moderately tolerant taxa also recorded. Many of these species can survive in-stream conditions with very little oxygen, either because they can gather this from the surface or possess other physiological adaptations, suggesting a low dissolved oxygen content in the stream water. This may be a result primarily of organic pollution precipitating algal blooms. However, the near- total dominance of *Oligochaetae*, exceptionally tolerant to various stressors even amongst this grouping, suggests high inorganic contamination levels were also present. It is important to note that upstream sites recorded almost no species from the 'pollution sensitive' grouping 3, with the sole exception of a single *Atydae* shrimp at site 3, which would appear on all counts to be an anomalous result.

Biofiltration in and around the Muara Angke Wetland

Site 5, identified in the study methodology as being heavily influenced by the ecology of the remnant Muara Angke Wetland, possessed a very different taxonomic composition. As is clearly evidenced from a brief overview of the data presented in Table 1 and is further visualized in figure 1, this study site was dominated almost entirely by taxonomic groups identified as being highly sensitive to pollution-related stressors in the surrounding water. Further analysis of the taxa recorded indicates that this is not simply a result of variations in habitat conditions. Gilled snails (both *Pachychilidae* and *Ampullariidae*) identified previously as the dominant group at this site, unlike their cousins, the Pouch Snails, breathe using an aquatic gill system, and depend on high levels of oxygen in the surrounding water, thus making them highly sensitive to both organic and inorganic contaminants (Voshel 2002). Individuals from at least two distinct species were identified at various points around study site 5, suggesting several different ecological niches are available for this pollution-sensitive group of creatures and decreasing the likelihood that their abundance can be explained simply by an unusual surplus of a particular food source, for example. Similarly, the presence of crustacean taxa *Atydae* and *Parathelphusidae*, both dependent on gill systems to breathe, indicates an improved water quality given the sensitivities of these species to pollutants, suspended particulates, and oxygen (Harmon 2008).

Despite not being recorded in our dataset, as mentioned previously, it is essential to note that each of these taxa at site 5 displayed considerable variations in size and, by extension, age, with both the largest and some of the smallest individuals collected from any study site being identified within the same species. This was most pronounced amongst the *Ampullariidae* taxon of gilled snails but also amongst *Atydae* and the other recorded *Mesogastropoda*. This clearly indicates a wide range of ages amongst individuals and would suggest that significant pollution levels are not accumulating over time within their bodies, allowing them to reach greater ages than taxa at upstream sites, which are unlikely to survive in highly polluted conditions for as long (Mason 2002). Similarly, identifying predatory species from the taxa *Odanatae*, and even the more tolerant *Nepidae* group, indicates that pollution from the surrounding water is not accumulating in sufficient concentrations in the bodies of prey species to be harmful. The increasing concentration of inorganic pollutants through higher levels of the food web has been well documented in a variety of aquatic and terrestrial environments (Farag et al. 1998; Hsu et al. 2007), and such the presence of a more complex food web at this study site can be taken as clear evidence of improved ecosystem health and function.

Finally, it is worth noting that, although the same number of different taxa were identified within the natural wetland area and the adjacent constructed wetland, the abundances of those species were significantly higher at points where the water had also passed through the planted mangroves of the artificial wetland area. This suggests that a combination of natural and constructed wetlands may offer the maximum potential for improving water quality, and thereby ecosystem functioning. Having established the positive influence of the Muara Angke wetland on even the most degraded urban watercourses, follow-up studies should be undertaken to establish the optimal system for wetland-based bio-filtration in this area using a combination of natural and managed or constructed wetland infrastructures.

To summarize, several important conclusions can be drawn from the results discussed above. First of all, despite the methodological limitations of this study, it is clear that the presence of wetland areas has a significant positive impact on the quality of water in the Ciliwung/Angke watershed, even after substantial reductions in the wetland area and the significant degradation of river water from upstream activities and the urban habitat. All indicators identified in the study methodologies (diversity, abundance, size of individuals, and pollution sensitivity) were found to support that the Muara Angke wetland area continues to act as a biological filter for the surrounding megacity. Therefore, it is crucial that existing measures intended to ensure protection for the wetland be reinforced and extended where possible to ensure significant ecosystems services to the surrounding area and further afield are not lost. Whilst quantifying the reach and extent of these benefits is beyond the scope of this preliminary research, it is likely to include significant improvements to local soil and water quality, as well as benefits for the health of the Jakarta Bay ecosystem, and regulating the interaction between Jakarta's urban, terrestrial and marine environments more generally. Interestingly, and perhaps unexpectedly, the results recorded at Sites 3 and 4 suggest that the far less extensive green spaces in these areas may already be delivering some measure of ecosystems services to the Ciliwung river, at the very least increasing the input of biological materials.

ACKNOWLEDGEMENTS

The first author wishes to thank Dr. Robert Francis, project supervisor, whose insight and support was invaluable in undertaking a relatively risky research project in an unknown area and whose guidance in transforming raw data into presentable statistics was particularly key in realizing this research. We appreciate Cendriko Dimas Putra from the Faculty of Biology, Universitas Nasional, Jakarta, and the Jakarta Green Monster, who spent endless hours arranging documents in government institutions, facilitating fieldwork in an incredibly complex urban environment, and helping us collect samples. RUJAK center for Urban Studies, Jakarta, whose ongoing support for all projects about the transformation of Jakarta into a sustainable metropolis remains one of the city's greatest hopes, and whose generosity in allowing the use of their office space and data was invaluable during this time. We acknowledge the government of the Republic of Indonesia, without which this research would not have been possible, particularly the Ministry of Environment and Forestry; Ministry of Research, Technology, and Higher Education, Government of Indonesia.

REFERENCES

Adakole J, Annune P. 2003. Benthic macroinvertebrates as indicators of environmental quality of an urban stream, Zaria, Northern Nigeria, J Aquat Sci 18 (2): 85-92. DOI: 10.4314/jas.v18i2.19948.

Aerts J, Major D, Bowman M, Dircke P, Marfai M, Abidin H, Ward P, Botzen W, Ali F. 2010. Development of water stress index as a tool for the assessment of water stress areas in the Metropolitan Jakarta, 16th Annual International Sustainable Development Research Conference, Hong Kong.

Allen D, Smith K, Darwall W. 2012. The Status and Distribution of Freshwater Biodiversity in Indo-Burma. IUCN, Cambridge.

Anderson N, Sedell J. 1979. Detritus processing by macroinvertebrates in stream ecosystems. Ann Rev Entomol 24 (1): 351-377. DOI: 10.1146/annurev.en.24.010179.002031.

Azrina M, Yap C, Ismail A. R, Ismail A, Tan S. 2006. Anthropogenic impacts on the distribution and biodiversity of benthic macroinvertebrates and water quality of the Langat River, Peninsular Malaysia. Ecotoxicol Environ Saf 64 (3): 337-347. DOI: 10.1016/j.ecoenv.2005.04.003.

Burhanuddin. 1993. A study on mangrove fish at Handeuleum Group and Panaitan Island of Ujung Kulon National Park. Proceeding of Workshop on Mangrove Fisheries and Connections, Ipoh, Malaysia, August 1991.

Cairns J, Prall J. 1993. A History of Biological Monitoring Using Benthos Macroinvertebrates. In: Rosenberg D, Resh V (eds) Freshwater Biomonitoring and Benthic Macroinvertebrates. Chapman and Hall, New York, USA.

Chu H, Chen N, Yeung M, Tam N, Wong Y. 1998. Tidetank system simulating mangrove wetland for removal of nutrients and heavy metals from wastewater. Water Sci Technol 38 (1): 361-368. DOI: 10.2166/wst.1998.0085.

Diaz J, Dowdell S, Stein V. 2013. The effect of anthropogenic inputs on benthic stream invertebrates in a tropical montane stream, Dartmouth. Stud Trop Ecol 23: 52.

Dodson S. 2001. Zooplankton communities of restored depressional wetlands in Wisonsin, USA. Wetlands 21 (2): 292. DOI: 10.1672/0277-5212(2001)021[0292:ZCORDW]2.0.CO;2.

Emere M, Nasiru C. 2008. Macroinvertebrates as indicators of the water quality of an urbanised stream, Kaduna, Nigeria. Nat Sci 7 (1): 1-7.

Farag A, Woodward D, Goldstein J, Brumbaugh W, Meyer J. 1998. Concentrations of metals associated with mining waste in sediments, biofilm, benthic macroinvertebrates, and fish from the Coeur d'Alene River Basin, Idaho. Arch Environ Contam Toxicol 34 (2): 119-127. DOI: 10.1007/s002449900295.

Ferianita-Fachrul M, Salim MR, Said MIM. 2011. The effect of water quality disturbances on macrozoobenthos communities in Jakarta bay waters, Indonesia. The Fifth IOC/WESTPAC International Scientific Symposium - Ocean Sciences at the Dawn of a New Millenium, Seoul, Republic of Korea.

Firaus R. 2013. Benefits of Green Space for Air Quality Improvement and GHG Emissions Reduction in Jakarta. International Institute of Urban Management, Erasmus University, Rotterdam.

Giesen W, Wulffraat S, Zieren M, Sholten L. 2007. Mangrove Guidebook for South East Asia. Wetlands International/FAO Press, Bangkok, Thailand.

Harmon S. 2008. Effects of pollution on freshwater organisms Water Environ Res 82 (10): 1945-2000. DOI: 10.2175/106143008X328851.

Hartnoll R. 1988. Ecoethology of mangroves, behavioural adaption to intertidal. Life 151: 477-489. DOI: 10.1007/978-1-4899-3737-7_30.

Hodkinson I, Jackson J. 2005. Terrestrial and aquatic invertebrates as bioindicators for environmental monitoring, with particular reference to mountain ecosystems. Environ Manag 35 (5): 649-666. DOI: 10.1007/s00267-004-0211-x.

Hsu M, Selveraj K, Agoramoorthy G. 2007. Taiwan's heavy metal pollution threatens terrestrial biota. Environ Poll 143 (2): 327-334. DOI: 10.1016/j.envpol.2005.11.023.

Ligtvoet W, Hughes R, Wulffraat S. 1996. Recreating mangrove forest near Jakarta, Conserving biodiversity in an urban environment. Land Water Intl 84: 8-11.

Lin K, Yo S. 2008. The effect of organic pollution on the abundance of aquatic oligochaetes in an Urban Water Basin, Taiwan. Hydrobiologia 596 (1): 213-223. DOI: 10.1007/s10750-007-9098-x.

Magee P. 1993. Detrital Accumulation and Processing in Wetlands. Fish Wildlife Leaflet 13.3.14. DOI: 10.1007/BF03161297.

Mason C. 2002. Biology of Freshwater Pollution. Pearson, London, UK.

Mastaller M. 1997. Mangroves - The Forgotten Forest Between Land and Sea. Tropical Press, Kuala Lumpur, Malaysia.

Nur Y, Fasi S, Wirjoatmoto N, Han Q. 2001. Towards Wise Coastal Management Practice in a Tropical Megacity - Jakarta. Ocean Coast Manag 44 (5-6): 335-353. DOI: 10.1016/S0964-5691(01)00054-0.

Ogbeibu A, Oribhabor B. 2002. Ecological impact of river impoundment using benthic macroinvertebrates as indicators. Water Res 36 (10): 2427-2436. DOI: 10.1016/S0043-1354(01)00489-4.

Palupi B. 2007. Muara Angke's flora and fauna. The Jakarta Post. http://www.thejakartapost.com/news/2007/08/07/muaraangke039s-floraandfauna.html.

Rosenberg D, Resh V. 1993. Introduction to Freshwater Biomonitoring and Benthic Macroinvertebrates. Chapman and Hall, New York.

Sasekumar A, Chong V, Leh M, D'Cruz R. 1992. Mangroves as a habitat for fish and prawns. Hydrobiologica 247: 195-207. DOI: 10.1007/978-94-017-3288-8_21.

Schläpfer F, Schmid B, Seidl I. 1999. Expert estimates about effects of biodiversity on ecosystem processes and services. Oikos 84: 346-352. DOI: 10.2307/3546733.

Sharma R, Rawat J. 2009. Monitoring of aquatic macroinvertebrates as bioindicator for assessing the health of wetlands: A Case Study in the Central Himalayas, India. Ecol Indicat 9 (1): 118-128. DOI: 10.1016/j.ecolind.2008.02.004.

Supartono J, Haluan MFA, Sondita, Manuwoto. 2016. Jakarta North Coast development impact on fishery activities. Asian J Sci Res 9 (1): 13-23. DOI: 10.3923/ajsr.2016.13.23.

Swari A, Arfiati D, Marsoedi. 2014. Type and abundance of macrozoobenthos as a pollution indicator in Ken Dedes River at Singosari Subdistrict, Malang Regency, East Java, Indonesia. Intl J Engin Sci 3: 43.

Tilley D, Badrinarayanan H, Rosati R, Son J. 2002. Constructed wetlands as recirculation filters in largescale shrimp aquaculture. Aquacult Eng 26 (2): 81-109. DOI: 10.1016/S0144-8609(02)00010-9.

Voshell J. 2002. A Guide to Common Freshwater Invertebrates of North America. The McDonald & Woodward Publishing Company, Blacksburg, VA.

Warwick P. 1993. A new method for detecting pollution effects on marine macrobenthic communities. Mar Biol 92 (4): 557-562.

Weston D. 1990. Quantitative examination of macrozoobenthic community changes along an organic enrichment gradient. Mar Ecol Prog Ser 61 (3): 233-244. DOI: 10.3354/meps061233.

Wong Y, Tam N, Chen G, Ma H. 1997. Response of *Aegiceras cornicaltum* to synthetic sewage under simulated tidal conditions. Hydrobiologia 352: 89. DOI: 10.1007/978-94-011-5234-1_10.

Ye Y, Tam N, Wong Y. 2001. Livestock wastewater treatment by a mangrove potcultivation system and the effect of salinity on the nutrient removal efficiency. Mar Pollut Bull 42 (6): 512-520. DOI: 10.1016/S0025-326X(00)00196-X.

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 74-78

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070203

Analysis of importance value index of unlogged and logged peat swamp forest in Nenasi Forest Reserve, Peninsular Malaysia

MOHD HASMADI ISMAIL[1,♥], MOHD FADLI AHMAD FUAD[2], PAKHRIAZAD HASSAN ZAKI[3], NOOR JANATUN NAIM JEMALI[4]
[1,3]Faculty of Forestry, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia. Tel: +60-389467220, ♥email: mhasmadi@upm.edu.my
[2]Department of Forestry Peninsular Malaysia, Jalan Sultan Salahuddin, 50660 Kuala Lumpur, Malaysia.
[4]Faculty of Earth Science, Universiti Malaysia Kelantan, 17600 Jeli, Kelantan, Malaysia.

Manuscript received: 10 July 2017. Revision accepted: 20 August 2017.

Abstract. *Ismail MH, Fuad MFA, Zaki PH, Jemali NJN. 2017. Analysis of importance value index of unlogged and logged peat swamp forest in Nenasi Forest Reserve, Peninsular Malaysia. Bonorowo Wetlands 7: 74-78.* Peat swamp forests are highly significant globally, both for their diverse and threatened species, and represent unique wetland ecosystems. Apart from its critical role in providing habitat for wildlife, the tropical peat swamp forest also acts as a gene bank that harbors potentially useful varieties of plant species. The composition of trees of the peat swamp forest in Nenasi Pahang State, Peninsular Malaysia, were investigated, especially in un-logged and logged-over forests. The objectives of this study are two folds; (i) to identify and compare the dominant tree species in the two types of forests and (ii) to calculate the importance value index (IVI) of the vegetation in the study areas. Two plots of 50m by 20m were established in different forest types. This quadrate was subdivided at each site using 25m by 20m and 5m by 5m, respectively. The results showed that the unlogged peat swamp forest is higher in tree species compared to logged peat swamp forest. There were 10 species distributed among 9 families in the unlogged forest and 7 species in 7 families in the logged forest. The most dominant species identified in unlogged peat swamp forests were *Litsea sp., Syzygium sp.,* and *Santiria laevigata.* The IVI for these species were 71.21, 51.13, and 42.49. In logged peat swamp forests, the dominant species are *Shorea platycarpa, Pometia pinnata,* and *Xylopia fusca.* The IVI of these species were 87.38, 52.66, and 47.55, respectively. The study concludes that about 40 percent of the tree composition in the logged peat swamp forests has declined compared to unlogged peat swamp forests.

Keywords: Peat swamp forest, tree composition, importance value index

INTRODUCTION

Wetlands or peatlands are habitats where the water table is at or near the land surface or where the land is covered by shallow water for a sufficient length of time to cause anaerobic conditions within the root zone of plants (Barnes et al., 2002). In recent years, the problem of ecological degradation and destruction of peat swamp forests and wetlands has attracted scientific communities worldwide. Current research has shown this ecosystem its significance not just as a global carbon store, but its value for biodiversity remains poorly understood (Posa et al., 2011). The forest's biodiversity is poorly understood, and its importance is underappreciated. Vast areas of peat swamp forest have been degraded and disappearing due to logging, fire, and conversion to agriculture and industry.

Presently about 50% of the peat swamp forests in Southeast Asia have been cleared and drained for agriculture (Hooijer et al. 2010). The peat-swamp forests in Malaysia are fragile and sensitive to development. Nowadays, some parts of peat swamp forests in Malaysia are being extensively cleared for agriculture and other development projects (Gasim et al., 2007). Activities such as logging, plantation, and development in the long term will lead to deterioration in the quality of peat swamp biodiversity and forest ecosystems. In Malaysia, 44% of remnant peat swamp forests are moderately or severely disturbed, with the highest proportion in Sarawak (54%). In Peninsular Malaysia, only 18% of peat swamp forest appears to be moderately or severely disturbed (Wetland International-Malaysia 2010). Due to various treats on the peat swamp forest, study on the compositions, conservation status, structural and environmental characteristics of the plant communities in these forests is required.

In general, the importance value index (IVI) measures how dominant a species is in a given forest area. The importance value index (IVI) of tree species was determined as the sum of relative frequency, relative density, and relative dominance (Curtis and McIntosh, 1950). Each of these values is expressed as a percent and ranges from 0 to 100. Due to the massive degraded peat swamp forest, preserving the forest remnants is essential, especially in conservation or reserve areas. Consequently, it is necessary to study the vegetation to elucidate the characteristics and regeneration processes of the remaining peat swamp forest. Therefore, the objectives of this study are two folds (i) to identify the presence/ absence of dominant tree species in unlogged and logged peat swamp forest, and (ii) to calculate the importance value index (IVI) and species diversity in both sites.

MATERIALS AND METHODS

Description of the study area

The peat swamp forests in the southeast Pahang area comprise four forest reserves; Pekan, Nenasi, Kedondong, and Resak cover 87,045 ha (Figure 1). The peat swamp forests harbor a unique flora and fauna, provide benefits and services of national interest, and support the livelihood of the aborigines (*Orang Asli*) communities (Mohd Azmi 2009). All these resources are significant not only for the ecological functioning of the forest but also for future biodiversity conservation and sustainable development of the whole area.

Specifically, the study area is located in two different forest conditions (unlogged and logged forest) but lies in the same locality and similar topographic features. The elevation of the study area is about 25m to 27m above sea level. The site's distance is about 25 km to Kuala Rompin and 55 km to Pekan, Pahang. Nenasi Forest Reserve has an area of 20,546 ha, between Bebar and Merchong Rivers.

Plot establishment and data collection

Two plots were established in unlogged and logged forests. At each site, 50m by 20m quadrate (Q1) was established, and within this quadrate, 25m by 20m (Q2) and 5m by 5m (Q3) plots were established. As each subplot's corner (border) markers, ironwood poles of 5cm x 5cm x 130cm were used as each subplot's corner (border). Every tree in the quadrate was tagged. The quadrate and its subdivision are shown in Figure 2. The peat swamp forest in plot number 1 is un-logged, while plot number 2 is considered the logged forest logged in 2006. A Garmin GPS was used to locate the coordinates of the plots, and other tools such as diameter tape for dbh measurement, Haga altimeter (tree height), densitometer (percentage canopy cover), and magnetic compass (plot establishment) were used during data collection.

Many ways have been used to classify vegetation. Some are based on vegetation physiognomy, vegetation structure, or environmental factors. Some of these approaches are useful at vast scales. For this study classification of trees was based on the tree classification by the Forestry Department of Peninsular Malaysia for Pre-Felling or Post-Felling inventories (Table 1).

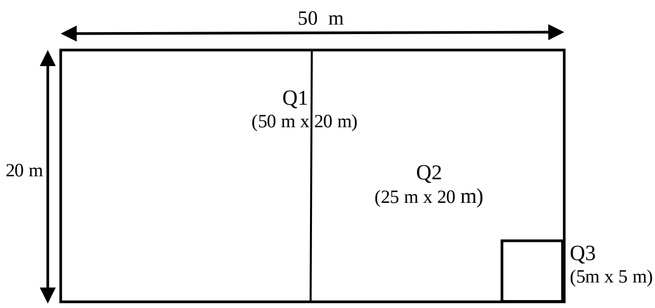


Figure 2. The layout of the quadrate (50 m x 20 m)

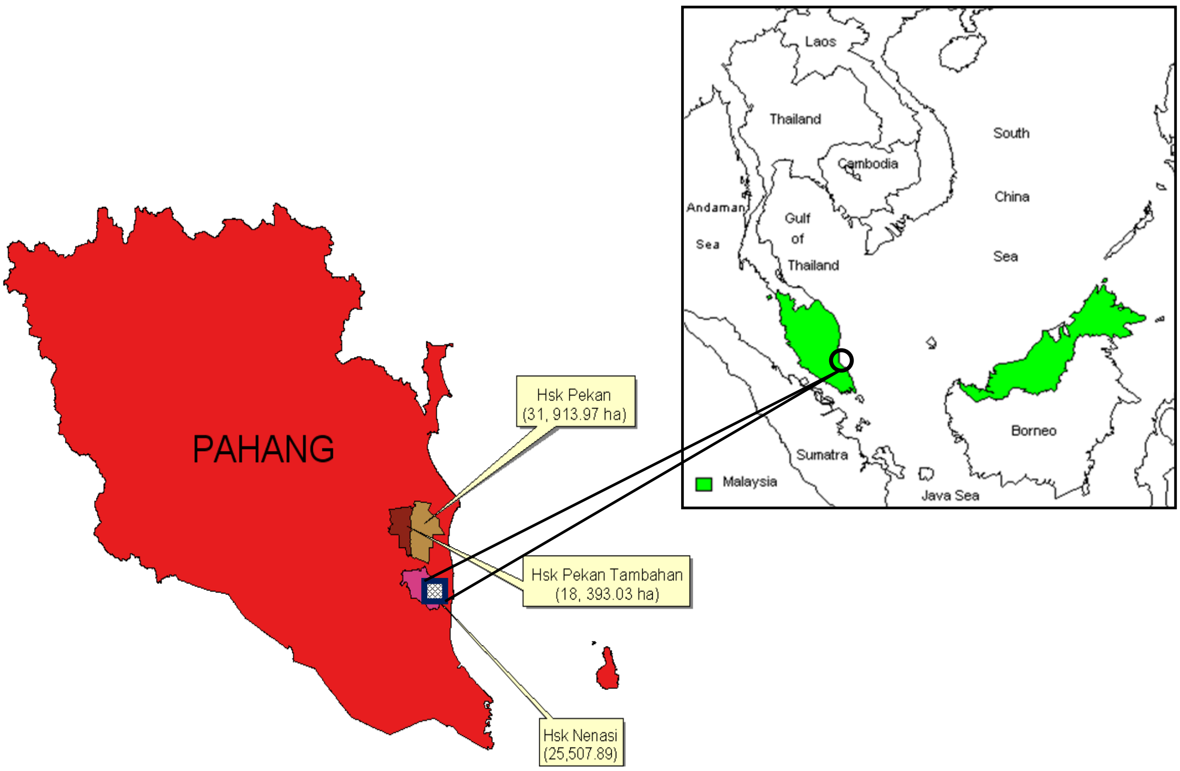


Figure 1. A location of the study area in Pahang state, Peninsular Malaysia

Table 1. Tree classification used in the study

Quadrate	Vegetation layers	dbh (cm)
Q1 (50 m X 20 m)	Big and small tree	> 30.0
Q2 (25 m X 20 m)	Big and small poles	5.0 - 29.9
Q3 (5 m X 5 m)	Sapling	1.5 - 4.9

Data analysis

The Importance Value Index (IVI) of Cottam and Curtis (1956), Cox (1967), and Mueller-Dombois and Ellenberg (1974) was used to describe and compare the species dominant in the plots. The species with the highest IV index were considered the most "important" in a plot. This index is used to determine the overall importance of each species in the community structure. The IVI of the taxon of each plot is defined as the sum of its relative density and relative dominance, which describes the dominance of a species in the whole plot. The IVI is determined for both plots by adding its relative frequency. The relationship, RD+RCC+RF, shows the importance value (IVI) for each species, and hence its value varies from 0 to 300. The species having the highest IVI is considered dominant in the community (Arshad et al., 2002; Noraimy et al., 2014). The following equations are used:

$$\text{Importance value (IVI)} = RD + RCC + RF$$

$$\text{Relative density (RD)} = \frac{\text{The density of a species}}{\text{Total density of all species}} \times 100$$

$$\text{Relative canopy cover (RCC)} = \frac{\text{Canopy cover of a species}}{\text{Total cover of all species}} \times 100$$

$$\text{Relative frequency (RF)} = \frac{\text{Frequency value of the species}}{\text{Sum of frequency of all species studied}} \times 100$$

RESULTS AND DISCUSSION

Tree analysis

The total numbers of families and species were determined for each. Overall, 39 tree species belonging to 12 families were identified. Total trees, species, families, total number of individuals per unit plot, and canopy height were greater in unlogged peat swamp forest (Table 2 and Table 3). A total of 27 species has been recorded in unlogged peat swamp forest and 12 in logged forest. Species *Syzygium sp.* was dominated most in an unlogged plot with 5 trees. The second dominant species was *Myristica sp.* with only 3 trees. In the logged forest, the dominant species recorded were *Shorea platycarpa* (3), *Pometia pinnata* (2), *and Goniothalamus sp.* (2). Approximately 60% (unlogged) were classified under big and small trees. While only 20% for the logged forest. The largest group of trees (in the unlogged forest) was big and small, but in logged, a group of big and small poles represents 60%. A huge impact occurred on the several tree presence in both sites. The results showed that the massive trees decline was at the logged forest, in which only 12 trees were present compared to unlogged peat swamp forest by 27 trees. The impact of tree harvesting about 6 years ago has caused a significant decline in the presence of the tree (about 40% different between the two plots).

The distribution of dbh classes and several trees for both plots and each quadrate are shown in Figure 5. In unlogged peat swamp forests, the tree stand is intact and shows good composition for each dbh range, and this is typical of all types of forest areas. However, in logged forest, due to the previous harvesting, the distributions are more toward the dbh range between 5.0 cm - 29.9 cm, respectively.

Peat swamp forests contain several valuable timber species and sometimes at high densities. However, this forest has been intensively exploited. There is very little information on the long-term effects of logging on flora and fauna of peat swamp forests (Hadisuparto, 1996). For instance, logging operation with a mechanization system leads to a lack of regeneration of ramin trees (Gonystylus bancanus), the most valuable timber species in peat swamps. The species is now listed as vulnerable on the IUCN Red List. Rashid and Ibrahim (1994) reported that the logging operation has led to more intensive extraction and more significant damage to the residual forest up to 50%.

The most dominant species found in unlogged peat swamp forests were *Litsea sp.*, *Syzygium sp.*, and *Santiria laevigata.* The importance value index (IVI) for these species were 71.21, 51.13, and 42.49, respectively. In logged peat swamp forest, the dominant species are *Shorea platycarpa*, followed by *Pometia pinnata*, *Xylopia fusca*, and *Syzygium sp.* The IVI of these species was 87.38, 52.66, and 47.55. The contribution of the dominant species (relative density) such as *Litsea sp.* and *Xylopia fusca* to the area (unlogged peat swamp forest) were 25.85%, 16.13 %, and 12.9 %. In logged peat swamp forest, the dominant species' relative density was 25% for *Shorea platycarpa* and equal for *Pometia pinnata, Xylopia fusca,* and *Syzygium sp*. (16.67%), and also similar for other species such as *Gluta velutia, Macaranga pruinose*, and *Durio carinatus* (8.33%).

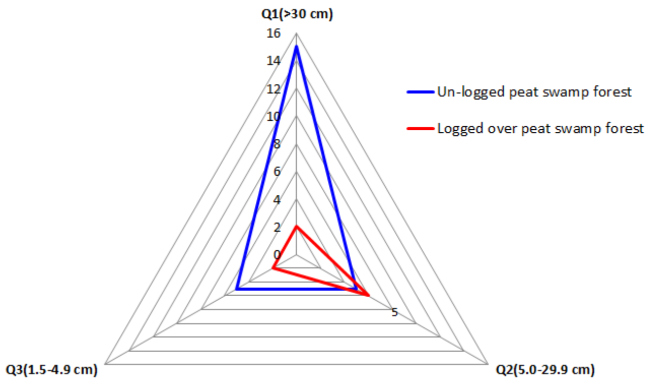


Figure 5. Comparison of the total tree at both site and vegetation layer

Table 2. List of species in unlogged peat swamp forest

Quadrate	Species	Family	dbh (cm)	Height (m)	Crown size (m)	Vegetation Layer
Q1 50m x 20m	*Dacryodes macrocarpa*	Burseraceae	42	6	5	Big and small trees
	Alstonia spatulata	Apocynaceae	31	7	3.6	
	Syzygium zeylenicum	Myrtaceae	36	5.5	3.5	
	Koompasia malaccensis	Leguminoceae	40	15	4.5	
	Syzygium zeylenicum	Myrtaceae	38	5	3.8	
	Instia bijuga	Leguminoceae	48	11	5.0	
	Artocarpus kemando	Moraceae	43	15	8.2	
	Litsea costata	Lauraceae	44	7.6	5.7	
	Santria laervigata	Burseraceae	36	5.6	3.8	
	Xylopia malayana	Annonaceae	39.4	8.3	4.8	
	Alstonia spatulata	Apocynaceae	38	14	5.6	
	Campnosperma coriaceum	Anacardiaceae	45.9	13	6.3	
	Horsfielddia crassifolia	Myristicaceae	36	5.6	4.8	
	Campnosperma coriaceum.	Anacardiaceae	42.8	4.8	5.7	
	Myristica lowiana	Myristicaceae	45.9	7.2	8.5	
Q2 25m x 20m	*Myristica gigantea*	Myristicaceae	12.2	5.2	2.6	Big and small poles
	Syzygium kiahii	Myrtaceae	18.7	6.3	4.8	
	Syzygium napiforme	Myrtaceae	21	5.8	7.0	
	Santiria rubiginosa	Burseraceae	11	2.4	2.4	
	Litsea grandis	Lauraceae	19.2	8.0	5.6	
Q3 5m x 5m	*Goniothalamus sp.*	Annonaceae	2	0.8	-	Saplings
	Myristica gigantea	Myristicaceae	0.3	0.4	-	
	Syzygium lineatum	Myrtaceae	0.2	0.2	-	
	Instia palembanica	Fabaceae	0.2	0.7	-	
	Litsea grandis	Lauraceae	0.2	0.8	-	
	Instia bijuga	Fabaceae	0.4	0.7	-	
	Litsea teysmannii	Lauraceae	0.8	1.0	-	
	Total tree: 27					

Table 3. List of species in logged peat swamp forest

Quadrate	Species	Family	dbh (cm)	Height (m)	Crown size (m)	Vegetation Layer
Q1 50m x 20m	*Pometia pinnata*	Sapindaceae	31.4	4	6	Big and small trees
	Durio carinatus	Moraceae	38.2	6	10.6	
Q2 25m x 20m	*Goniothalamus malayanaus*	Annonaceae	22.5	9	3.5	Big and small poles
	Shorea platycarpa	Dipterocarpaceae	23	10	4	
	Goniothalamus sp.	Annonaceae	9.5	8	2.8	
	Gluta velutina	Anacardiaceae	10.1	9	2.3	
	Shorea platycarpa	Dipterocarpaceae	10.7	7	4.5	
	Pometia pinnata	Sapindaceae	25.6	11	2.7	
	Shorea platycarpa	Dipterocarpaceae	1.7	7.6	8	
Q3 5m x 5m	*Macaranga pruinosa*	Euphorbiaceae	0.9	1	-	Saplings
	Syzygium kiahii	Myrtaceae	0.7	1.5	-	
	Syzygium napiforme	Myrtaceae	2.4	0.8	-	
	Total tree: 12					

Unlogged peat swamp forest is significantly rich in plant species (10 species compared to the logged forest is 7 species). A study by Bruenig and Droste (1995) indicates that selective logging causes changes in forest structure and composition. This may explain that logging activity has opened the forest gap in the logged forest. This is vulnerable to more destruction by the fire where the soil substrate is highly flammable when dry (Langner et al., 2007). Consequently, according to Yeager et al. (2003), the effects of fire on peat swamp forest vegetation are similar to those in other forest types, where the burned forest has lower canopy cover, decreased species richness, and reduced tree and sapling density compared with unburned forest.

Table 4. Quantitative analysis for IVI (Unlogged peat swamp forest)

Species	RD (%)	RCC (%)	RF (%)	IVI
Santiria laevigata	12.9	16.69	12.9	42.49
Syzygium napiforme	16.13	18.87	16.13	51.13
Koompasia malaccensis	3.22	4.44	3.22	10.88
Instia bijuga	9.67	4.94	9.68	24.29
Artocarpus kemando	3.22	8.10	3.22	14.54
Litsea grandis	25.85	19.56	25.8	71.21
Xylopia fusca	9.67	4.74	9.68	24.13
Alstonia pneumatophora	6.45	9.09	6.47	22.01
Campnosperma coriaceum	3.22	6.22	3.22	12.66
Gymnacranthera euginiifolia	9.67	7.31	9.68	26.66
Total	**100**	**100**	**100**	**300**

Note: RD = relative density, RCC= relative canopy cover, RF= relative frequency, IVI = important value index

Table 5. Quantitative analysis for IVI (Logged peat swamp forest)

Species	RD (%)	RCC (%)	RF (%)	IVI
Pometia pinnata	16.67	19.59	16.4	52.66
Durio carinatus	8.33	23.88	8.33	40.54
Xylopia fusca	16.67	14.18	16.7	47.55
Shorea platycarpa	25	37.17	25.21	87.38
Gluta velutina	8.33	5.18	8.33	21.84
Macaranga pruinosa	8.33	0	8.33	16.96
Syzygium gracile	16.67	0	16.4	33.07
Total	**100**	**100**	**100**	**300**

Note: RD = relative density, RCC= relative canopy cover, RF= relative frequency, IVI = importance value index

Conclusion

Previously, the vast majority of the peat swamp areas were covered by forest. Still, logging and large-scale conversion of peat forests to agricultural land have occurred within the last few decades. Because peat swamp forests initially supported high densities of tree species, biodiversity losses have occurred through timber extraction. This study showed that the unlogged peat swamp forest in Nenasi, Pahang, has a more diverse population than the logged peat swamp forest in the same locality. Differences in several individual trees, species, families, canopy cover and dbh can be attributed to harvesting. The number of trees, species, families, individuals per unit plot, and canopy height were more significant in the un-logged peat swamp forest. The impact of tree harvesting has caused a significant decline in the presence of the tree. Research and possible restoration are urgently needed into the biodiversity and ecology of Malaysia's remaining peat swamp forests. The study concludes that about 40 percent of the tree composition in the logged peat swamp forests has declined compared to unlogged peat swamp forests. Conservation and rehabilitation of the peat swamp forest will be crucial in preserving biodiversity.

REFERENCES

Arshad M, Uddin S, Reahman Rao A. 2002. Phytosociological assessment of natural reserve of National Park Lalsuhanna, Punjab, Pakistan. Asian J Plant Sci 1: 174-175. DOI: 10.3923/ajps.2002.174.175.

Barnes K, Ellery W, Kindness A. 2002. A preliminary analysis of water chemistry of biodiversity of a 1 ha old world rainforest in Brunei, Borneo. Biodiv Conserv 13: 2067-2088. DOI: 10.1023/B:BIOC.0000040001.72686.e8.

Bruenig EF, Droste HJ. 1995. Structure, dynamics, and management of rainforests on nutrient-deficient soils in Sarawak. In Primack RB, Lovejoy TE (eds) Ecology, Conservation, and Management of Southeast Asian Rainforests. Yale University Press, USA.

Cottam G, Curtis JT. 1956. The use of distance measures in phytosociological sampling. Ecology 37 (3): 451-460. DOI: 10.2307/1930167.

Cox GW. 1967. Laboratory Manual of General Ecology. Wm. C. Brown Co. Publisher, Iowa.

Curtis JT, McIntosh RP. 1950. The interrelations of certain analytic and synthetic phytosociological characters. Ecology 31 (3): 434-455

Gasim MB, Ismail BS, Toriman E, Mir S I, Tan CC. 2007. A physico-chemical assessment of the Bebar River Pahang, Malaysia. Global J Environ Res 1 (1): 7-11.

Hadisuparto H. 1996. The effects of timber harvesting and forest conversion on peat swamp forest dynamics and environment in west Kalimantan. In: Edwards DS, Booth WE, Choy SC (eds) Tropical Rainforest Research, Current Issues. Kluwer, The Nederland. DOI: 10.1007/978-94-009-1685-2_40.

Hooijer A, Page S, Canadell JG, Silvius M, Kwadijk J, Wosten H, Jauhiainen J. 2010. Current and future CO_2 emissions from drained peatlands in Southeast Asia. Biogeosciences 7 (5): 1505-1514. DOI: 10.5194/bg-7-1505-2010.

Kunwar R M, Sharma SP. 2004. Quantitative analysis of tree species in two community forests of Dolpa district, mid-west Nepal. Himalayan J Sci 2 (3): 23-28. DOI: 10.3126/hjs.v2i3.226.

Langner A, Miettinen J, Siegert F. 2007. Land cover change 2002-2005 in Borneo and the role of fire derived from MODIS imagery. Glob Change Biol 13 (11): 2329-2340. DOI: 10.1111/j.1365-2486.2007.01442.x.

Mohd Azmi MI, Cullen R, Awang Noor AG. 2009. The existence value of peat swamp forest in Peninsular Malaysia. NZARES Conference, Tahuna Conference Centre-Nelson. New Zealand, 7-28 August, 2009.

Mueller-Dombois D, Ellenberg H. 1974. Aims and Methods of Vegetation Ecology. John Wiley and Sons, New York.

Noraimy R, Mohd Hasmadi I, Norizah K, Pakhriazad HZ. 2014. Effect of skid trails on the regeneration of commercial tree species at Balah Forest Reserve, Kelantan, Malaysia. Biodiversitas 15 (2): 239-243. DOI: 10.13057/biodiv/d150218.

Noraimy R, Norizah K, Mohd Hasmadi I. 2014. Examining the rate of vegetation diversity under abandoned skid trails in Peninsular Malaysia forest. J Agric Crop Res 2 (8): 165-172.

Posa MRC, Wijedasa LS, Corlett RT. 2011. Biodiversity and conservation of tropical peat swamp forests. BioScience 61 (1): 49-57. DOI: 10.1525/bio.2011.61.1.10.

Rashid MFA, Ibrahim S. 1994. Skyline as an alternative logging method in peat swamp forest in Peninsular Malaysia. In: Ibrahim WRW, Appanah S, Rashid MFA (eds) Proceedings of the Symposium on Harvesting and Silviculture for Sustainable Forestry in the Tropics. FRIM, Kepong, Malaysia.

Wetlands International. 2010. A quick scan of peatlands in Malaysia. Wetlands International-Malaysia, Petaling Jaya, Malaysia.

Yeager CP, Marshall AJ, Stickler CM, Chapman CA. 2003. Effects of fires on peat swamp and lowland dipterocarp forests in Kalimantan, Indonesia. Trop Biodiv 8 (1): 121-138.

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 79-82

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070204

Short Communication: The potential of Sulfate Reducing Bacteria of ex-coal mine sediment pond as sulfate reducing agents of acid land in Samarinda, Indonesia

EKO KUSUMAWATI[1,♥], SUDRAJAT[2], IKA PURNAMASARI[3], BINA CRISTYANTI PANGGABEAN[1], MAIDA APRIYANTI[1]

[1]Laboratory of Microbiology and Molecular Genetics, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Mulawarman. Samarinda 75123, East Kalimantan, Indonesia. ♥email: eko.kusumawati11@gmail.com

[2] Laboratory of Animal Anatomy and Microtechnique, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Mulawarman. Samarinda 75123, East Kalimantan, Indonesia

[3]Laboratory of Economics and Business, Department of Mathematics, Faculty of Mathematics and Natural Sciences, Universitas Mulawarman. Samarinda 75123, East Kalimantan, Indonesia

Manuscript received: 20 May 2017. Revision accepted: 9 September 2017.

Abstract. *Kusumawati E, Sudrajat, Purnamasari I, Panggabean BC, Apriyanti M. 2017. Short Communication: The potential of Sulfate Reducing Bacteria of ex-coal mine sediment pond as sulfate-reducing agents of acid land in Samarinda, Indonesia. Bonorowo Wetlands 7: 79-82.* The study aims to determine the effect of pH medium on the growth of sulfate-reducing bacteria taken from the e-coal mine sediment pond and determine its potential as a reducing sulfate agent of acid ex-coal mine land in Samarinda East Kalimantan, Indonesia. This study used six SRB isolated from an ex-coal mine sediment pond in Samarinda. The SRB potency test in reducing sulfate was conducted by growing the SRB on Postgate liquid medium at different pHs of 2, 4, and 6 by adding acid soils on each treatment. The results showed that sulfate reducing bacteria isolated from ex-coal mine sediment pond in Samarinda, i.e., sp.1 (*Desulfococcus* sp.), sp.2 (*Desulfotomaculum* sp.), sp.3 (*Desulfobacter* sp.), sp.4 (*Desulfobulbus* sp.), sp.5 (*Desulfobacterium* sp.) and sp.6 (*Desulfotomaculum* sp.) had potential as sulfate reducing agent of acid land. The efficiency of sulfate reduction was 89%, 91%, and 91% in the pH of 2, 4, and 6, respectively. This indicated that the highest sulfate reduction is in the medium with pH 4 and 6. In addition, sp.5 (*Desulfobacterium* sp.) growing on medium at pH 4 had the best sulfate reduction efficiency (93%) compared with other SRB isolates.

Keywords: pH of the medium, sulfate reducing bacteria, acid land

INTRODUCTION

Mining activities such as coal mining can provide economic benefits and cause environmental and soil ecosystems damages (Tala'ohu et al., 1995). One element of the causes of the impact is the generated waste as a byproduct of the remnant of the mining and processing, which are often in large volume and various kinds. United Nations Environment Programme (UNEP) classifies the impacts of mining activities, including the destruction of habitat and biodiversity at the mine site, the landscape change or loss of land use, and B3 waste and chemicals (Fahruddin 2010). The most severe mining activity problem is the phenomenon of acid mine drainage (AMD) or acid rock drainage (ARD) due to the oxidation of sulfur minerals. This will give a series of interrelated effects, namely decreasing pH, disturbing the availability and the balance of soil nutrients, and increasing the solubility of micronutrients which generally is a metal element (Havlin et al. 1999).

One environmentally friendly method is bioremediation, namely, a process to restore the environment using microorganisms as contaminants eliminating. A group of microbes that can improve the quality of post-mine land is sulfate-reducing bacteria (SRB). In metabolic activities, SRB can convert sulfate to H2S. This gas will immediately bind to the metals found in many post-mining land and precipitate in the form of metal sulfides reductive (Hards and Higgins 2004). However, the main problem often encountered in applying microorganisms for bioremediation is the decrease or the loss of potential microbial. To improve the effectiveness of microorganisms in bioremediation, the following two strategies can be done. First, bio-stimulation is a technique to add specific nutrients to stimulate the activity of local (indigenous) microbes. Atlas and Bartha (1992) stated that the bio-simulation process has successfully controlled the oil spills in water and the contamination of hydrocarbons (PAHs) in the soil. Lieberg and Cutright (1999) stated that nutrients often used in this technique are phosphorus and nitrogen. Second, bio-augmentation is a technique to introduce specific microbes in the remediated area. In addition, environmental influences such as pH, temperature, and soil moisture were also very influential in determining the success of the bioremediation process.

Based on the introduction, the problem in this research is how the influence of pH of the medium on the sulfate reduction activity of sulfate reducing bacteria isolated from ex-coal mine sediment pond in Samarinda on acid land (TMT) on the location of the ex-coal mine is. The purpose of this study was to determine the effect of medium pH on the potential sulfate reducing bacteria isolated from ex-coal mine sediment pond in Samarinda as a sulfate reducing agent in acid land (TMT) on the location of the ex-coal mining is. The obtained information will develop bacteria as a potential environmentally friendly bioremediation agent.

MATERIALS AND METHODS

Sampling

Sulfate-reducing bacteria (SRB) were isolated from an ex-coal mine sediment pond in Samarinda, East Kalimantan. Bacteria were isolated on Postgate liquid medium (Atlas and Park, 1993) containing (g/l) Na lactate (3.5), $Mg.SO_4$ (2.0), NH_4Cl (0.2), KH_2PO_4 (0.5), $FeSO_4.7H_2O$ (0.5) and pH 4 and sterilized at 121^oC with 1 atmosphere pressure for 15 minutes. SRB growth was characterized by the emergence of colonies in dark brown to black at the bottom of the tube.

Sulfate-reducing bacteria activity test on liquid media

Six SRB isolates result from the isolation of bacteria from ex-coal mine land in Samarinda. The isolates comprised of a mixture of six isolates based on the early identification, i.e., sp.1 (*Desulfococcus* sp.), sp.2 (*Desulfotomaculum* sp.), sp.3 (*Desulfobacter* sp.), sp.4 (*Desulfobulbus* sp.), sp.5 (*Desulfobacterium* sp.) and sp.6 (*Desulfotomaculum* sp.). Each pure isolate SRB (1 ml) was inoculated into a liquid Postgate medium enriched with 2 N sulfuric acid solution as much as 5% (v/v). The cultures were incubated in 25 ml tubes filled up to total volume. Each SRB isolate was used to treat with a consortium of all bacterial treatments. Experiments were carried out in a completely randomized design with three replications. The sulfate measurements were done at the beginning and twenty days of treatment. The control group was treated with Postgate B medium enriched with 2 N sulfuric acid solution as much as 5% (v/v) without SRB inoculation. The efficiency of each treatment was calculated using the formula stated by Widyati (2006), namely:

<u>(Initial sulfate concentration) - (final sulfate concentration) x 100%</u>
(Initial concentration)

Sulfate-reducing bacteria activity test against acid ex-coal mine soil with pH of 2, 4, 6

The composition of bacteria used in the trial experiments test was similar to SRB in Postgate liquid media. SRB isolates used in this study are maintained in a Postgate liquid medium. Each of these pure SRB isolates (1 ml) was inoculated into a liquid Postgate media with different pH, i.e., pH 2, 4, and 6. Each media was previously filled with 5 grams of acid ex-coal mine soils. The cultures were incubated in a 10 ml screw-capped tube filled to the brim. Each SRB isolate was used to treat with a consortium of all bacterial treatments. Experiments were carried out in a completely randomized design with three replications. The measurements of pH and sulfate were done at the beginning of treatment and after twenty days of treatment. The control group was treated with Postgate B medium enriched with 2 N sulfuric acid solution as much as 5% (v/v) without SRB inoculation. The efficiency of each treatment was calculated using the formula stated by Widyati (2006), namely:

<u>(Initial sulfate concentration) - (final sulfate concentration) x 100%</u>
(Initial concentration)

RESULTS AND DISCUSSION

SRB activity test results on the liquid Postgate media can be seen in Table 1. Table 1 shows that the treatment not inoculated with SRB sulfate concentrations only slightly decreased while the sulfate in the treatment inoculated with SRB sulfate concentrations varied on the 20th day after incubation. When it was calculated with the formula of efficiency of Widyati (2006, 2007), the lowest efficiency value was obtained at sp.1 and sp.3 with the amount of 74%, while the highest efficiency value was obtained at sp.5 with 91%, but on the controls which were not inoculated with SRB, the efficiency only decreased by 1% within 20 days. In this study, the sulfate concentration decreased because the SRB can use sulfate as an electron acceptor for metabolic activities (Doshi 2006). Because the sulfur accepts electrons, it will change to sulfide so that its concentration in the culture decreases.

SRB activity test was also conducted on samples of coal mine acid soil to measure the content of sulfate in the soil using ex-coal mine soil with varying pH, namely pH 2, 4, and 6. The measurement results on sulfate content changes on ex-coal mine land by SRB activity can be seen in Table 2.

Data in Table 2 show that sulfate reduction occurred in all treatments. The efficiency test on sulfate reduction is done by SRB activity in reducing ex-coal mine sulfuric acid soil after 20 days of treatment. It results that it is more significant at pH 4 and 6 than at pH 2. The sulfate reduction efficiencies ranging from pH 2, pH 4, pH 6 are respectively 89%, 91%, and 91%. This shows that sulfate reduction is most significant at pH 4 and 6. Table 1 also shows that the controls remain sulfate reduction occurred when the control contained only sterile media and acid soil, coal mines without getting additional isolates SRB. This indicates that in acid soil samples, there is also the content of indigenous sulfate-reducing bacteria in those samples. It was alleged that the sulfate-reducing bacteria are more efficient to reduce sulfate at pH 2 than at pH 4 and pH 6, so the efficiency of the control sulfate reduction treatment at pH 2 is 92%, which is higher than at pH 4 (87%) and pH 6 (91%).

Table 1. The concentration of sulfate at the beginning and the end of the test was done by the activity of sulfate to reduce bacteria in a liquid medium Postgate

Treatment	Initial sulfate concentration in the medium (mg/L)	Final sulfate concentration (mg/L)	Reduction of sulfate (mg/L)	Efficiency (%)
Control	128,668	127,405	1,263	1%
Sp.1 (*Desulfococcus sp.*)	127,835	33,450	94,385	74%
Sp.2 (*Desulfotomaculum sp.*)	128,630	31,479	97,151	76%
Sp.3 (*Desulfobacter sp.*)	126,878	32,704	94,174	74%
Sp.4 (*Desulfobulbus sp.*)	129,365	32,944	96,421	75%
Sp.5 (*Desulfobacterium sp.*)	128,043	12,023	116,020	91%
Sp.6 (*Desulfotomaculum sp.*)	127,772	14,092	113,680	89%
Consortium	129,507	32,129	97,377	75%

Table 2. Initial to final sulfate concentration on sulfate reducing bacteria activity test against coal mine acid soil with pH medium using different liquid Postgate medium

pH	Treatment	Initial sulfate concentration in the medium (mg/L)	Final sulfate concentration (mg/L)	Reduction of sulfate (mg/L)	Efficiency (%)
2	Control	202,437	15,561	186,876	92
	Sp.1 (*Desulfococcus sp.*)	220,080	24,090	195,990	89
	Sp.2 (*Desulfotomaculum sp.*)	212,917	23,106	189,810	89
	Sp.3 (*Desulfobacter sp.*)	234,273	23,726	210,547	90
	Sp.4 (*Desulfobulbus sp.*)	216,287	21,489	194,798	90
	Sp.5 (*Desulfobacterium sp.*)	215,313	29,115	186,199	86
	Sp.6 (*Desulfotomaculum sp.*)	222,023	28,318	193,705	87
	Consortium	210,480	20,580	189,900	90
					89
4	Control	196,563	24,627	171,936	87
	Sp.1 (*Desulfococcus sp.*)	224,930	19,381	205,549	91
	Sp.2 (*Desulfotomaculum sp.*)	207,207	17,841	189,366	91
	Sp.3 (*Desulfobacter sp.*)	212,080	18,229	193,851	91
	Sp.4 (*Desulfobulbus sp.*)	205,117	15,867	189,250	92
	Sp.5 (*Desulfobacterium sp.*)	219,733	16,313	203,421	93
	Sp.6 (*Desulfotomaculum sp.*)	249,217	22,382	226,834	91
	Consortium	215,213	23,404	191,809	89
					91
6	Control	213,857	19,319	194,538	91
	Sp.1 (*Desulfococcus sp.*)	206,230	21,570	184,660	90
	Sp.2 (*Desulfotomaculum sp.*)	248,897	22,241	226,656	91
	Sp.3 (*Desulfobacter sp.*)	189,877	19,147	170,730	90
	Sp.4 (*Desulfobulbus sp.*)	207,483	15,999	191,484	92
	Sp.5 (*Desulfobacterium sp.*)	189,453	16,898	172,555	91
	Sp.6 (*Desulfotomaculum sp.*)	202,990	15,891	187,099	92
	Consortium	200,970	15,153	185,817	92
					91

In making sulfate reduction, SRB uses sulfate as an energy source, i.e., as an electron acceptor, and uses organic materials as a carbon source (C). The carbon acts as a donor, the electrons in the metabolism, and the cell's building blocks (Groudev et al. 2001). Djurle (2004) adds that the SRB uses electron donor H_2 and the C source (CO_2), obtained from organic materials. The reaction of sulfate reduction by SRB, according to Van Houten et al. (1994, 1995), is as follows:

$$SO_4^{2-} + H_2 + 2 H^+ \rightarrow H_2S + 4H_2O$$

In this research, the initial pH medium used is 2, 4, and 6; after being treated with the increase in the pH, it ranges 5-6. This shows that the SRB effectively increases the pH in each treatment. This occurrence may be because sulfuric acid is so strong that adding this compound into the environment will influence acidity (pH). The more the reduction of sulfate, the higher the PH. Increasing pH for SRB activity reducing the sulfate will have a double impact, i.e., to produce H_2S and bicarbonate ion (HCO). H_2S results from reducing sulfate (sulfate decreases, the pH will increase), and bicarbonate acts to increase the pH

(Frank 2000). Based on the normality test to find out whether a value is a significantly different comparator or equal to the average value of each treatment, if the sample average sig <0.05, there were no significant differences in the samples with comparative values, and the data could not be received. The normality test showed abnormal results, so a non-parametric data analysis was used. The Kruskal-Wallis test analyzed data to analyze the sulfate reduction using a single factor, namely pH or SRB course. The test results showed that the sulfate reduction in pH or SRB treatment alone is not significantly different because sig is > 0.05, and the data is rejected. Then, the data analysis was done by Friedman test to determine the effect of treatment in two factors: pH and SRB. The results of the Friedman test showed that the reduction of sulfate to the treatment of two factors, namely pH and SRB, was significantly different because sig was <0.05.

In conclusion, after doing this research, we concluded that six isolates of sulfate reducing bacteria isolated from ex-coal mine sediment pond in Samarinda, namely sp.1 (*Desulfococcus* sp.), sp.2 (*Desulfotomaculum* sp.), sp.3 (*Desulfobacter* sp.), sp.4 (*Desulfobulbus* sp.), sp.5 (*Desulfobacterium* sp.) and sp.6 (*Desulfotomaculum* sp.) have potential as sulfate reducing agent in ex-coal mine acid soil (TMT) with efficiency sulfate reduction ranging from pH 2, pH 4, pH 6 respectively 89%, 91%, and 91%. This indicates that the sulfate reduction is highest in the medium with pH 4 and 6. It is also noted that sp.5 (*Desulfobacterium* sp.) on medium with pH 4 has the best ability of sulfate reduction efficiency compared to other SRB isolates at 93%. These results align with the Postgate activity test on sulfate reduction in liquid media, showing that treatment with sp.5 (*Desulfobacterium* sp.) gives the best results, i.e., 91% compared with other treatments.

ACKNOWLEDGEMENTS

Thanks to the Faculty of Mathematics and Natural Sciences and LP2M, Universitas Mulawarman, Samarinda, Indonesia, for all facilities and assistance of research that have been given so that research can be undergone.

REFERENCES

Atlas RM, Bartha R. 1992. Hydrocarbon biodegradation and oil spill bioremediation. In: Marshall KC (ed) Microbial Ecology. Vol. 12. Plenum Press. New York, USA. DOI: 10.1007/978-1-4684-7609-5_6.

Djurle C. 2004. Development of Model for Simulation of Biological Sulphate Reduction with Hydrogen as Energy Source. [Thesis]. Department of Chemical Engineering, Lund Institute of Technology, The Netherlands.

Doshi SM. 2006. Bioremediation of Acid Rock Drainage using Sulfate Reducing Bacteria. U.S. Environmental Protection Agency Office of Solid Waste and Emergency Response Office of Superfund Remediation and Technology Innovation, Washington, D.C.

Fahruddin. 2010. Bioteknologi Lingkungan. Penerbit Alfabeta, Bandung. [Indonesian]

Frank F. 2000. Bioremediation by sulfate reducing bacteria of acid mine drainage. Department of Environmental Sciences, UC Berkeley, CA, USA.

Groudev SN, Komnitsas K, Spasova II, Paspaliaris I. 2001. Treatment of AMD by a natural wetland. Miner Eng 12: 261-270. DOI: 10.1016/S0892-6875(99)00004-7.

Hards BC, Higgins JP. 2004. Bioremediation of Acid Rock Drainage Using SRB. Jacques Whit Environment Limited, Ontario.

Havlin JL, Beaton JD, Tisdale SL, Nelson WL. 1999. Soil Fertility and Fertilizers An Introduction to Nutrient Management. 6 th ed. Prentice Hall, Upper Saddle River, New Jersey.

Lieberg EW, Cutright TJ. 1999. The investigation of enhanced bioremediation through the addition of macro dan micro nutrisis in PAHs contaminated soil. J Intl Biodet Biodegrad 44 (1): 55-64. DOI: 10.1016/S0964-8305(99)00060-8.

Tala'ohu SH, Moersidi S, Sukristiyonubowo, Gunawan S. 1995. Sifat fisikokimia tanah timbunan batubara (PT BA) di Tanjung Enim, Sumatera Selatan. Prosiding Pertemuan Pembahasan dan Komunikasi Hasil Penelitian Tanah dan Agroklimat. Buku IV. Bidang Konservasi Tanah dan Air serta Agroklimat. Puslitbangtanak. Cisarua, 26-28 September 1995. [Indonesian]

van Houten RT, Hulshoff Pol LW, Lettinga G. 1994. Biological sulphate reduction using gas-lift reactors fed with hydrogen and carbon dioxide as energy and carbon source. Biotechnol Bioeng 44 (5): 586-594. DOI: 10.1002/bit.260440505.

van Houten RT, van der Spoel H, van Aelst AC, Hulshoff Pol LW, Lettinga G. 1995b. Biological sulfate reduction using synthesis gas as energy and carbon source. Biotechnol Bioeng 50 (2): 136-144. DOI: 10.1002/(SICI)1097-0290(19960420)50:2<136::AID-BIT3>3.0.CO;2-N.

Widyati E. 2006. Bioremediasi Tanah Pasca tambang Batubara dengan Sludge Industri Kertas untuk Memacu Revegetasi Lahan. [Disertasi]. Sekolah Pascasarjana IPB. Bogor. [Indonesian]

Widyati E. 2007. Pemanfaatna bakteri pereduksi sulfat untuk bioremediasi tanah bekas tambang batubara. Biodiversitas 8 (4): 283-286. DOI: 10.13057/biodiv/d080408.

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 83-94

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070205

Analysis of wave energy reduction and sediment stabilization by mangroves in Gazi Bay, Kenya

MAINA DAVID NDIRANGU[1], ROBERT MUTUGI CHIRA[1,♥], VIRGINIA WANG'ONDU,[1] JAMES G. KAIRO[2]
[1]School of Biological Sciences, University of Nairobi. P.O. Box 30197 (00100), Nairobi, Kenya. ♥email: rchira@uonbi.ac.ke rchira@uonbi.ac.ke
[2]Kenya Marine and Fisheries Research Institute. Mombasa, Kenya

Manuscript received: 5 May 2017. Revision accepted: 29 September 2017.

Abstract. *Ndirangu MD, Chira RM, Wang'ondu V, Kairo JG. 2017. Wave energy reduction and sediment stabilization by mangroves in Gazi Bay, Kenya. Bonorowo Wetlands 7: 83-94.* Mangrove forests provide natural protection to the coast by attenuating wave energy and stabilizing sediments. The efficiency of coastal protection is likely to decline with increased degradation and losses of mangrove forests. Nevertheless, there are few empirical studies to test these hypotheses. The study's objective was to investigate how wave energy and sediment stabilization vary with tree density in a mono-species stand of mangroves (*Sonneratia alba*) at Gazi Bay, Kenya. Seven belt transects were randomly selected along 900 m stretches of shoreline with homogenous emergent wave energy. Structural parameters, including; tree density, pneumatophores density, and basal areas, were quantified using two quadrat measuring 25 m by 20 m along each transect. Three intertidal stations with five sampling points were sampled using uniform plaster of Paris clod cards for wave energy. Sediment stability was measured using improvised sinking metal disks made from bicycle spokes, and sediment accretion was monitored by Surface Elevation Tables (SETs). All transects showed significant difference in pneumatophores density ($F_{(2.39)}$ =25.15), tree density ($F_{(2.33)}$ =24.79), and basal area ($F_{(2.39)}$ = 29.66). The wave energy sampled by tree stems between stations and the correlation of wave energy reduction against tree density/ha, pneumatophores density, and basal areas per m^2 also showed a significant difference. Regression analysis showed a significant difference in the sediment stability against tree density (R2 = 61%) and basal areas (R2 = 72.8%), while there was no significant difference between sediment stability and pneumatophores density (R2= 47%). Regression analysis between mean sediment accretion rates against all parameters was not significant. This study will help the managers and the government on the merit of using mangroves as bio-shields in protecting coastlines against erosion and stabilizing sediment in the wake of much anticipated global changing climate and sea-level rise.

Keywords. Mangroves, Gazi Bay, Kenya, sediment stabilization, wave energy

INTRODUCTION

Mangrove forests are essential ecosystems that stabilize coastlines and protect life and property (Keryn et al., 2011). The forest extends to the intertidal areas of sheltered tropical and subtropical shores. They are confined mainly to the equatorial region between latitude 30^0 north and 30^0south (Spalding et al. 2010). Globally, mangroves are estimated to cover 14-24 million ha, spread over 122 territories (Giri et al., 2010). The largest under mangrove forest is in Southeast Asia, which occupies 34-42% of global mangroves (Spalding et al., 2010).

Nigeria has the highest density of mangroves in the African continent with 4.7%, followed by Mozambique and Madagascar with 2.3% and 2.0%, respectively (Spalding et al. 2010). Considering the multiple goods and services provided by the mangrove ecosystem, there is increased effort to restore degraded mangrove forests globally (Gilman et al., 2008). Efforts to replant degraded mangrove forests were started in Kenya in the 1990s and are still ongoing (Kairo, 1995).

In Kenya, there is some information on human-induced mangrove deforestation and transformation (Abuodha and Kairo 2001). However, only little is known about the effects of degradation on ecosystem functions, such as shoreline protection. For example, at the Gazi pilot area, removing the fringing mangroves has led to the erosion of adjacent coconut plantations (Dahdouh-Guebas et al. 2000). Some 30-40% of nearshore mangroves at Gazi comprise Sonneratia alba forest, mainly adapted to frequent inundation (Neukermans et al. 2008).

In the context of climate change, mangroves build new land by accreting sediment either *in-situ* or exogenous (Krauss et al. 2013), thus balancing relative sea-level rise (Bell and Lovelock 2013). In the recent past, mangrove forests have provided shoreline protection (Bell and Lovelock 2013). The forest attenuates wave energy and binds sediment by its roots (Mazda et al. 1997). The previous study reported that the Caribbean coast of Belize, Honduras, and Panama (McKee et al. 2007) had shown sediment capture of 4.1 ± 2.2 cm by fringing mangrove forest, which is likely to counter the global mean rates of eustatic sea-level rise of 1.5-2 mm $year^{-1}$ (Krauss et al. 2003)

This study measured wave attenuation by mangroves at Gazi Bay, Kenya, to establish baseline data on the protective functions of mangroves in the area. In Kenya, mangroves cover was approximately 45,590 ha and distributed along 600 km of the coastline by 2010 (Kirui et al. 2012). The most significant number of mangroves in Kenya is found in Lamu, 33,500 ha, Kwale district, 8375 ha, Kilifi district, 5570ha, Tana River district, 3045 ha, and

Mombasa district, 2490 ha (Abuodha and Kairo 2001) (Figure 1).

The objectives of this research were (i) to determine the effect of trees and pneumatophores density on wave energy reduction; (ii) to assess the impact of tree and pneumatophores density on sediment stability and accretion.

MATERIALS AND METHODS

Study areas

The study was performed at Gazi Bay, approximately 60 km from Mombasa, Kenya, on the south coast of Kenya ($4^0$25'S and 4^0 27'S; $39^0$50'E and 39˙50ºE). With a surface area of 18km^2, the Gazi Bay has an estimated mangrove cover of 615 ha (Kairo et al. 2001). The Chale peninsula shelters the bay to the east and a fringing reef to the south, where a further distance on-shore, a degraded and semi-pristine fringing mangrove forest occurs (Figure 1). The study site was severely degraded following deforestation in the 1970s and early 1980s through unsustainable harvesting of industrial wood fuel. The width of the fringing mangrove forest ranges between 40-90 m in width and 800 m long. The narrow stretch of fringing mangrove forest is inundated daily during spring and neap tide (Kairo 1995).

Hydrology and rainfall

The climate of Gazi Bay resembles that of the Kenyan coast, which is influenced by both southeast and northeast monsoons. Total annual precipitation exhibits a bimodal precipitation pattern, ranging between 1000 mm to 1600 mm (Kairo, 1995). During spring, the tidal range in Gazi Bay falls around ~4.0 m (Kirui et al., 2012). Long rains are under the influence of the southeast monsoon, starting in April to August, while northeast monsoon influences the short rains

beginning from October to December. The average annual temperature is 26°C, and the humidity is approximately 65-80% (Bosire et al., 2003).

The Gazi Bay is an estuary of two seasonal rivers, Kidogoweni River and Mkurumuji River, which depend on rainfall experienced inland. River Mkurumuji is more prominent with a catchment of 175km^2 inland and has high flow rates; a maximum and minimum flow of 0.02m^3/s and 5.90 m^{3,} respectively, while groundwater seepage is restricted to small parts (Kitheka et al. 2003). The hydrology of the more significant Mkurumuji river contributes positively to allochthonous nutrients together with the upcoming anthropogenic inputs from agricultural farms and mining industries during the rainy season (Kitheka et al., 2003). Human activities, mainly sugar plantations at Ramisi and Titanium mining in Kwale County, may negatively affect allochthonous sediment input due to damming upstream in both rivers.

Geology

Geologically, the Kenyan coast comprises rocks, mostly marls, and limestone, represented by sandstones, clays, conglomerates, and gravels (GOK 2009). The building industry extensively exploits these well-developed reef complexes, coral reefs, coral rubble, and sandstones (GOK 2009). Fringing reef crests dominate the Kenyan coast forming a natural barrier to the wave energy from the ocean with a shoreline comprising of reef terraces and mangroves, with a tidal flat in Gazi Bay fronting the mangroves (GOK 2010). The study site is conducted on a low shore beach that receives daily inundation (Watson inundation class 1) and has moderate wave exposure and sandy sediments. The highest spring tide is approximately 4 m above the datum (Kitheka et al. 2003).

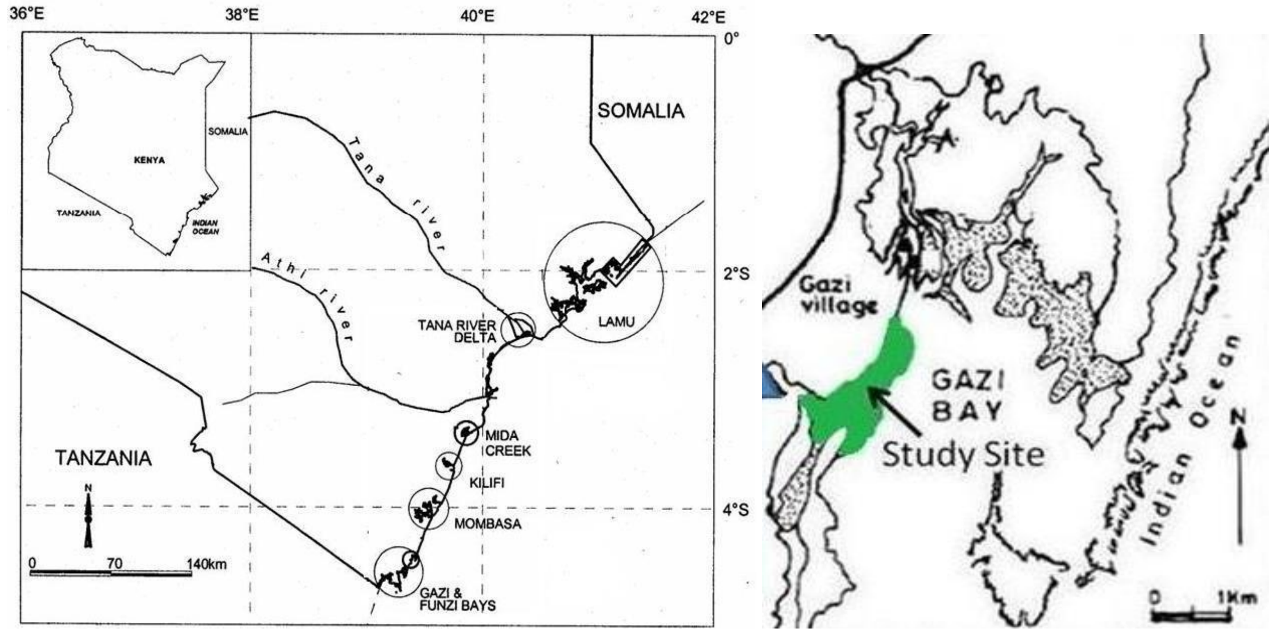


Figure 1. A map of the Kenyan Coastline showing the study site comprising of fringing mangroves in Gazi Bay (Bosire et al. 2003)

Mangroves of Gazi

Gazi Bay possesses all the nine mangrove species described in the western Indian Ocean region. The dominant species that make up more than 70% of the formation are *Ceriops tagal* (Perr) C.B Robinson and *Rhizophora mucronata* (Lam) (Kairo et al., 2008). A typical pattern of species that occurs from the sea to the land are *S. alba, R. Mucronata, Bruguiera gymnorrhiza*, (L) Lam *C. tagal*, *Avicennia marina* (Forsk) Vierh, *Xylocarpus granatum* (Koen), *Lumnitzera racemosa* (Willd), *Xylocarpus muluccensis* (Lam) Roem and *Heritiera Littoralis* (Dryand) (Dahdouh-Guebas et al. 2000). The massive tidal regime controls these species. Animal species range from mollusks; to sesarmid crabs which are keystone species due to their high rate of nutrient cycling (Bosire et al. 2008). Other common species of crabs include *Uca annulipes* and *Uca inverse* (Skove et al. 2002).

Epibiotic communities of algal species encountered in seaward and landward zones include *Enteromorpha ramulose*, *Polysiphonia* sp., *Hypnea* sp., and *Caloglossa leprieuri* attached to pneumatophores of *S. alba* at different pneumatophores height. Poriferans are found growing on the pneumatophores of *S. Alba* stands with *Tedania digitata vulcanis* as a typical example (Crona et al. 2006).

Gazi Mangrove forests are habitats for juvenile fish, either planktivorous or benthic feeders. Common juvenile species of fish located in the fringing mangrove forest are *Gerres oyena* and *lutjanus fulviflamma* (Kirui et al., 2012). Also, the forest forms a suitable habitat for different species of snakes, residents, and migratory birds that commonly feed in nearshore environments (Kairo 1995).

Social-economic activities

Gazi Bay has a population of approximately 4825 people according to the national census of 2009, the majority being women and children. The men (90%) are involved in artisanal fishing and mangrove cutting for sale to Mombasa and other coastal towns (Hamsa 2013). Approximately 90% of mangroves harvested are used for building houses, tannins, provision of wood fuel, ribs for boat manufacture and furniture, traditional medicine (Dahdouh-Guebas et al. 2000). Meanwhile, women are mostly engaged in small businesses and weaving makuti (roof thatches made from coconut fronds) farming (Dahdouh-Guebas et al., 2000). Mangroves have also been used extensively by undergraduate and postgraduate research students from various disciplines to address various research questions (Kairo et al., 2009).

Recently, Mikoko Pamoja, a community-based mangrove conservation initiative, has started mangrove restoration and protection in the bay. Together with KMFRI and Universities in the UK, their attempt for mangrove restoration and preservation is made by selling carbon credits in the voluntary carbon market (Hamsa 2013).

Project design

Change in wave energy was measured across seven belt transects selected randomly and perpendicular to the shoreline in a mono-species of fringing of *Sonneratia forest* (Figure 2). The seven transects had varying tree and pneumatophores densities and were denoted with letters K, L, M, N, O, and P. Among the seven transects, transect Q had no mangrove cover and was selected as a control. The belt transect was set short from river Mkurumuji, heading north towards the Bay. This selection was made to incorporate vegetation structural differences of fringing mangroves.

The positioning of transects was done in areas where the bandwidth of the mangrove trees stretch was ≥ 80m. This positioning was made because the bandwidth of the mangrove forest was not uniform at the study site. It was the most extensive stretch of mangrove cover that existed and allowed a sufficient number of transects.

Sampling design

The density of mangrove trees was quantified in seven; 80 m long belt transects by utilizing two plots, each measuring 25 m long and 20 m width. The plots were systematically selected along each transect set perpendicular to the shoreline. The first plot was established 20 m from the lowest mangrove tree on the seaward side, while the second plot was set 5 m from the first plot toward the landward side. Pneumatophores density was determined by systematically selecting five quadrants, each measuring 1m by1m nested in each of the more prominent plots of 25m by 20m. At each corner, the four plots of 1 m by 1 m were set, one at the middle of each of the two plots. A total of ten plots per transect were selected to estimate pneumatophore density per transect. A total of 12 plots were selected to calculate mangrove trees and pneumatophores density, respectively, at the study site.

Wave energy was measured at three stations marked A, B, and C across seven transects set perpendicular to the shoreline. A total of 21 stations were characterized in the study area. Station A was ~20 m from the last mangrove trees on the seaward side, which measured incident wave energy before mangrove influence; station B was 50 m from station A while station C was 80 m from station A. Station B, and C measured incident wave energy passing through the mangrove forest. Each station was marked with five wooden poles of 10cm girth, 1.5m high, driven 70 cm into the ground.

Paris clods were used to measure wave energy. The clods were prepared by mixing 11.4ml of freshwater with 14.3g plaster of Paris powder manufactured by LAFARGE PRESTIA (*www*.lafargeprestia.fr) (Figure 3). The powder was slowly added to water and stirred with a spoon. The slurry was poured into plastic ice cube trays. The trays were tapped several times vigorously to dislodge air bubbles and allowed to harden for between 20-30 minutes before removing them. The cards were oven-dried for 48 hours at 40 degrees centigrade following this step. The final weight of the clod after exposure to the field was determined by subtracting the total weight of the clod plus the plastic plate and the silicon cement before exposure. The basic assumption was that the weight of the plastic and that of the silicon cement remained constant and did not change due to corrosion by seawater.

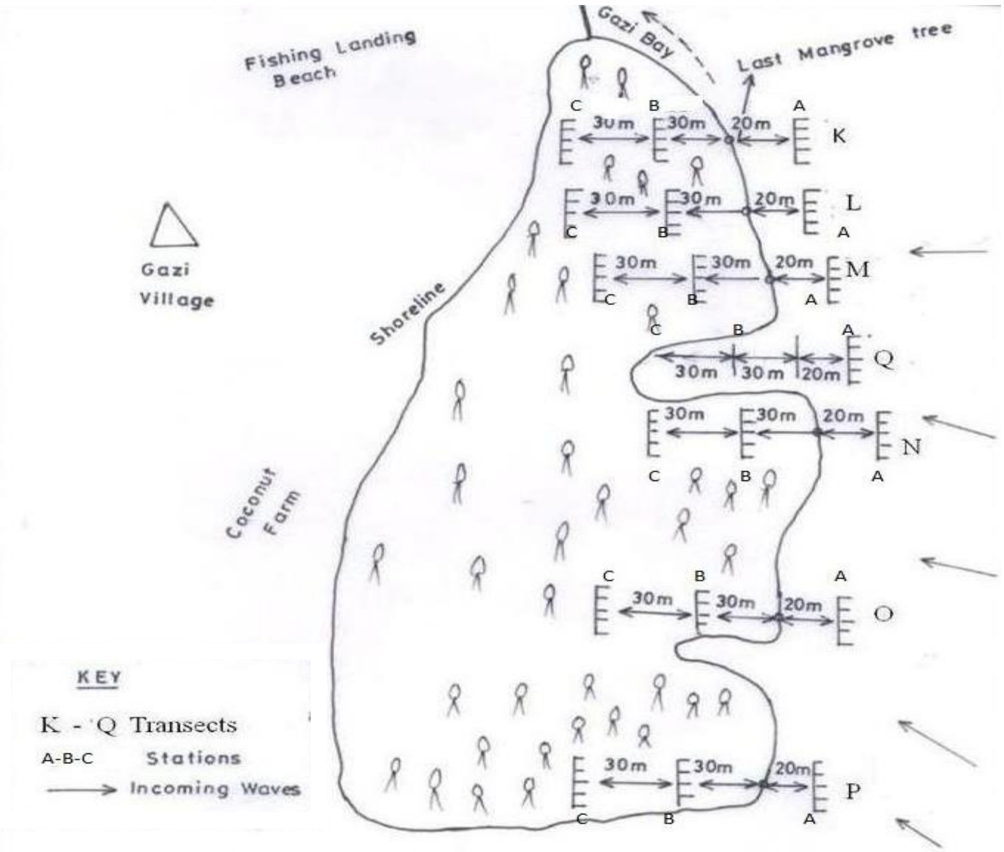


Figure 2. Schematic diagram showing the relative position of stations in each transect at the study site (not drawn to scale)

The clod cards were then sanded at the bottom to attain a uniform weight of 14 ± 1.5 g within a batch. They were glued to a 3 cm×8 cm plastic plate with silicone cement (No-Nonsense Ltd.BA 228RT). These cards were taken to the field and mounted on the poles at 15 cm and 50 cm from the substrate surface, respectively, as shown in Figure 4. The clod cards mounted at 15 cm measured wave energy reduction by pneumatophores, while the clod cards mounted at 50 cm measured wave energy reduction by mangrove tree stems.

Soil samples were collected at 10 points extending the three stations for each transect: station A on the seaward side, station B in the middle of mangrove forest, and C on the landward side. The soil samples were obtained using a cover at approximately 5 cm depth. The points at which the samples were collected cover a distance of 25 m and each station and are separated 5 corerm away from each other. The ten samples for each station were pooled in a one-labeled plastic bag and mixed thoroughly to make one composite.

The sediment stability was measured using cheap and inexpensive bicycle spokes fitted with metal disks at the three stations in the same transects set perpendicular to the shoreline where wave energy was determined. As many as ten replicate points, which were 2.5 m apart, were systematically selected, covering a25 m along with each station. The spokes were fitted with the metal disks weighing 2.8 g and 1.7 cm in diameter and inserted approximately 5 cm bicycle in the sediment. The spokes held the metal disks and left them at the substrate surface so that unstable sediment could cover them as sediment settles down during the alternating high and low tide regimes. A total of 30 spokes per transect were used in six transects with contrasting mangrove tree densities. One transects without tree cover was established perpendicular to the shore as a control. The bicycle spokes and the metal disc were inexpensive for estimating sediment stability because the materials used were cheap and locally available. We suggest that the metal disk was denser than seawater and sunk during high tide. Second, the dimension of the spoke, such as thickness, did not modify the site by reducing water velocity, thus facilitating sediment to settle. Furthermore, all spokes and metal discs at station A before the mangroves on the seaward side were used to control all transects.

Measurement of sediment accretion was done using 3 m long and 1 cm diameter stainless steel rods. Two metal rods were set 50 cm apart and driven 280 cm into the sediment at station B, which was almost in the middle of the transect. A portion of 20 cm ± 0.1 cm of each rod was left projecting perpendicular above the substrate surface. Leveling was done using the spirit level to ensure both steel rods were at the same height. Thus, sediment accretion was estimated at six sites in six transects set perpendicular to the shoreline.

The use of steel rods has the advantage of determining accretion and erosion and gives information about subsurface processes down to the maximum level at which the pins are driven. The use of rods in the surface elevation tables (R-SET) is advantageous since it is cheap regarding cost, high-precision, simple, and is accessible to economically developing countries with coastal wetlands. In this experiment, a wooden board 4.2 cm × 2.2 cm and 50 cm long smoothened flat, and the square was used to determine sediment accretion. The wooden board was placed on the two rods projecting above the substrate. Ten replicate points were marked along the entire length of the wooden board. The wooden board was set on the two rods projecting on the substrate surface to determine surface elevation changes at each transect.

Data collection methods

Within the two plots measuring 25 m by 20 m along each transect, diameter at breast height (DBH) of mangrove tree stem was measured at D130 or diameters at 1.3 m above the ground (Cintron and Schaeffer-Novelli, 1984). All mangrove tree stems in diameter D130 ≥ 2.5 m were measured and recorded. Stand density per hectare for each transect was calculated using the relation. **Di = ni/Ai.** *Where;* $\mathbf{D_i}$ is the density for species **i,** $\mathbf{n_i}$ is the total number of individuals counted for species, and $\mathbf{A_i}$ is the total area sampled (Brower and Carl., 1990). Basal areas were calculated using the formula; BA= 0.00007854×DBH2 (where DBH is the diameter at breast height). Ten pneumatophores were selected randomly in ten quadrats per transect measuring 1m× 1 m. Their height and diameter at ½ heights were measured using a standard ruler and Vanier calipers, respectively. The density of the pneumatophores was calculated per meter square in each transect and recorded.

Wave energy was sampled twice covering northeast monsoon season by exposing clod cards to wave action for 48 hours on the 8th to 10th October 2012 during spring. A replicate sampling of wave energy was taken during neap tide on the 16th -18th November 2012. During the southeast monsoon season, wave energy sampling was done by exposing clod cards for 48 hours on the 1st to 3rd March 2013 during spring tide. A replicate sampling of wave energy was retaken on the 25th to 27th May 2013. During the four sampling regimes, clod cards were mounted and retrieved during low water.

Figure 3. Oven-dried clod cards attached to plastic plates with silicon cement before exposure in the field (Photo by MDN)

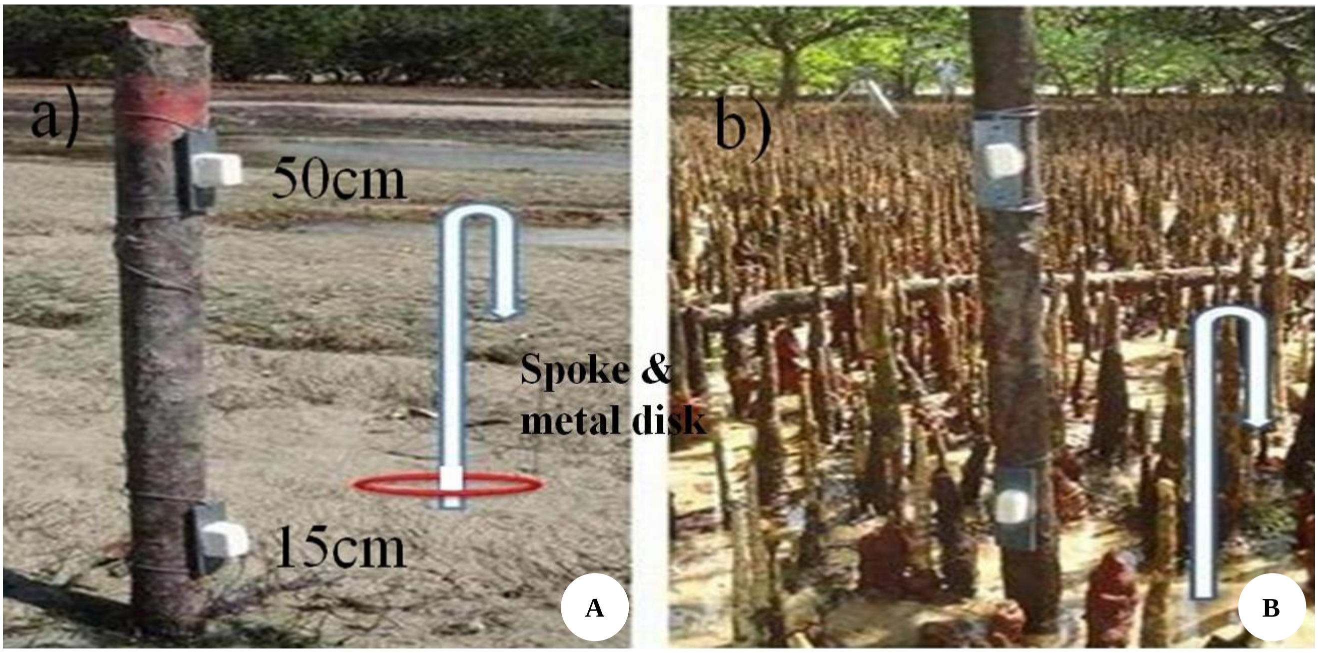


Figure 4. A setup showing (A) station A on the seaward side; (B) station B in the mangrove; with upper (50 cm) and lower (15 cm) clod card, a bicycle spoke, and a metal disk (Photo by MDN)

Figure 5. Surface elevation station measuring sediment accretion in a *Sonneratia alba* forest at Gazi Bay, Kenya (Photo by MDN)

The underlying assumptions were; First, station A would be impacted more by high water velocity in all transects due to the absence of mangrove forest structures, thus experiencing high dissolution rates of clod cards. Secondly, lower clod cards sampled the energy absorbed by pneumatophores, while the upper clod card sampled the hydrological energy consumed by the tree stems and pneumatophores. Also, the magnitude of the tide could not change within the same tide cycle during clod card exposure.

Data collection of the soil samples was done in the laboratory. After drying, the samples were weighed; oven-dried at 80°C for 24 hours to determine the percentage moisture content. A sub-sample of 25 g from each station was accurately weighed from the dried soil samples for grain size analysis. The second set of samples weighing 25 g was labeled and preserved for organic matter analysis.

The sediment (25 g) was put in a labeled beaker and treated with (6.2g/1 dilution) of 250 ml water and 10 ml aqueous sodium hexametaphosphate ((NaPO3)6). The solutions were stirred for 10 minutes, left for four hours, and then stirred for 10 more minutes. The content of the beaker was flushed through a 63 µm sieve with fresh water until no further silt was lost. The wet sediment was carefully flushed into a pre-weighed foil boat and oven-dried at 80 0 C for 12 hours. Next, the dry content was weighed to determine the weight of silt. The dry sediment was then flushed through a 500 µm sieve. The sediment grain size >500 µm was weighed and recorded and used to calculate the weight of the 63-500 µm fraction. From the oven-dried preserved sample of 25 g, a sub-sample of 10 g was weighed and put in pre-weighed aluminum crucibles and set in a muffle furnace for combustion at 440 0 C for 6 hours. The procedure was followed by cooling the samples in a desiccator and weighing them to determine the organic matter in the sediment.

The sediment stability was monitored by estimating the depth at which the metal disks sunk in the sediment. Every month, the depth of sinking metal disks was estimated using a standard ruler during low tide. New spokes and metal disks were reset for the presiding month to avoid confounding due to changes in the disks' mass due to the corrosive nature of seawater. Sediment accretion was estimated using a standard ruler by measuring the height from the substrate to the wooden board placed on the two rods. Ten replicate measurements were taken and recorded once each month for all the six transects from the ten points marked on the wooden board (Figure 5). The measurements were taken for 8 months, covering 240 days

Data analysis methods

Data were analyzed using MINITAB 14.0 software package. One-way analysis of variance (ANOVA) was performed to test the variations in three parameters: tree density, pneumatophores density, and basal areas between transects. The same data analysis was also performed to examine how wave energy varied between the three stations in different transects with varying tree and pneumatophores densities. Pearson's correlation analysis was carried out to investigate the significance of the forest structure, namely, tree density, pneumatophores density, and basal areas against wave energy.

Correlation analysis was conducted to test for significant differences between silt, fine sand, coarse sand, and organic matter against tree and pneumatophore density. ANOVA was used to test for variation in sediment stability during the two sampling regimes and differences between stations in different transects. Regression analysis was carried out to check for sediment stability and sediment accretion against tree density/ha and pneumatophores density m^2.

RESULTS AND DISCUSSION

Forest structure at the study site

Transect M had the highest pneumatophores density 174±21.1 Ind. /m^2. Tree density and basal areas were highest in transect L, 690±330 Ind/ha, and 11.5±2.6/$m^{2,}$ respectively, whereas transect O had the lowest structural attributes in the study site. All transects showed significant difference in tree density ($F_{(2,\ 33)}$ =24.79, p = 0.000) pneumatophores density (F ($_{2,\ 39)}$ =25.15, p = 0.000), and basal area ($F_{(2,39)}$ = 29.66, p = 0.01). Table 1 summarizes the structural parameters of mangrove forests in the study area.

Effect of tree and pneumatophores density on wave energy reduction

Wave energy sampled by stems in all station A on the seaward side before the mangrove influence had the highest mean in wave energy expressing 81.4 ± 5.1% (SE), reducing gradually from station B (73.1 ± 4.9% (SE)) to C (65.5 ± 5.5% (SE)). Transect Q with no mangrove cover had the highest mean in wave energy sampled by tree stems, expressing a mean of 84.3 ± 1.7% (SE) in station A, 81.5± 1.4%, and 77.9 ± 2.0% (SE) in station B and C, respectively. The mean wave energy sampled by mangrove stems was 73.4 ± 4.8, while the mean of the control station Q had 81.2 ± 2.9. The mean wave energy expressed in the control denoted with letter Q (without mangrove trees) was higher by 7.8% than the mean wave energy reduction in the six transects with mangrove tree cover. Table 2 summarized the results for wave energy reduction along with stations A, B, and C in all transects sampled at 50 cm by tree stems and the mean of the three stations per transect.

Wave energy sampled at 15 cm, which expressed energy reduction by pneumatophores in all station A on the seaward side before mangrove influence, had the highest mean of wave energy expressing 84.7 ± 2.7% (SE). Results for wave energy reduction across stations A, B, and C in all transects sampled at 15 cm by pneumatophores and the mean of the three stations per transect are shown in (Table 3).

The energy from stations B to C has gradually reduced at an average of 77.5 ±2.8 and 73.5 ± 2.8% (SE), respectively. Transect Q (Control) without pneumatophores cover had the highest mean in wave energy in station A, expressing 86.4 ± 2.5, while station B and C expressed 84.3 ± 1.5 and 81.3 ± 1.6% (SE), respectively. On average, wave energy reduced by pneumatophores in the six transects with varying pneumatophore cover was 79.0 ± 1.9. Meanwhile, the mean wave energy measured at the three stations (A, B, and C) in transect without pneumatophore cover (control transect) had 84.0 ± 2.9. Therefore, the difference in wave energy reduction in areas covered by pneumatophores and those without (the control transects) was 5%. ANOVA showed a significant difference in the wave energy reduction by pneumatophores between stations A, without pneumatophores cover, and stations B and C with pneumatophores cover ($F_{(2.18)}$ =17.05, p = 0.000).

Total energy reduced by mangrove trees reached 7.8%, while the energy reduced by pneumatophores was 5.0%. The entire wave energy reduced by both tree density and pneumatophore density was 12.8%.

To get the relation between wave energy change across the mangrove forest and better understand the effect of tree density on the wave energy, a plot of wave energy on the Y-axis and transects with different tree densities on the X-axis was plotted. The wave energy reduction measured at 50 cm above the substrate (energy reduction by mangrove tree stems) is shown in Figure 7. The result showed wave energy decreasing with increasing tree density.

Table 1. Mean values ± SD of structural characteristics of the mangrove forest at Gazi Bay, Kenya

Transect	Tree height (m)	Basal area (ha)	Tree Density (stems/ha)	Pneumatophores Density(m^2)
K	6.5±0.7	8.1±7.6	320±300	57.2±19.8
L	5.4±0.2	11.5±2.6	690±330	146.3±1.3
M	6.1±0.3	18.1±1.3	580±240	174.9±21.1
N	5.5±0.1	8±1.3	496±208	88.6±19.8
O	4.2±0.5	1.2±0.6	92±28	38.4±24.4
P	5.2±0.3	6.8±3.2	510±50	173.7±37.9
Mean	5.5±0.8	7.3±3.4	448±212.3	113.2±59.8

Table 2. Mean values ±SE of wave energy reduction by mangrove stems (sampled at 50 cm in the three stations; A (Open areas in the seaward side) and station B and C in the mangrove

	Weight loss of Clods cards (%)			Wave energy reduction by mangrove tree stems
Transect	A	B	C	Mean wave energy per transects
K	87.6±0.5	78.7±1.6	69.9±7.5	78.7±8.2
L	84.0±1.2	72.0±1.2	61.8±1.2	72.6±10.3
M	81.6±1.2	74.9±7.9	75.3±1.9	77.3±3.5
N	80.7±2	72.1±1.6	64.4±2.3	72.4±7.5
O	85.3±1.5	78.8±0.6	66.6±2.4	76.9±8.7
P	69.4±1.6	62.3±4.5	55.1±8.3	62.3±6.6
Mean wt. loss	81.4±5.1	73.1±4.9	65.5±5.5	73.4±4.8
Control (Q)	84.3±1.7	81.5±1.4	77.9±2.0	81.2±2.9

Note: One-way analysis of variance (ANOVA) showed a significant difference in wave energy sampled by tree stems between stations A, B, and C ($F_{(2,\ 18)}$ = 10.92, p = 0.001).

The plot for hydrological energy sampled at 15 cm above (energy reduction by pneumatophores density) against pneumatophores density in the seven transects showed the same trend as the energy reduction sampled by tree density where wave energy-reduced with increasing pneumatophore density (Figure 8).

The results for Pearson's correlation of wave energy showed significant negative correlation coefficient for the three forest structures namely; Wave energy against tree density/ha (r^2 = -0.594, p = 0.000), wave energy against pneumatophores density/m^2, (r^2 = -0.794, p = 0.0000) and wave energy against Basal area (m^2) (r^2 = -0.451, p = 0.000).

Sediment grain size and organic matter distribution

The analysis of grain size showed a higher percentage of silt in transect M, K, and L expressing 15.2 ± 5.8%, 15.0 ± 2.2%, and 8.8 ±2 1.2% (SE) while coarse sand was highest in transects P, O, and N expressing 39.7 ± 0.1, 18.1 ± 0.5 and 12.8 ± 0.9 (SE) respectively. All transects expressed mean fine sand as the highest proportion of sediment by 79.5±4.7 % (SE). Coarse sand was least frequent in all transects except transect P, which had a mean of 39.7 ± 0.1%. Transect Q displayed the highest percentage of fine sand with 93.1 ± 0.4% (SE).

Pearson product-moment correlation showed no significant difference between mean silt and tree density (r^2 = 0.530, p = 0.221), fine sand and tree density (r^2 = -0.307, p = 0.503) and coarse sand and tree density (r^2 = 0.083, p = 0.860) among transect in the study site. Correlation between mean silt and pneumatophores density ($r^{2\,=}$ 0.484, p = 0.271), fine sand and pneumatophores density (r^2 = -0.580, p = 0.171) and coarse sand (r^2 = 0.358, p = 0.431) was also not significant in all transect.

Organic matter distribution was highest in transect K, M, and N expressing 2.1 ± 0.5, 1.9 ± 0.4, and 1.6 ± 0.5 (SE) respectively, while the control transects (Q) with no mangrove cover expressed the lowest percentage of 0.8 ± 0.1 (SE). No significant correlation of organic matter between tree density/ha (r = 0.521, p = 0.230) and also in pneumatophores density/m^2 (r = 0.367, p = 0.419). Results for sediment grain sizes and organic matter content during the study period are summarized in (Table 4).

Effect of tree and pneumatophores density on sediment stability and accretion

Mean sinking depths of metal disks in the entire sampling period were pooled per stations A, B, and C covering both southeast and northeast monsoon seasons. All stations before the mangrove influence on the seaward side (station A) displayed the highest means of sinking depths of metal disks of 14.6 ± 3.1mm (SE). The depth reduced gradually from station B to C, which attained a sinking depth of 8.2 ± 2.7 mm (SE) and 7.4 ± 3.5 mm (SE), respectively. In contrast, near river Mkurumuji, transect P recorded a slightly high sinking depth in station C on the landward side with 7.35 ± 0.8 (SE) than stations B and C, which had similar values of 5.78 ±0.6 (SE). On the other hand, Transect Q with no mangrove cover registered the highest mean depth of 23.2 ± 2.3 mm (SE) in the three stations. Unlike most stations in another transect with varying tree cover, station A in the open area (control transect Q) had the least depth of 19.9 ± 2.4 (SE) compared with stations B and C, which recorded 21.9 ± 2.2 and 27.7 ± 2.9 (SE) respectively. Transect M had the least mean depth of 2.9 ± 1.1 1.6 (SE) in all transects.

ANOVA showed a significant difference in sinking depth of metal disks between stations A B and C in transects with varying mangrove tree cover ($F_{(2, 15)}$ = 4.57, p = 0.028). The results for sediment stability comparing different stations along seven transects are shown in (Figure 9).

Table 3. Mean values ±SE of wave energy reduction by pneumatophores in the three stations; A (Open areas on the seaward side) and station B and C in the mangrove

	Weight of Clods cards (%)			Wave energy reduction by pneumatophores
Transect	A	B	C	Mean wave energy reduction per transects
K	85.5±0.6	75.7±1.8	68.3±1.2	76.5±9.8
L	80.3±2	72.9±1.9	71.2±1.8	74.8±5.5
M	88.5±0.9	82.3±1.2	75.4±1.6	82.1±7.4
N	85.5±2	75.5±1.4	73.5±1.6	78.2±7.3
O	87.8±1.6	80.9±0.5	74.2±0.9	80.9±7.7
P	81.0±8.8	77.8±7.3	78.3±2.8	79.0±1.9
Mean wave energy	84.7±2.7	77.5±2.8	73.5±2.8	78.6±2.3
Control	86.4±2.5	84.3±1.5	81.3±1.6	84.0±2.9

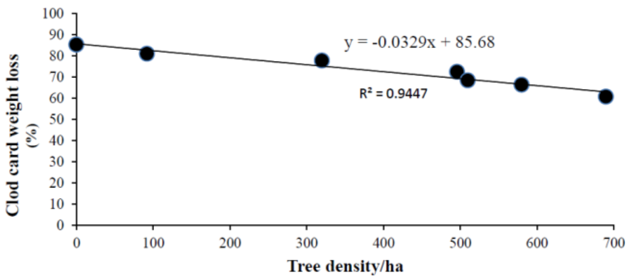


Figure 7. Linear relationship between wave energy against tree density/ha in seven transects

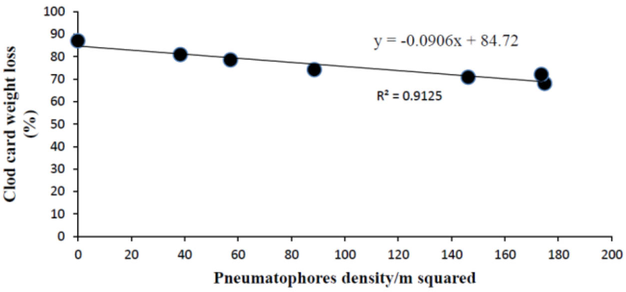


Figure 8. Linear relationship between wave energy against pneumatophore densities in seven transects

Effect of tree and pneumatophores density on sinking metal disks

The means for sinking metal disks of different stations were determined, and results compared with tree density/ha and pneumatophores density per m^2. The depth of metal disks decreased with an increase in mangrove tree density (Figure 10).

A plot of sinking metal disk against pneumatophores density per m^2 showed similar treads with the one plotted against tree density/ha. The depth of metal disks decreased with increased pneumatophores density per m^2 (Figure 11).

The regression analysis showed a significant difference in sediment stability against tree density (R^2= 61%, P = 0.028) and basal areas (R^2 = 72.8%, p = 0.015) whilst there was no significant difference between sediment stability and pneumatophores density (R^2= 47%, p = 0.089).

Sediment accretion rates in six transects showed a net positive gain of 6.3±2.1mm (±SE). For 330 days. When extrapolated to annual rates, the transects showed values equal to a mean of-of 5.4±1.7mm (±SE). The mean range between the highest and the lowest accreting transect was 12.2mm recorded between transect M and transect O, respectively. Mean sediment accretion rates were highest in transects K 13.3 ± 1.8mm (±SE), followed by transect M with 11.5 ±2.1 (mm) (±SE). The lowest mean accretion rates were recorded in transect P with 1.4±1.2mm (±SE) and transect O with 1.1±2.8mm (±SE) within the same period. Sediment accretion rates as shown in (Figure 12).

Regression analysis between mean sediment accretion rates against forest structures showed no significant difference namely; tree density/ ha showed no significant (R^2 = 7.2%, p = 0.608), basal areas (R^2 = 8.9%, p = 0.566) and pneumatophores density/m^2 (R^2= 11.1%, p= 0.415) In addition, regression analysis between sediment accretion and wave energy reduction along transects was also not significant (R^2= 16.7, p = 0.489).

Table 4. Percentage of sediment grain sizes classes and organic matter content in the seven transects (Mean values ± SE)

Transect	Silt (%)	Fine sand (%)	Coarse sand (%)	Organic matter (%)
K	15.0 ± 5.8	81.7 ± 4.7	3.3 ± 1.0	2.1 ± 0.5
L	8.8 ± 1.9	87.6 ± 0.9	3.5 ± 0.2	1.4 ± 0.2
M	15.2 ± 2.2	79.6 ± 2.9	5.3 ± 0.1	1.9 ± 0.4
Q (control)	3.8 ±0.5	93.1 ± 0.4	3.5 ± 2.1	0.8 ± 0.1
N	5.6 ± 3.3	81.6 ± 2.3	12.8 ±0.9	1.6 ± 0.5
O	2.4 ± 1.4	79.5 ± 0.8	18.1 ± 0.5	1.1 ± 0.3
P	7.0 ± 2.3	53.4 ± 2.4	39.7 ± 0.1	1.2 ± 0.1

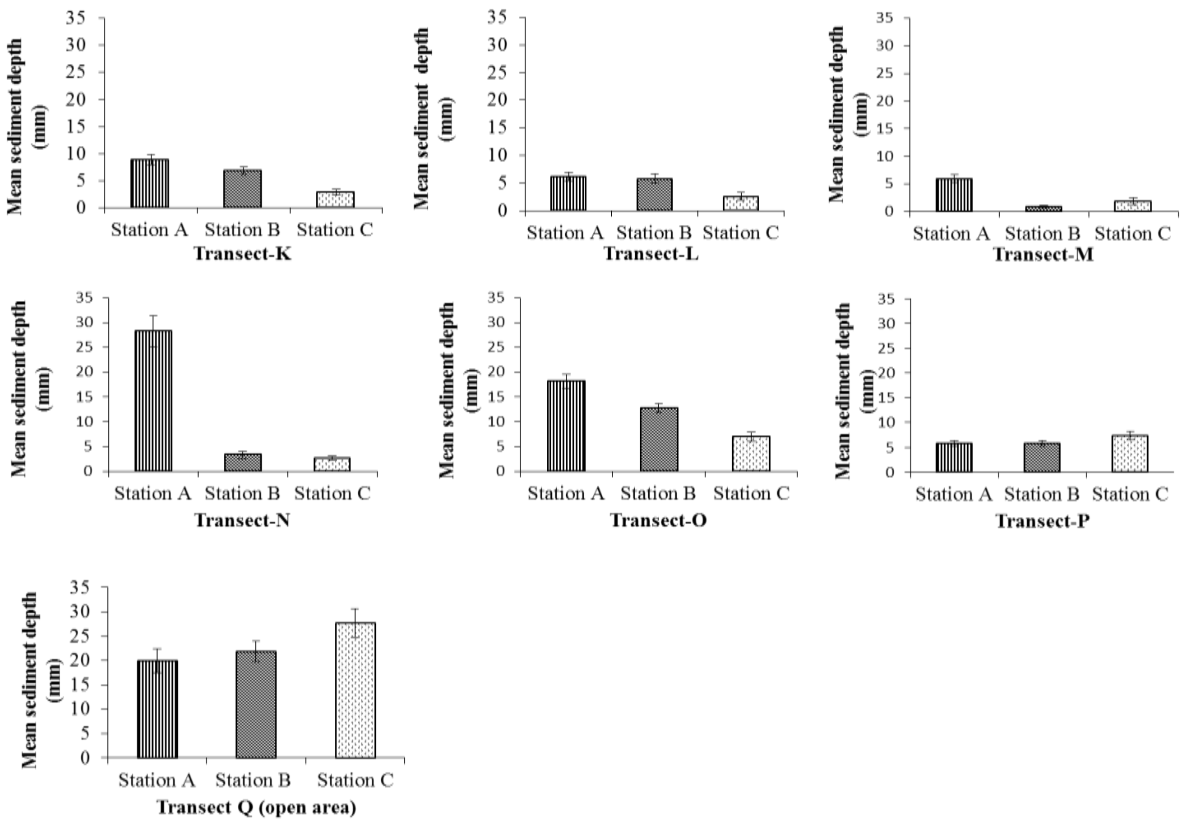


Figure 9. Mean values (± SE) of sinking depth of metal discs across station A, B, and C along the seven transects

Discussion

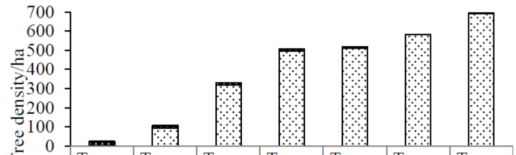
Provision of natural defenses by mangroves to both human beings and artificial structures along the coast has in the recent past been given a lot of attention following the lessons learned during the 26th December 2004 tsunami in Asia. Many degraded mangrove sites have witnessed accelerated shoreline erosion along the Kenya coast, prompting the government and local communities to put up artificial structures to mitigate the problem with little or no success. However, the restoration attempt guided by scientific findings has provided a lasting and sustainable solution by restocking the degraded sites with suitable species. Increasing the mangrove tree densities has led to stability and functioning of the ecosystem by reducing shoreline erosion and binding of sediment by mangrove forest structures.

Effect of tree and pneumatophores density on the wave energy

This study demonstrated that wave energy was gradually reduced along 80 m transects with different tree and pneumatophore m densities. The data was obtained by comparing dissolution rates of percentage loss of clods card as a proxy to wave energy attenuation across the reforested mangrove forest. The energy reduction by tree stems alone was 7.8%, while pneumatophores' energy reduction was 5%. In total, energy reduction by the two mangrove forest structures was 12.8%. Similar studies in Vinh Quang village and Red River (Song Hong) Vietnam showed that 100 m of intact mangrove attenuate wave energy up to 45% (Mazda et al. 1997). This study has proved that mangroves are suitable for coastal vegetation defense to protect shoreline erosion against average wave energy.

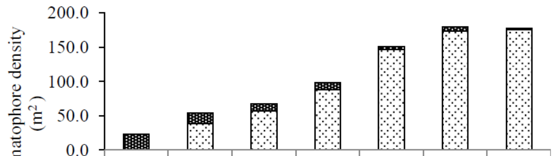
The clod cards exposed to water current in the sea were used to express a loss of their weight as a function of water velocity since plaster of Paris objects erodes with the friction with the water. The higher the water velocity, the higher the clod card weight loss. Therefore, it is interesting to compare the relationship between weight losses of the clod cards versus tree and pneumatophores density along transects of the known distance across the mangrove forest. Change in wave energy was plotted as the weight loss of clods (in the y-axis) versus transects with varying pneumatophores density and tree density (in the x-axis). The regression slope was the change in wave energy as tree and pneumatophores density varied among transects.

High wave energy was recorded at 15 cm above the substrate as demonstrated by high clod card weight loss at this height with a mean of 78.3 ± 2.3% (SE) compared to the means of clod cards mounted at 50 cm, which recorded 73.4 ± 2.4 in transect that had mangrove trees. A similar power of high wave energy was experienced at 15 cm above the substrate in the control transect without mangrove cover, registering a mean of 84.0 ±2.9% (SE). In comparison, the energy sampled at 50 cm was 81.2 ± 2.9% (SE). However, the capacity to reduce wave energy depends on forest width, tree density, and tree species (Alongi 2009). Mangrove reforestation initiatives at Gazi have seen an increase in tree density of fringing *Sonneratia alba* from diminishing levels to 448 ±212.3 (SD) at the time of the study. In this study, the site comprised of one species (*Sonneratia alba*), the semi-pristine nature of the forest stands coupled with narrow width limited the ability to reduce considerable wave energy contrary to Vinh Quang village along with Red River (Song Hong) Vietnam with pristine fringing mangrove (Mazda et al. 1997).



	Transect Q	Transect O	Transect K	Transect N	Transect P	Transect M	Transect L
Mean sediment depth (mm)	23.18	15.63	10.21	9.43	6.30	2.91	4.83
tree density/ha	0	92	320	496	510	580	690

Figure 10. Mean of sinking depth of metal disks against; Tree density/ha across fringing mangrove forest



	Transect Q	Ttransect O	Transect K	Transect N	Transect L	Transect P	Transect M
Mean sediment depth (mm)	23.18	15.63	10.21	9.43	4.83	6.30	2.91
Pneu. Density/square meter	0.0	38.4	57.2	88.6	146.3	173.7	174.9

Figure 11. Means of sinking depth of metal disks against pneumatophores density/m^2 across fringing mangrove forest

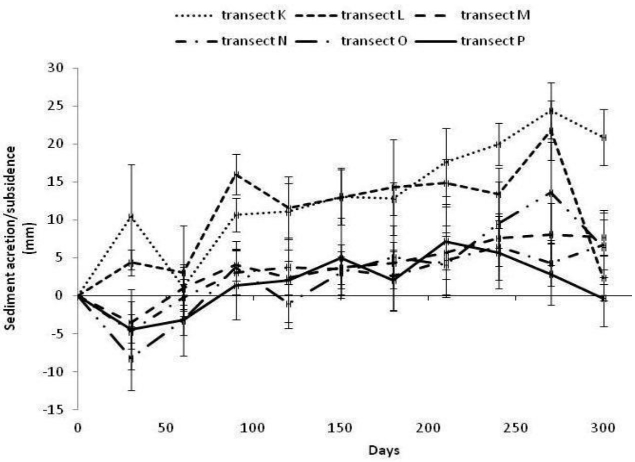


Figure 12. Mean values ± SE of sediment accretion rate in study area observed for 330 days

The effect of tree and pneumatophores density on wave energy reduction among different transects demonstrated that increasing tree density enhances the frictional drag by the mangrove forest structures. Pneumatophores of *Sonneratia alba* taper off upwards, thus reducing their efficiency to maintain high drag force, explaining why more wave energy was experienced at 15 cm due to pneumatophore influence than energy sampled at 50 cm due to mangrove tree density (Mazda et al. 1997).

Effect of tree and pneumatophores density on sediment stability and accretion

Sinking depth of metal disks along transects with different tree densities demonstrated that sediment gains stability with increasing tree and pneumatophores density. The deeper the metal disks sunk in the sediment, the more unstable the sediment was. Therefore, this study showed sinking depth of metal disk reducing gradually with distance from the open areas on the seaward side towards the shoreline as tree and pneumatophores densities varied among transects. The trends in sediment stability demonstrated in this study are similar to those reported in mangroves along the Cairns boardwalk in wet tropical Australia within a fringing *Rhizophora* sp. (Furukawa and Wolanski, 1996). The sediment transport mechanisms in both sites are dominated by hydrodynamics from waves, and sediment gained stability with increasing distance from the seaward side towards the shore. The sediment particles settled as wave energy-reduced across the mangrove forest, demonstrating the net effect of forest width and tree density on sediment stability as reported elsewhere in the world (Alongi 2009).

Where mangrove tree stems and pneumatophores densities were high, the depth of sinking metal disks in the sediments were less due to reduced wave energy, thus facilitating trapping and holding of sediment. The trapping mechanism resulted from high micro-turbulence created by the flows around the trees structures such as tree stems and pneumatophores (Wolanski 1995; Mazda et al. 1997) during flood tide and settled down at slake water when current flows are low during ebb tide.

The study showed station all A (before the mangroves) in all forested transect having the highest means of sinking depth of metal disks indicating high sediment instability, which decreased with increasing distance across station B through station C. The observation witnessed in this study on sediment stability is accurate of studies done by Balke et al. (2013), who found sediment stability rates increasing with distance from the seaward side towards the shore. In the control transect without mangrove, there was less micro-turbulence due to the absence of mangrove, trees thus making the sinking depths of metal disks increase with distance along with the intertidal complex, an indication of high sediment instability.

The study has demonstrated that degradation of fringing mangrove forests alters sediment stability due to alteration in their physical roles, a condition that has been witnessed elsewhere in the world (Furukawa and Wolanski, 1996). Graphical relationship of sinking depth of metal disks against tree and pneumatophores density showed the mean depth sinking metal disks decreasing with increasing tree stems/ha and pneumatophores density/$m^{2,}$ highlighting that the replanted fringing forest remains the best option to stabilizing the highly eroded study site at Gazi Bay.

The levels of sinking depth of metal disks improved with reduced exposure to strong wave energy with mangrove forest structures offering a bio-shield by creating frictional drag as reported elsewhere (Furukawa and Wolanski, 1996). In this study, high wave energy was observed from transect without mangrove cover (Control transect) and reduced gradually to reasonably high forested transect. Reducing wave energy levels by mangrove forest structures (tree and pneumatophores density) improves sediment stability along with the inter-tidal complex. Also, transects with high organic matter content and reasonably high silt levels had a low sinking depth of metal disk, indicating improving sediment physio-chemical characteristics (Macnae 1968). This condition makes the mangrove forest a fully functional system (Bosire et al., 2008).

The study showed sediment accretion rates of 5.4 ± 1.7 mm/year^{-1} (SE), which is more or less similar to that reported in Kosrae (FSM) at Yale and Utwe river basin of 4.5 ± 1.1 and 9.1 ± 2.1 (SE) mm, respectively. Various levels of sediment accretion or subsidence due to the influence of mangrove forest structures such as tree and pneumatophores densities using sediment elevation tables (SETS) method have been reported in many studies worldwide.

The stems and pneumatophores create frictional drag-reducing the current flow from wave energy, thereby creating eddies that encourage sedimentation when cohesive particles flocculate mainly due to the presence of clay particles (Furukawa et al. 1997). Our study has shown that reforestation efforts in this site have boosted the regulatory services offered by mangroves by modifying the physical characteristics of the sediment. The phenomena have been observed in transects M, K, and L, which expressed the highest clay particles, which bind sediment together, thus supporting the sediment accretion. Transect with the smallest clay particles as witnessed at transect O, which had the lowest tree and pneumatophores density, expressed the least mean accretion rates. Thereby, the study shows that by increasing tree stems through reforestation, sediment is less prone to resuspension by waves, thus reducing shoreline erosion compared to other earlier efforts of mitigating shoreline protection through the construction of gabions.

Conclusion

Using the inexpensive clod card dissolution method to quantify wave energy attenuation as a regulatory service, this study has established baseline data to help monitor and quantitatively estimate the role of mangrove forests in shoreline protection and how this changes with this tree density. Evidence of the study site regaining stability after replanting mangroves is a more sustainable and cost-effective method than putting up complex engineering structures. The approach can easily be replicated elsewhere in many sites along the Kenyan coast and western Indian

Ocean. Mangroves are potentially used as a bio-shield that shall provide other ecosystem benefits such as increased biodiversity aesthetic beauty. They will have potential application in disaster risk reduction in the long term. At the study site in Gazi Bay, mangroves proved to reduce on average 7.8% of wave energy in transects with trees compared to open areas without trees. In contrast, sediment stability correlated positively with an increase in tree and pneumatophores density as wave energy reduces. Stable sediments have shown accretion rates improve as the density of the forest increases over time through reforestation efforts and the shoreline erosion gradually decreases and resists further coastline changes. The primary challenge in the study was the lack of baseline data and other comparative studies on the use of mangrove forests as bio-shields to provide coastal protection against average wave energy impacts along the Kenya coast and the Western Indian Ocean region.

REFERENCES

Abuodha PAW, Kairo JG. 2001. Human-induced stresses on mangrove swamps along the Kenyan coast. Hydrobiologia 458 (1): 255-265. DOI: 10.1023/A:1013130916811.

Alongi DM. 2009. The Energetics of Mangrove Forests. Australian Institute of Marine Science, Townsville, Queensland, Australia.

Balke T, Webb EL, den Elzen E, Galli D, Herman PM, Bouma TJ. 2013. Seedling establishment in a dynamic sedimentary environment: a conceptual framework using mangroves. J Appl Ecol 50 (3): 740-747. DOI: 10.1111/1365-2664.12067.

Bell J, Lovelock CE. 2013. Insuring mangrove forests for their role in mitigating coastal erosion and storm-surge : An Australian case study. Wetlands 33 (2): 279-289. DOI: 10.1007/s13157-013-0382-4.

Bosire JO, Dahdouh-guebas F, Kairo JG, Koedam N. 2003. Colonization of non-planted mangrove species into restored mangrove stands in Gazi Bay. Aquat Bot 76 (4): 267-279. DOI: 10.1016/S0304-3770(03)00054-8.

Bosire JO, Dahdouh-guebas F, Walton M, Crona BI, Lewis RR, Field C, Koedam N. 2008. Functionality of restored mangroves: A review. Aquat Bot 89 (2): 251-259. DOI: 10.1016/j.aquabot.2008.03.010.

Brower JE, Zar JH, Carl N. 1990. Field and Laboratory Techniques for General Ecology. 3rd ed. Wm.C. Brown Publishers, New York.

Cintron G, Schaeffer-Novelli Y. 1984. Methods for studying mangrove structure. The Mangrove Ecosystem: Research Methods. UNESCO, Paris.

Crona BI, Holmgren S, Ronnback P. 2006. Re-establishment of epibiotic communities in reforested mangroves of Gazi Bay, Kenya. Wetlands Ecol 14 (6): 345-360. DOI: 10.1007/s11273-006-9005-7.

Dahdouh-Guebas F, Mathenge C, Koedam N. 2000. Utilization of mangroves wood products around Minda Creek (Kenya) among subsistence and commercial users. Econ Bot 54: 513-527. DOI: 10.1007/BF02866549.

Furukawa K, Wolanski E, Mueller H. 1997. Currents and sediment transport in mangrove. Estuar Coast Shelf Sci 44 (3): 301-310. DOI: 10.1006/ecss.1996.0120.

Furukawa K, Wolanski E. 1996. Sedimentation in mangrove forest. Mangroves Salt Marshes 1 (1): 3-10. DOI: 10.1023/A:1025973426404.

Gilman EL, Ellison J, Duke NC, Field C. 2008. Threats to mangroves from climate change and adaptation options : A review. Aquat Bot 89 (2): 237-250. DOI: 10.1016/j.aquabot.2007.12.009.

Giri C, Ochieng E, Tieszen LL, Zhu Z, Singh A, Loveland T, Masek J, Duke N. 2010. Status and distribution of mangrove forests of the world using earth. Glob Ecol Biogeogr 20 (1): 236-255. DOI: 10.1111/j.1466-8238.2010.00584.x.

GOK. 2010. National Climate Change Response Strategy. Ministry of Environment and Mineral Resources Nairobi, Kenya.

Hamsa J. 2013. Mangroves and Livelihood: An Assessment of Livelihood Project in Mangroves Ecosystem along The Kenyan Coast. [Thesis]. Vrije University Brussels, Belgium.

Kairo JG, Bosire J, Koedam N. 2001. Restoration and management of mangrove systems - a lesson for and from the East African region. South Afr J Bot 67 (3): 383-389. DOI: 10.1016/S0254-6299(15)31153-4.

Kairo JG, Lang JKS, Dahdouh-guebas F, Bosire J, Karachi M. 2008. Structural development and productivity of replanted mangrove plantations in Kenya. For Ecol Manag 255 (7): 2670-2677. DOI: 10.1016/j.foreco.2008.01.031.

Kairo JG, Wanjiru C, Ochiewo J. 2009. Net pay: Economic analysis of a replanted mangrove plantation in Kenya. J Sustain For 28 (3-5): 395-414. DOI: 10.1080/10549810902791523.

Kairo JG. 1995. Artificial Regeneration and Sustainable Yield Management of Mangrove Forests at Gazi Bay. [Thesis]. Botany Department, University of Nairobi, Nairobi, Kenya.

Keryn B, Gedan M, Kirwan L, Eric W, Edward BB. 2011. The present and future roles of coastal wetland vegetation in protecting shorelines: answering recent challenges to the paradigm. Clim Change 106 (1): 7-29. DOI: 10.1007/s10584-010-0003-7.

Kirui KB, Kairo JG, Bosire J, Viergever KM, Rudra S, Huxham M, Briers RA. 2012. Ocean coastal management mapping of mangrove forest land cover change along the Kenya coastline using Landsat imagery. Ocean Coast Manag 30: 1-6. DOI: 10.1016/j.ocecoaman.2011.12.004.

Kitheka JU, Ongwenyi GS, Mavuti KM. 2003. Fluxes and exchange of suspended sediment in tidal inlets draining a degraded mangrove forest in Kenya. Estuar Coast Shelf Sci 56 (3-4): 655-667. DOI: 10.1016/S0272-7714(02)00217-2.

Krauss W, Mckee KL, Lovelock CE, Cahoon DR, Saintilan N, Reef R, Chen L. 2013. How mangrove forests adjust to rising sea level. New Phytol 202 (1): 19-34. DOI: 10.1111/nph.12605.

Macnae W. 1968. A General account of the flora and fauna of mangrove swamps and forests in the Indo-West-Pacific region. In: Russel FS, Yonge M (eds) Advances in Marine Biology 6: 74-165. DOI: 10.1016/S0065-2881(08)60438-1.

Mazda Y, Wolanski E, King B, Sase A, Ohtsuka D, Magi M. 1997. Drag force due to vegetation in mangrove swamps. Mangroves Salt Marshes 1 (3): 193-199. DOI: 10.1023/A:1009949411068.

Mckee KL, Cahoon DR, Feller IC. 2007. Caribbean mangroves adjust to rising sea levels through biotic controls on change in soil elevation. Glob Ecol Biogeogr 16 (5): 545-556. DOI: 10.1111/j.1466-8238.2007.00317.x.

Neukermans G, Kairo JG, Koedam N, Neukermans G, Roosevelt AFD. 2008. Mangrove species and stand mapping in Gazi Bay (Kenya) using Quickbird Satellite Imagery. Spatial Sci 53 (1): 75-86. DOI: 10.1080/14498596.2008.9635137.

Skove MW, Vannini M, Shunula JP, Hartnoll RG, Cannicci S. 2002. Quantifying the density of mangrove crabs: Ocypodidae and Grapsidae. Mar Biol 141 (4): 725-732. DOI: 10.1007/s00227-002-0867-9.

Spalding M, Kainuma M, Collins L. 2010. World Atlas of Mangroves. Earthscan, London, UK. DOI: 10.4324/9781849776608.

Valiela I, Jennifer BL, Joanna YK. 2001. Mangrove forests: one of the world's s threatened major tropical environments. Bioscience 51 (10): 807-815. DOI: 10.1641/0006-3568(2001)051[0807:MFOOTW]2.0.CO;2.

Williams N. 2005. Tsunami insight to mangrove value. Curr Biol 15 (3): R73. DOI: 10.1016/j.cub.2005.01.015.

Wolanski E. 1995. Transport of sediment in mangrove swamps. Hydrobiologia 295: 31-42. DOI: 10.1007/978-94-011-0289-6_5.

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 95-107

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070206

The role of indigenous knowledge and practice in water and soil conservation management in Albuko *Woreda*, Ethiopia

ADDIS TAYE, TEBAREK LIKA MEGENTO♥
School of Graduate Studies, Addis Ababa University. Addis Ababa, Ethiopia. ♥email: tebroza@yahoo.com, tebarekl@yahoo.com

Manuscript received: 4 July 2017. Revision accepted: 11 October 2017.

Abstract *Taye A, Megento TL. 2017.* The *role of indigenous knowledge and practice on water and soil conservation management in Albuko Woreda, Ethiopia. Bonorowo Wetlands 7: 95-107.* The objectives of this study were to assess various aspects of indigenous knowledge and practice in water and soil conservation measures introduced in Albuko Woreda/district, South Wollo, Ethiopia, and investigate how farmers have adopted indigenous soil and water conservation measures. This study also aimed to examine farmers' views on land degradation and assess their adoption behavior of soil and water conservation knowledge. The data for the study originated mainly from farmers in the study area. Structured questionnaire survey and focus group discussion methods were applied to collect the necessary information from farm households. A total of 140 heads of families were questioned. The received data was analyzed using descriptive statistics and quantitative and qualitative data analysis. Even though some soil and water conservation methods were introduced to combat land degradation in Ethiopia, adopting these practices remains below expectations. Most farmers preferred soil (stone) bund, water diversion ditch and contour ploughing for soil and water conservation, crop rotation, and mixed cropping for soil fertility amendment measures instead of traditional cutoff drain and fallowing. Farmers faced several constraints in adopting soil and water conservation measures. The main problems related to conservation structures include the source of pests, inconveniency during ox ploughing, reduction of farmland, labor intensiveness, difficulty in implementation, and costliness. In contrast, farmers were knowledgeable about various indigenous soil and water conservation measures but implemented only some of them. They understand the effects of erosion on crop productivity. Finally, farmers need to be trained on the impact of soil erosion and available conservation measures. Information on the ineffectiveness of some traditional conservation measures has to be disseminated among farmers.

Keywords: Farmer, indigenous knowledge, land degradation, soil, and water conservation

INTRODUCTION

Culture and the respective systems of indigenous peoples have been widely misunderstood or even dismissed by development planning experts in the past. This issue was raised recently in a final statement by the Brundtland Commission (WCED): "Some traditional lifestyles are threatened with virtual extinction by insensitive development over which the indigenous people have no participation. Their traditional rights should be recognized, and they should be given a more decisive voice in formulating policies about resource development in their areas" (Our Common Future 1987).

Many scientific and social researchers associated with developing assistance policies are now beginning to recognize the decisive role that indigenous peoples and their knowledge of the ecosystem contribute to in the success of development projects and strategies (Lalonde 1991). It is reasonable to assume that essential global development assistance activities including; local participation, capacity building, and sustainable resource management can be enhanced in cost-effective programs and strategies that understand and work with indigenous knowledge and decision-making systems. Africa may be an ideal continent to learn about and begin thoughtfully integrating indigenous expertise with development planning techniques. Despite the rich diversity and number of indigenous cultural groups throughout Africa, much of the remaining areas of Africa today are threatened by growing environmental and social pressures. In this unexploited, remote, or sparsely populated area, the indigenous societies tend to live traditional lifestyles (Lalonde 1991).

Indigenous knowledge (IK) has attracted the interest of researchers nowadays. Accordingly, various investigators tried to recognize how conservation of natural resources such as water and soil can be done through indigenous knowledge of communities compared to modern ways. In this regard, (IIRR 1996) argues that IK is adapted to local culture and environment, based on experience, habitually established over centuries of use, dynamic, and changing. Likewise, Ajibade (2003) conceptualizes IK as it has been built upon and passed from one generation to generation and improves within a particular culture or ethnic group, and tries to continue objectives in a specific environmental background.

The densely residential areas of Northern Ethiopia are among those with the highest rate of soil loss because the environment is highly degraded compared to the Southern part of the country. The country's forest reserves are predicted to be 2.5-3% of the total land, where approximately 100,000 hectares of forest are lost annually. About 1 billion tons of topsoil are also believed to be eroded annually (Berisso 1995). In line with this, Tsighe

(1995) asserts that the average soil erosion is 42 tones/hectare/ year in the croplands.

It is well-known that soils with low fertility cannot allow sufficient crop cover to sustain life. Low humus content and erosion of such soil decrease infiltration and moisture-holding capacity of the soil. Thus, it is increasingly recognized that adequate conservation of soil resources is a precondition for sustainable rural development strategies, mainly in the highlands of Ethiopia. Most of the projects for soil conservation, however, planned at the center and implemented at the local level, show little attention to whether the local population could apply techniques on their farm fields. Similar to the above experience, farmers did not want to expand new methods to the rest of the farm fields at the end of the project. This is partially attributed to the costly nature and problem of adaptability of the latest technology otherwise not contextualized. Furthermore, planners in the area assume that the local population is responsible for the overall maintenance of the structures built by a project (Tsighe 1995).

Most highland areas of Ethiopia are characterized by high population, high rainfall, and sloppy lands. Maintaining the fertility of the productive soil and rehabilitating degraded arable lands are the primary concern of many stakeholders in highland areas (Amede et al. 2001).

Likewise, soil erosion is one of the major agricultural problems in the Ethiopian highlands. The highlands occupy 44% of the country's total area, 95% of the land under crops and 90% of the total population, and 75% livestock. Deforestation, overgrazing, and cultivation of slopes not suited to agriculture, together with the farming practice that does not include conservation measures are the primary causes for soil erosion in much of Ethiopia's highland areas. Population pressure and soil erosion in the regions are critical causes for declining arable lands. The decrease in the productivity of arable lands in the highlands is due to the washing away of the fertile topsoil by water erosion (Amede et al. 2001). Hence, this research assessed the indigenous knowledge applied in the Albuko *woreda* of the South Wollo.

The specific objectives of this study are to (i) identify the traditional and indigenous soil and water conservation practices in the *woreda*; (ii) examine the extent of indigenous water conservation measures (soil water conservation) practices as they appeared in the *woreda*; (iii) to assess the most commonly and rarely used indigenous practices of soil water conservation in the *woreda*; (iv) assess the socio-cultural, economic, biophysical system and constraints for the implementation and maintenance of indigenous soil and water conservation practices; and; (v) analyze farmers' knowledge towards determining the degree of land degradation in the *woreda*.

MATERIALS AND METHODS

Description of the study area

Location

The Amhara national, the regional state, is located in the country's central, northeastern and northwestern parts, between 9^o-13^o 45''N and 36^o-40^o 30''E (Figure 1). According to the latest GIS mapping, the Amhara regional state covers about 157,647 km^{2,} or around 14% of the country's land area. The highlands of the Amhara region (>1500 m asl.) account for 73% of the region's land area, with the lowlands sharing 27% (MoA 2002).

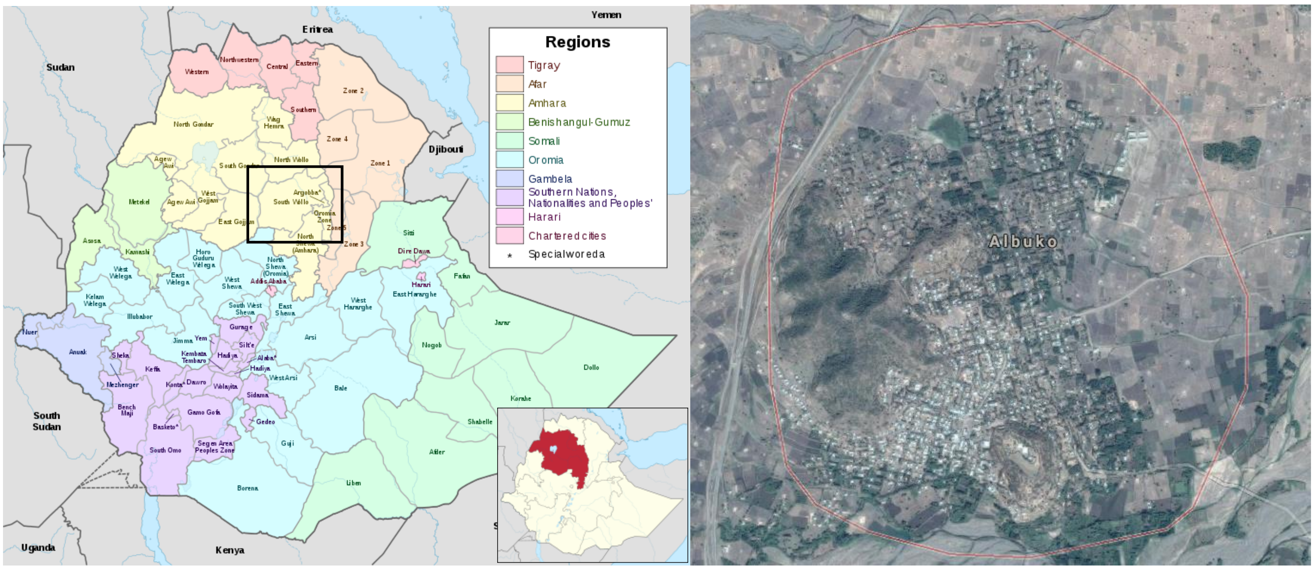


Figure 1. Study site in Albuko *Woreda*, South Wollo, Amhara, Ethiopia

Albuko *Woreda* is situated in the northeastern highlands of the Amhara region, at a distance of about 37km from Dessie in the southeast direction, on 11000` N and 39039` E. Weyna Dega defines the characteristic of the agro-climatic zone to which the site belongs to Dega. The range of altitude lies from 2530-2860 m.a.s.l. The primary type of soils in the area are Phaeozems/Lithosols 56%, Phaeozems 37.6%, Gleysols 2.5%, Fluvisols 2.2%, and Regosols 1.7% coverage. The catchment's mean annual (1982-1989, 1992-1993) precipitation is 1211 mm with a bimodal characteristic regime. The mean yearly erosivity amounts to 4267 MJ mm ha-1 hr-1 (Bosshart 1999; SCRP 2000). The catchment is low-potential, intensively cultivated, and part of the ox-plow cereal belt in the highlands of northeastern escarpment (SCRP 1982).

Population

According to figures published by the Central Statistical Agency in July 2011 (CSA 2011), this *woreda* has an estimated total population of 82,311, consisting of 41,010 males and 41,301 females. Albuko has a population density of approximately 172.7 people per square kilometer from an entire area of 476.68 square kilometers, which is higher than the Zonal average of 158.7 and the regional average of 119.9 people per square kilometer.

Topography

According to the Geological Map of Ethiopia (Ministry of Mines 1973, in Tesfaye 1988), the research area consists of alkaline olivine basalts and tuffs of the Ashangi Groups (Paleocene-Oligocene-Miocene), which are part of the volcanic trap series. The area is part of the Afar Margin, and the tectonics is dominated by graben faulting. Results of Weigel's (1986) soil survey demonstrated that the Albuko area is dominated by steep (25-55%) and very steep (more than 55%) slopes, which cover two-thirds of the survey area.

Temperature

The minimum air and soil surface temperatures were 3-4°C, and the maximum was 28-35°C, respectively (SCRP 2000). April to June are the warmest months with a mean monthly minimum soil temperature of 12.0°C. The maximum soil temperature goes up to 16°C between November and January.

Rainfall

According to SCRP (2000), the rainfall of the research area is mainly influenced by the Inter-Tropical Convergence Zone (ITCZ) and the sub-tropical pressure cells, resulting in the area's annual rainfall variable. The primary rainy season (*Kremet*) occurs typically from June to October, with the peak rainfall in August, then declining markedly in September and October.

According to monthly rainfall distributions, Albuko is bimodal, having *Belg* (slight rain between February and May) and *Kremt* (the primary rainy season between July and September). The rainfall distribution during the *Belg* season is between 69.5 mm and 112.7 mm, with peak rainfall in April and peak erosivity of 658 MJ mm ha-1 in March. The rainfall distribution during the *Kremt* season is between 140.9 mm and 287.8 mm, with peak rainfall in August and a peak erosivity value of 905 MJ mm ha-1.

Farming system

Small-scale farming is considered the most prevalent method in the catchments for maintaining a subsistence economy at the household level. Farmers usually own draught animals. Thus, plowing is traditionally done with the traditional tool called *maresha*. The crops are produced during two cropping seasons, *meher* and *belg*. During belg season, the most important crops are barley, whereas emmer wheat, maize, horse bean, and *teff* are the most important ones in the *meher* season. No fixed cropping sequence is employed because each plant is grown on suitable sites. Planting is started immediately after the first adequate rainfall in the rainy season on the land that has been plowed. Different tillage and land preparation requirement before cultivation is needed for each crop. Cereals require the highest number of plowings, whereas pulses are plowed only once or twice. Seedbed which prepared for *teff* plantation needs some tillage frequency than others. Fields are left to lay fallow in areas where alternative fields are available and when the soil is depleted (Hanggi 1997).

Soil and water degradation

Water and soil degradation, recurrent drought, small farm plots, high population density, and input shortage, including draught animals and improved seed, are the primary agricultural problems of the *woreda*. These agricultural production problems are enhanced with inadequate research technology and extension support delivery. Cash income for household financial requirements is mainly generated from the sale of livestock and crop products. Households facing seasonal food shortages receive cash or food transfer, either "for work"(through a public work program to employ beneficiaries in soil water conservation works, building roads and other infrastructures) or "for free," from productive safety net program (PSNP).

Types of data and source

This study is based on data obtained from primary and secondary sources. The literature review entirely depends on secondary sources. All analysis depends on primary and secondary data, collected through unstructured personal interviews and questionnaires from randomly selected households, and secondary data obtained from various published and unpublished sources of the governmental and non-governmental organizations. Books, journals, internet sources, research reports, archives, and records were employed for acquiring the necessary information.

Household survey

A formal survey questionnaire was used to gather primary household data at the village level. Preliminary data was also generated by interviewing local extension agents. Also, direct field observations and some informal discussions with farmers groups, village elders, and extension workers were conducted to verify information of interest to this study. This questionnaire was prepared, tested, and amended to fulfill the objectives of the present study.

Sampling technique

If resources and time are available, the entire population should be studied to provide the most accurate results. Nevertheless, resources are never abundant, and time is never unlimited. This urges for taking samples from the population. This method is a convenient and cost-saving approach to a research study (Asraf and Brewer 2004).

In the study area, there are 14 *kebeles (lowest administrative units in Ethiopia)*. All of them are identified and listed down from which three sample *kebeles*, namely Maybar, Soba, and Tulutosigne, were selected using the purposive sampling method. The total households of Maybar, Soba, and Tulutosigne *kebeles* were 612, 693, and 711, respectively (Table 1). Seven percent of each *kebele*'s households were selected using a simple random sampling technique, making the total sample 140.

Given the relative homogeneity of the subsistence farmers in the study villages (*kebeles*) regarding physical environmental factors and resource endowments, this number was considered maximum, which could be efficiently handled within the research time and budget.

Interview

Interviews at the household level were carried out by going to each interviewee's homestead by the researcher. A randomly selected sample substitute was included in cases where the selected interviewees may not be interviewed for various reasons. The purpose of this interview was to acquire primary household data. Informal discussions with village elders and farmers groups were conducted using a checklist of topics to guide the sessions. This interview was conducted to cross-check household data and obtain reliable data for the entire study.

Key informants interview

The final level of the informal interview was conducted with village extension workers. The interviews were intended to gather information about the type of soil conservation and fertility practice, farming practices, extent of using market inputs such as fertilizers, improved seeds, and the nature of extension services offered and to establish a communication gap between farmers and extension workers.

Table 1. Total household heads of the three *kebeles* and sample size taken

Study villages (*kebeles*)	Average elevation (m asl.)	Climatic zone	Total no. of households	Sample size (7%)
Maybar	2530-2860	Temperate	612	42
Soba	2530-2860	Temperate	693	48
Tulutosigne	2530-2860	Temperate	711	50
Total			2016	140

Focus Group Discussion

Focus group discussions with six to eight discussants in each group were carried out on different local people's indigenous soil conservation mechanisms issues. The discussions focused on native and modern soil conservation mechanisms.

Data analysis

The survey generated qualitative and quantitative data; the first task was to summarize, categorize, and code all qualitative responses into numeric values and then enter them into the SPSS statistical program. The quantitative data was for analysis of various parameters.

Information obtained from unstructured interviews and informal interviews with different seniors in the village and extension officers was mainly verbal/narrative information. These were written down during the survey and summarized. This information is more qualitative and supports the coded qualitative and quantitative data analysis. Descriptive statistics such as sum, mean, and percentages are presented in tables to enable interpretation and quick visual comparisons of variables within the study area.

RESULTS AND DISCUSSION

Demographic characteristics

Family size of the households

As shown in Table 2, 38% of the respondents have 1-3 family size, 41% of the household heads have 4-6 family size and 15%, and 6% of the households have 7-9 and above nine family size, respectively. This shows that the household heads have a large family size that contributes to the positive impact on the soil water conservation practice in the *woreda*. Family size helps efficiently take on some soil and water conservation practice measures in their farmlands.

The mean household size is 6.03 persons. The average household size in Maybar, Soba, and Tulutosigne village are 5.5, 5.9, and 6.7, respectively. The household size refers to the number of individuals currently living under one roof, excluding children of some households who may have established their families.

Sex of the household members

The entire population of the 140 households is 844, consisting of 416 males and 428 females (Table 3). Of the whole 140 sample households, the result indicates that 49.3% of the household members are males. Comparing the regional figure based on figures published by the Central Statistical Agency in July 2011, this *woreda* has a total population of 82,311, of whom 41,010 were males, and 41,301 were females. A similar result is shown from the survey concerning male and female population proportion in the study area, specifically in three *kebeles*.

Age of the heads of household

As we can see from Table 4, 45% of the respondents were between 20-34 years old, 37% of the respondents between 35-49, whereas 10% and 8% of the household age group were from 50-64 years and more than 65 years old, respectively. This phenomenon reflects that almost all the household heads are adults and obtain enough experience in their lifetime. This might contribute to implementing the indigenous soil and water conservation programs. The mean household age is 42, similar to Maybar, Soba, and Tulutosigne villages.

Education level of the heads of household

Education is one of the fundamental human needs that all human beings deserve for the proper understanding of social, economic, political, and natural environments in which individual lives. In this study, concerning the educational background of the respondents, as we can see from Table 5, about 33.6% of the household heads are illiterate. Meanwhile, 42.9%, 15.7%,7.1%, and 7% of the respondents have 1-4, 5-8, 9-12, and above 12 grades of education, respectively. The above result indicates that most of the respondents have a low level of educational qualification that might become a challenge for adopting new soil water conservation technologies.

Landholding

Land availability often affects farming practice and the land degradation process. The majority of the agricultural land in the study area has been subdivided into the smallest landholdings, which are no longer economically viable for smallholders' subsistence. Farmers in the study cannot expand landholdings because the frontier is limited. Furthermore, the availability of arable land has shrunk dramatically from 0.42 to 0.23 ha per capita in the past 30 years (CSA 2005).

As Table 6 shows, almost all respondents have their farmlands. Nevertheless, the farmland size of the first 27 respondents is decreasing from year to year. The main reason for the reduction of their farm size is attributed to land degradation (presented in Figure 2) and need for housing by themselves for their children, and unanticipated road construction by the government. On the other hand, 69.3% of the respondents said their farmland is scattered. Regarding farmland cultivation, 76.4% of the respondents agree that they are cultivating their farm, whereas the rest, about 23.6%, are not developing all of their lands. The researcher took a photograph to show the degraded land (which is described by the farmer as one of the reasons for farmland size decreasing) in the *woreda* (Figure 2).

It is apparent from the preceding observation that there is intense pressure on the villages' land due to limited arable and grazing land, increasing family size, and lack of off-farm employment. Rapid population growth and limited arable land have increased pressure on the forest and grazing land resources besides aggravating soil degradation via extended farming into steep and marginal areas, shortened fallow system, tilling more frequently, and unchecked depletion of the inherent soil nutrients (Gizachew 1995).

In the study area, farmers cannot expand landholdings because the frontier is limited, and the availability of arable land has shrunk dramatically from time to time. Farmers' responses also revealed the existing land shortage in the study area. As we can see from Table 7, each household has an average of 0.79 ha of land. From 140 total interviewed farmers, more than 80% of households reported that their current landholdings are too small compared to the area needs of the family, and they are not in a position to inherit land to their children. Young male farmers inherited property from their parents when they got married. However, with married children living with parents until their death, multigenerational households have become more common as household landholdings have decreased. As shown in Table 6, over 90% of sampled households have less than 1.0 hectares of land.

Table 2. Family size distribution of respondents (N=140)

Family size	Maybar		Soba		Tulutosign		Total	
1-3	16	38	19	40	18	36	53	38
4-6	17	41	21	44	20	40	58	41
7-9	6	14	6	12	9	18	21	15
>9	3	7	2	4	3	6	8	6
Total	42	100	48	100	50	100	140	100
Mean	5.5		5.9		6.7		6.03	

Table 3. The demographic composition of the sample households, number of individuals by sex

Sex	Maybar		Soba		Tulutosign		Total	
Male	125	51.4	137	48.8	154	48.1	416	49.3
Female	118	48.6	144	51.2	166	51.9	428	50.7
Total	243	100	281	100	320	100	844	100

Table 4. Age distribution of respondents (N=140)

Age	Frequency	Percent	Mean of household age
20-34	63	45	42
35-49	52	37	
50-64	14	10	
>65	11	8	
Total	140	100	

Table 5. Educational level of the sample respondents (N=140)

Educational level	Freq.	Percent	Valid percent	Cumulative percent
Illiterate	47	33.6	33.6	33.6
1-4	60	42.9	42.9	76.4
5-8	22	15.7	15.7	92.1
9-12	10	7.1	7.1	99.3
>12	1	.7	.7	100.0
Total	140	100.0	100.0	

Table 6. Information on land ownership, scattered land, and cultivation status

Response of household	Own farmland		Scattered farmland		Cultivate farmland		Change on the size of farmland	
	Freq.	Percent	Freq.	Percent	Freq.	Percent	Freq.	Percent
Yes	140	100	97	69.3	107	76.4	27	19.3
No	0	0	43	30.7	33	23.6	113	80.7
Total	140	100	140	100	140	100	140	100

Table 7. Landholding of the household in Albuko *Woreda*, Ethiopia

Size of land in Hectare	Frequency	Percent	Mean	SD
0-0.2	22	15.7	0.79	0.34
0.3-0.5	10	7.1		
0.6-0.8	60	42.9		
0.9-1	47	33.6		
>1	1	.7		
Total	140	100.0		

Cropping system

As suggested by Table 8, the major crops grown in the area are barely wheat, bean, *teff*, pea, and maize. Traditionally, in the study significant, the major cereals grown by most of the farmers are barely wheat or bean, which accounts for 30.7%, 23.6%, and 16.4%, respectively. By referring to Table 8, we can conclude that the area is a temperate zone.

The most common feature of the cropping systems in the study area is crop rotation, which is practiced widely. Crop rotation is done by growing two or three different crops on the same field, one after the other and within a time frame of a year or two or three years, repeating this rotation over and over again. In the agricultural sense, this is practiced to help the soil recover after growing one crop, and if fallow is used in the turns instead of a plant, it does so even more.

As we can see from Table 9, about 24% of the respondents reported that they practice changing to another crop each year. Meanwhile, the rest of the respondents said that they did switch to other crops and fallow (16.3%), practice fallow (9.3%), and plant the same crop each year (6.4%). Out of interviewed farmers, 43.7% of the respondents practiced intercropping. Intercropping, mainly utilizing legumes, is a deliberate measure to maintain soil fertility. For example, in all *kebeles*, intercropping always involves incorporating a legume, such as a bean, along with other crops. Legumes emphasize enhancing soil fertility since inorganic fertilizer use has fallen drastically because of the high prices. To practice such a system, different crops are intercropped, including maize + bean, maize + potato, maize + cabbage, and bean + potatoes mainly are mostly two practical advantages of the intercropping system: mixing legumes with a grain crop, especially maize. Firstly, legumes are nitrogen-fixing plants; therefore, by intercropping the two, farmers do not even have to apply fertilizer or reduce it since most of them cannot afford to buy fertilizer. Lastly, legumes are a cover crop, so they suppress the growth of weeds and minimize the problematic task of weeding.

Figure 2. Degraded land in Albuko *Woreda*, South Wollo, Amhara, Ethiopia

Table 8. Major crops are grown in order of importance by the farmers in Albuko *Woreda*, Ethiopia

Types of major crops grown in order of importance	Frequency	Percent
Barely	43	30.7
Wheat	33	23.6
Bean	23	16.4
Teff	17	12.1
Pea	13	9.3
Maiz	11	7.9
Total	140	100

Table 9. Crop rotation practice by the farmers in Albuko *Woreda*, Ethiopia

Crop rotation practice	Yes		No		Percent	
	Freq.	Percent	Freq.	Percent	Freq.	Percent
Intercropping	61	43.7	79	56.3	140	100
Change to other crops each year	34	24.3	106	75.7	140	100
Change to other crops and fallow	23	16.3	117	83.7	140	100
Practice fallow	13	9.3	127	90.7	140	100
Plant same crop each year	9	6.4	131	93.6	140	100

Major uses of crop residue

As indicated in Table 10, the use of crop residues for livestock feeding was the most common practice in the study area. The Researchers' discussion with farmers and extension agents indicated that crop residue from small cereals (wheat, barley, and *teff*) and legumes (beans and faba bean) are taken from the crop field to the home or villages and stored for animal feed due to high animal feed shortage. In addition, maize and other stalk are mainly used for fuelwood and fencing, respectively. Animals grazed the leaf and leaf sheath right in the field. This practice shows that soil fertility declines most rapidly in the primary fields because crop residues from these regions are used for livestock feed. In contrast, animal manure is used only to maintain soil fertility.

Another potential measure to reduce soil/nutrient loss is mulching, covering the soil surface with crop residues. Through mulching, the hydraulic force of the raindrop on the soil particle will be reduced. Thus, soil detachment is minimized, but mulch is not applicable in the study villages' farming systems since the farmers use crop residue for different purposes, described in Table 10.

Soil fertility amendment methods

As shown in Table 11, almost all farmers apply inorganic fertilizer and farmyard manure to improve soil fertility in the study *kebeles*. This could explain the perception that the effect of soil fertility loss is a decrease in crop yields. It also demonstrated the deep concern to achieve better returns. Table 11 shows that soil fertility measures are being practiced. As we can depict from Table 11, about one hundred percent of the interviewed farmers used farmyard manure, but only to maintain soil fertility of homestead gardens in all study *kebeles*. The primary land-use systems in the community include homestead farms, where the most important crops such as wheat, *teff*, barley, and vegetables are grown.

Inorganic fertilizer

All of the interviewed farmers used chemical fertilizer as the sole source of improving soil fertility of outer fields where cereal and pulse crops are grown, as is depicted in Table 11. Significantly few farmers tended to use a combination of organic and inorganic fertilizers instead of purely relying on inorganic fertilizers, as was more familiar with the majority of farmers.

Table 10. Uses of crop residue by the farmers in Albuko *Woreda*, Ethiopia

Uses of crop residue	Yes		No		Total	
	Freq.	Percent	Freq.	Percent	Freq.	Percent
Animal feed	140	100	00	0	140	100
Fuelwood	140	100	00	0	140	100
Fencing	63	45	77	55	140	100
Burn	33	23.6	107	76.4	140	100
Other uses	5	3.6	135	96.4	140	100

Table 11. Soil fertility measures being practiced in Albuko *Woreda*, Ethiopia

Practices	Practiced		Percent		Total	
	Freq.	Percent	Freq.	Percent	Freq.	Percent
Use of farmyard manure	140	100	00	0	140	100
Use of inorganic fertilizer	140	100	00	0	140	100
Crop rotation	107	76.4	33	23.6	140	100
Intercropping	75	53.6	65	46.4	140	100
Agroforestry	63	45	77	55	140	100
Use of compost and mulch	57	40.7	83	59.3	140	100

Through discussion with local extension agents and farmers, we proved that the progressive increment of fertilizer cost after subsidies were being phased out was a major bottleneck to use an equal amount of fertilizer as farmers had before. However, in small-scale farming systems like the study area, where inorganic fertilizers are too expensive and unaffordable, the combined application of organic fertilizers with a small number of mineral fertilizers was found to be the promising route to improve the efficiency of mineral fertilizers and to achieve higher yields in smallholder farms. Long-term trials conducted in Kenya on organic and mineral fertilizer interaction also showed that maize grain yield was consistently top for 20 years in plots fertilized with mineral NP combined with farmyard manure than plots with sole mineral NP or farmyard manure (Palm et al. 1997).

One potential source of organic fertilizer is farmyard manure. Fortunately, in all *kebeles*, the availability of cow dung is much better than the abundant firewood found in the study area. However, as indicated above, farmers were not using manure for soil fertility replenishment of crop fields. The reason could be partly due to the lack of relevant knowledge regarding the combination of judicious use of mineral fertilizers and improved organic residue management via composting and application of farmyard manure.

Improving the content of organic matter of crop fields is very crucial to slow down the present trend of land degradation and the accompanied low productivity. Farmyard manure has a long residual effect and does not have to be applied every year. Farmyard manure could help trap the chemical fertilizer and make it more useful. Hence, increasing the supply of farmyard manure is a viable option in restoring soil fertility and enhancing soil organic matter among smallholders in the study area. Increased organic matter content will improve the efficiency of inorganic fertilizers through increased water availability, reduced nutrient loss from leaching and desertification, and increased microbial activity (Palm et al., 1997).

Agroforestry

As we can see from Table 11, one can conclude that the findings of this study also suggest that agroforestry is partially used as a means of maintaining soil fertility in the study area. In the study *kebeles*, scattered trees on cropland are also found, but the trees are widely spaced and probably have little effect on soil fertility maintenance. Agroforestry could play a potentially valuable role in enhancing soil organic matter and improving land productivity (Shibru 2010).

Soil organic matter helps retain essential nutrients, improves infiltration and water-holding capacity, and reduces erosion. Even when inorganic fertilizer is available, a minimum amount of organic matter is required for its efficient use. Given that external inputs may continue to be unaffordable, agroforestry could be useful in maintaining soil fertility. But further investigation is merited to establish agroforestry as a profitable investment and the best alternative for the sustainable increase of land productivity (Shibru 2010).

Livestock production

The type and the total number of livestock owned across all sample households are given in Table 12. Sample farmers rear livestock for various purposes, including draught power, milk, meat, eggs, transport, and other objectives. The primary feed sources for livestock in the study area include straw, grazing land, and maize stalk during its vegetative stage.

Livestock is an integral part of the farming system. As one can see from Table 12, every household has an ox, 2 sheep, and 2 chickens per head. In all *kebeles*, cattle are as much important. Oxen provide the significant power needed for farming. Sheep, goats, and poultry are substantial sources of cash and food. The few horses, mules, and donkeys available give the service of locomotives to transport goods and people.

Ownership of the livestock is also used to measure the wealth status of households. Hence the social stigma is attached to the number of animals owned regardless of economic value and the feed shortage.

As shown in Table 13, none of the respondents have their private grazing land. However, all respondents have communal grazing land for their lives- stocks. The reason for not having hay as their animal feed is that they prefer selling it and replacing it by buying the cheap type of animal feed to have lots of amount in feeding their livestock. As many as 37% of the respondents reported not using hay as their animal feed. Instead, crop residue is the most popular animal feed.

There is also a severe shortage of on-farm grazing land. For example, 71% of sampled households reported that grazing areas on their farm were decreasing because of farmland expansion. Respondents (68%) reported that grazing land was needed for increased crop production and tree planting. In contrast, the rest of the respondents (5.9%) said decreasing their grazing land for their house construction, and 9% did not know the actual cause (Table 14).

Despite the chronic feed shortages and grazing land scarcity, few farmers reported the adoption of more intensive methods of fodder production. This situation indicates the need for an active extension service to improve the current livestock management system. Cash income for household financial requirements is mainly generated from the sale of livestock and crop products. A limited number of households make off-farm income. For example, only 13% of all homes surveyed were engaged in off-farm activities. Hence, even though agricultural land is under intense pressure, a lack of alternatives that help to absorb the excess workforce and reduce the burden on the ground would likely worsen the problems of land degradation and nutrient loss on arable land. Also, the lack of non-farm activities can affect land management by the inability to fund the purchase of inputs and investments (Table 15).

Farmers' perception and attitude towards soil degradation

Farmers' perception of soil erosion as a potential hazard to agricultural production and sustainable agriculture is the most critical determinant of conservation measures. Those farmers who perceive soil erosion as a problem having adverse impacts on productivity and who expect positive returns from conservation are likely to adopt available conservation technologies (Gebremedhin and Swinton 2003). On the contrary, farmers will not benefit from controlling erosion when they do not acknowledge soil erosion as a problem. They will likely decide against adopting any conservation technologies.

Table 12. Type and number of livestock owned by the sample households (N=140) in Albuko *Woreda*, Ethiopia

Types of livestock	No. of animals	Mean
Oxen	157	1.1
Cows	130	0.9
Calves	163	1.2
Heifer	30	0.2
Horses	17	0.1
Mules	13	0.1
Donkeys	19	0.1
Goats	35	0.3
Sheep	223	2
Chicken	273	2

Table 13. Source of animal feed in order of importance in Albuko *Woreda*, Ethiopia

Animal feed	Yes		No		Total	
	Freq.	Percent	Freq.	Percent	Freq.	Percent
Communal land	140	100	00	00	140	100
Crop residue	140	100	00	00	140	100
Hay	103	73.6	37	26.4	140	100
Private grazing land	00	00	140	100	140	100
Other	97	69.3	43	30.7	140	100

Table 14. Trend of animal feed in Albuko *Woreda*, Ethiopia

Trend	Yes		No		Total	
	Freq.	Percent	Freq.	Percent	Freq.	Percent
Increasing	00	00	140	00	140	100
Decreasing by farmland expansion	99	71	41	29	140	100
Decreasing by both crop production and tree planting	88	62.8	52	37.2	140	100
Decreasing by housing	8	5.9	132	94.1	140	100
Do not know	13	9	127	91	140	100

Table 15. Livelihood of farmers in Albuko *Woreda*, Ethiopia

Livelihood	Yes		No		Total	
	Freq.	Percent	Freq.	Percent	Freq.	Percent
Off-farm activities	18	13	122	87	140	100
Sale of crop products	111	79.3	29	20.7	140	100
Sale of livestock	117	83.6	23	37.2	140	100
Both crop and livestock sale	8	5.9	132	94.1	140	100
Others	13	9	127	91	140	100

As shown in Table 16, only 35% of the sample farmers believed that overgrazing was the most severe cause of soil erosion, followed by 27% of the farmers who considered that deforestation caused the most erosion. Based on the result, we can say that overgrazing and deforestation are the most severe problems that can cause soil erosion in the selected study area. We only found that 17% of cultivation of steeply sloping land was the other cause that could challenge the loss of soil. Sixteen percent of the respondents answered that excess rainfall is the primary cause of soil degradation; in contrast, other respondents said that poor agricultural practices (15%), poor government policies (14%), and over-cultivation (13%) are the leading causes of soil degradation, respectively.

Chi-square tests on the effect of erosion on productivity (land suitability criteria by soil loss rate, soil fertility, and potential crop yield)

Concerning farmers' knowledge of land productivity and how it was affected by erosion, the sample respondents were questioned on what criteria they used to conserve and depict the type of suitable soils. So, to handle such an issue, a discussion with key informants was made and proved that farmers in the study area divided their land into several plots for various purposes. The farmer separates fields based on different critical criteria. For this study, three land suitability criteria were considered, i.e., soil fertility level, soil loss status, and crop yield production potential. In the study area, most field holdings tended to stretch from the very steep hill slope to gentle slope segments. Thereby, farmers were able to express their perceptions of each slope position.

The above result (Table 17) rejects the null hypothesis, suggesting that no causal relationship between land suitability and soil loss rate is denied based on the above chi-square test result. Ninety-five percent of the observed data proves a significant relationship between land suitability and soil loss rate in a 5% degree of error interval. The result indicates that land suitability dramatically affects soil fertility rates. Consequently, this result is congruent with what farmers knew about the speed of soil loss and level of slope position as positively related, suggesting that the level of soil loss rate is more significant when the slope of the land is very steep.

By examining the cross-tabulation of land suitability and soil fertility (Table 18), we reject the null hypothesis that suggests that land suitability does not affect soil fertility rate. However, the chi-square test proves otherwise. It means that land suitability dramatically changes the soil fertility rate. The chi-square test result shows that land suitability affects soil fertility by about 95%. This means, in a typical distributed data, the precision is 95% in a 5% error range. Therefore, this result is congruent with what farmers knew about the rate of soil loss and level of slope position as positively related. The finding suggests that the level of soil loss rate is very high when the slope of the land is very steep.

The cross-tabulation result of land suitability and crop yield (Table 19) rejects the null hypothesis, suggesting that land suitability does not affect crop yield. However, the chi-square test proves otherwise. This is to means that land suitability dramatically changes crop yield. To make it clear, the chi-square test result shows that land suitability affects crop yield by about 95%. This means, in a typical distributed data, the precision is 95% in a 5% error range. Therefore, this result is congruent with what farmers knew about the rate of crop yield and level of slope position as inversely related. This suggests that the level of crop yield rate is shallow when the slope of the land is very steep.

Table 16. The perceived major causes of soil erosion and their ranks in Albuko *Woreda*, Ethiopia

	Causes of soil degradation							
Rank	**Deforestation**	**Over grazing**	**Over cultivation**	**Agro poor practices**	**Steep slope cultivation**	**Excess rainfall**	**Poor policies**	**Others**
1st	27	35	13	15	19	16	14	3
2nd	8	9	45	26	31	18	3	0
3rd	31	12	17	27	23	19	11	0
4th	15	46	22	26	12	13	6	0
5th	19	11	15	26	17	35	12	5
6th	17	10	17	16	32	20	28	0
7th	23	17	8	4	8	19	33	28
8th	0	0	3	0	0	0	33	104
Total	140	140	140	140	140	140	140	140

Table 17. Chi-square analysis of land suitability by the soil loss rate

	Value	Df	Asymp. Sig. (2-sided)
Pearson Chi-Square	1.532a	2	.465
Likelihood Ratio	1.561	2	.458
Linear-by-Linear Association	.625	1	.429
N of Valid Cases	140		

Note**.** cells (.0%) have expected to count less than 5. The minimum expected count is 10.54.

Table 18. Chi-square analysis of land suitability criteria by soil fertility

	Value	Df	Asymp. Sig. (2-sided)
Pearson Chi-Square	3.911[a]	2	.141
Likelihood Ratio	4.312	2	.116
Linear-by-Linear Association	2.721	1	.099
N of Valid Cases	140		

Note: 2 cells (33.3%) have an expected count of less than 5. The minimum expected count is 4.64.

Table 19. Chi-Square analysis of land suitability criteria by crop yield

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	2.282a	2	.319
Likelihood Ratio	2.447	2	.294
Linear-by-Linear Association	1.689	1	.194
N of Valid Cases	140		

Note: 0 cells (0.0%) have expected count less than 5. The minimum expected count is 8.21.

The majority of farmers perceived that steep and very-steep slopes were landscape segments with a high risk of soil erosion and low soil fertility level, causing low crop yields. Gentle slopes ranked reasonably, showing moderate yield potential. The farmers’ description aligns with scientific knowledge that acknowledges the effects of slope steepness on land productivity. Other scientific experiments (Rockstöm et al. 1999) agree with farmers’ observation. The farmer’s knowledge is also aligned with findings by Steiner (1998) on farmers in Rwanda who associated soil suitability with slope position. Steeper slopes had shallower soils, whereas fine-textured soils dominated plateau and foot slopes, implying high fertility soils.

Soil and water conservation practices

In many parts of Ethiopia, farmers use native soil and water conservation practices as integral to their farming systems. Farmers have developed methods that have maintained productivity and contributed to long-term sustainability, while introduced measures have often been rejected or failed to achieve their technical objectives. Farmers have their criteria to evaluate the efficiency of soil water conservation measures. Their standards are often based on the quality of the natural resources (farmers are more likely to preserve those soils that will give the highest return on their investment), gender, the resource level of the household (particularly income levels and labor availability), education, cropping intensity as well as cultural traditions related to age. However, of great importance is the need to maintain or increase soil productivity.

The field survey results shown in Table 20 suggest that most of the farmers' indigenous knowledge recognized soil erosion problems and were willing to give their opinion about the conservation practice.

Drainage ditches, also known as traditional ditches, are widely used soil water conservation practices in the study area. Microchannels are constructed on cultivated farms to drain excess water and control soil erosion. Out of the total respondents, 20% applied indigenous drainage ditches. These are low-cost measures in which construction is part of the regular plowing activity. However, unlike the plow furrows, the ditches are made broader and deeper in dimension and usually run diagonally across the field. Farmers in the study area call the drainage ditches "*Feses.”*

Table 20. Indigenous knowledge in soil water conservation in Albuko *Woreda*, Ethiopia

Indigenous knowledge	Freq.	Percent	Valid Percent	Cumulative Percent
Traditional ditches ‘feses.’	28	20.0	20.0	20.0
Application of manure	17	12.1	12.1	32.1
Soil (stone) bund	29	20.7	20.7	52.9
Traditional cut-off drain ‘makor.’	11	7.9	7.9	60.7
Planting tree	18	12.9	12.9	73.6
Contour ploughing (curved plough)	25	17.9	17.9	91.4
Other	12	8.6	8.6	100.0
Total	140	100.0	100.0	

The application of manure in the study area is used by some farmers (12.1%) to improve the fertility of the soil. The best form of organic fertilizer is manure Soil and water conservation management consisting of animal dung and urine. Farmers used manure mainly near the homestead. During the focus group discussions with the key informants and DAs (Development Agents at *kebele* level), farmers (primarily those who were poor) have increased the use of manure because of the high current price of inorganic fertilizers.

Soil (stone) bund is a dam or ridge built across a slope along the contour. Soil bunds are made of soil or mud. The farmers construct the soil bunds for erosion control on moderately sloping areas. On steep eroded bare lands, stone terraces are the most used structures in the study area. As critical informants state during the interview session, the stone terraces are considered useful in erosion control in steep areas. From the survey, 20.7% of the respondents have constructed soil and stone bunds in the common eroded lands, particularly around the mountainous region; farmers were constructing bunds motivated by the cash they could earn from a safety net program.

Traditional cut-off drains become physical structures constructed by digging the soil deep to divert the runoff before reaching the farmland. The survey results show that 7.9% of the respondents used to cut off drains. The farmers construct such structures to prevent loss of seeds, fertilizer, and soil due to excessive runoff coming from uplands and dispose of the excess water for the field. However, according to farmers’ opinions, most of these structures accelerate soil erosion over time.

Planting trees and other non-crop plants such as sisal euphorbia and eucalyptus are sometimes planted along the contour with other conservation practices. This type of conservation method is applied by 12.9% of the respondents to reduce runoff and conserve the soil and water around the plants’ roots. Indigenous and newly introduced trees and shrubs are planted on overused eroded lands to make the land fully productive again. In specific areas, common highly degraded grounds are closed off to livestock to protect from grazing and planted with trees for regeneration.

Contour ploughing is tilling the land along the slope’s contours to reduce the runoff on the steeply sloping ground. It is used separately or combined with other conservation structures such as cut-off drains and plantation trees. From the sample farmers, 17.9% applied the composition in combination with cut-off waste; it is carried out using the ox-drawn plough. Hence, it is part of the standard farming activity; it needs no extra labor and time for construction. According to Mulat (2013), in several cases, most farmers in Ethiopia apply this method for soil conservation. The rest of 8.6% of respondents said they used different traditional techniques to prevent soil and water degradation. Teklu and Gezahegn (2003) argue that fallowing is abandoning the land for rejuvenation when the nutrients are exhausted. It is one of the traditional methods of leaving the cropland uncultivated for one or more years to recover soil fertility and minimize soil loss. In the study area, some of the respondents said that fallowing is used when the farmland is unproductive and loses its fertility.

Leaving the crop's residues on the field after harvest is another traditional practice used by the farmers in the area. The survey result shows that some farmers are implementing this measure to improve soil fertility and protect the land from erosion. For Teklu and Gezahegn (2003), after legumes or oil crops, the significant cereals are rotated mainly for soil fertility maintenance, disease control, and weed. Crop rotation becomes another widespread phenomenon where maize, groundnut, and bean are grown rotationally. The farmers apply crop rotation for different reasons, such as soil fertility, therefore, improved crop yield. As the scientific method developed, the farmers knew that alternating high residue-producing crops could achieve soil fertility with the low growing residue-producing crops.

Mixed cropping is widely practiced in the area. Farmers inter-plant two or more crops together with some root edible plants. The vast majority of the cases are a mix of maize and groundnuts. Mixed cropping in the area helped reduce erosion by having a crop on the land for a more extended year period. Also, it served to cultivate different crops on a single farmland period. However, the crops in the area have broadly similar growing seasons. Nevertheless, cereal crops may benefit from including leguminous plants to improve their nitrogen fixation.

Most farmers use surface mulches in their fields that provide a protective cover when crop cover does not exist. The benefit of protective covering has been widely appreciated, as was the improved infiltration rate managed by the approach and decreased evaporation rate. The further stated objective is the addition of nutrients to the soil by decomposition of the organic matter. However, the density of mulch in many fields was below the level required to be most effective as protective cover since the use of the residence as animal food was witnessed in many households of the area.

Problems related to soil water conservation measures

The respondents' significant issues related to conservation structures include a source of pests, inconveniency during ox ploughing, reduction of farmland, labor intensiveness, difficulty in implementation, and costliness. During the field survey, it was recorded that out of 140 respondents, about 39.7% indicated that soil (stone) bunds reduce farmlands, 35.8% responded inconveniency during oxen ploughing, while 24.5% revealed labor-intensive.

Regarding cut-off drains, 41.7% of respondents reported problems of costly, 34.6% said implementation difficulty, and 23.7% expressed labor intensiveness. Concerning waterways, 39.2% of the respondents indicated that it is expensive, 35.5% suggested it is difficult to implement, and 25.3% said it is labor-intensive. Respondents were also asked to compare the improved conservation practices with the traditional ones. Around 70% of the respondents indicated that conventional conservation practices better retain soil.

Conclusions

The fast population growth on the finite land resources continuously makes it difficult for developing countries to produce enough to feed their population. Attempt to generate often enough relied on the horizontal expansion of agricultural practices with substantial irreversible ecological degradations stemming from deforestation, overgrazing, and soil erosion. This scenario has culminated in poor land productivity with adverse economic, social, and political implications. Agricultural development in Ethiopia is hampered by land degradation; degradation, in turn, is threatening the overall sustainability of agricultural production. Soil erosion is a significant cause of land degradation in Ethiopia.

The Indigenous knowledge system is essential to maintain the present-day diverse and rich biodiversity among the estimated 80 ethnic groups under different agro-ecosystems in both the highland and lowland against Ethiopia's historical social and natural problems. Past research shows the role of indigenous knowledge in sustaining ecosystems while addressing socio-economic and environmental issues.

Despite the importance of the knowledge as shown by ethnoecology research, which started in the 1980s, the effort to integrate the experience with land development is not in-place. Previous research shows its importance in addressing soil infertility, labor, bio-diversity, land shortage, and increasing agricultural productivity to the extent of showing its importance in livelihood adaptation and climate change. Although modern technology is a challenge for the farmers, they have a good practice in handling soil and water by the different indigenous knowledge they employ (Table 20). Except for native terracing and soil and water conservation measures, a past study has not examined the spatial variation of practices in Ethiopia. Previous research has focused on farmers' attitudes, characterizing, and the role of the practices.

Based on the result of this study, out of the total respondents, 20% applied indigenous drainage ditches. Whereas 20.7% of the respondents have constructed soil and stone bunds in the common eroded lands, especially around the mountainous area, farmers were constructing bunds because of the cash they would earn from a safety net program. On the contrary, personal factors, socioeconomic factors and livestock holding capacity of household, institutional factors, biophysical factors, and degree of slope affect the probability of adopting both improved and traditional soil and water conservation measures.

Land availability often influences farming practices and hence affects the land degradation process. All the farmers cultivated their land with no room for expansion except in marginal areas. Sloppy grounds, which used to be grazing areas or tree plots, became under cultivation. Other alternatives that can reduce the pressure on the environment and provide a livelihood for farmers include non-farm employment.

Most farmers experienced soil erosion, a phenomenon related to the widespread onsite erosion indicators. They have sufficient knowledge of the water and soil conservation processes and the consequent on-site erosion impacts. They clearly understand various erosion indicators spread over the landscape and which adversely affect their soils. Rill and gully were most often mentioned indicators, followed by sheet wash (runoff flow paths), root exposure, and appearance of gravels in large proportion on croplands. They attributed the formation of these indicators to factors such as high rainfall, runoff from upslope fields, steep slopes, and poorly designed or ineffective soil water conservation measures, which they find themselves incapable of changing.

Farmers understand the harmful effects of erosion on crop productivity. They refer soil fertility levels and crop yield potential to a slope position. Fields on flat and gentle slopes were recognized to have the highest potential for crop production. Fields located on steep and very steep slopes were seen to be eroded hence the likelihood of not realizing high crop yields.

Likewise, farmers use a variety of indicators to understand and explain the condition of their land fertility. Farmers mentioned signs used to perceive and understand soil fertility decline identified by this study. Those signs include reduction of crop yield, stunted growth, change in crop color or crop leaves, crops unable to cope with weed dominance, high amount of straw yield compared to crop yield, and disappearance of grass and some local specific plant species. However, some of the biological and physical indicators are local and site-specific. Regarding indicators of soil fertility decline, there is an appreciable indigenous knowledge that can be tapped in developing plant species indicators.

Farmers perceived that increased crop yield could be realized, among other husbandry practices, by implementing soil water conservation measures. In addition to increased crop yield, soil water conservation measures improved soil fertility and soil-water retention. Farmers were knowledgeable about various soil water conservation rules, but few implemented them. Ridges, water diversion ditch, and contour ploughing were the most popular and traditional soil water conservation practices. Low appreciation of the agroforestry systems in the research area as a soil conservation measure suggests that farmers were more interested in the tree by-products (construction timber and wood fuel) than its scientifically perceived influence on soil and water conservation. Although farmers knew many soil water conservation measures, constraints to ensure widespread adoption were still experienced.

The most significant constraints to adopting soil water conservation were decreased farm size and inconvenience during farm operations, especially for free movement of oxen plough, followed by lack of capital and tools, labor shortage, and construction know-how.

REFERENCES

Ajibade LT. 2003. A methodology for collecting and evaluating farmers‟ indigenous environmental knowledge in developing countries. INDILINGA: Afr J Indigen Knowl Syst 2 (1): 99-113. DOI: 10.4314/indilinga.v2i1.47002.

Amede T, Belachew T, Geta E. 2001. Reversing the Degradation of Arable Land in Ethiopian Highlands. Managing African Soils No. 23. Areka Research Centre, London.

Asraf RM, Brewer JK. 2004. Conducting tests of hypotheses: The need for an adequate sample size. Austr Educ Res 31 (1): 79-94. DOI: 10.1007/BF03216806.

Berisso T. 1995. Deforestation and environmental degradation in Ethiopia: The case study of Jam Jam Province. Northeast Afr Stud 2 (2): 139-155. DOI: 10.1353/nas.1995.0010.

Bosshart U. 1999. Hydro-sedimentological characterization of Kori Sheleko and Hulet Wenz Catchments, Maybar and Andit Tid research units: Case studies of two SCRP micro-catchments in the northeastern highlands of Ethiopia. University of Berne, Switzerland.

CSA [Central Statistical Agency of Ethiopia]. 2005. Agricultural Sample Survey. Report on Area and Production. Central Statistical Agency of Ethiopia, Adis Ababa, Ethiopia.

CSA [Central Statistical Agency of Ethiopia]. 2011. Agricultural Sample Survey. Report on Area and Production. Central Statistical Agency of Ethiopia, Adis Ababa, Ethiopia.

Gizachew A. 1995. Soil erosion assessment: Approaches, the magnitude of the problem, and issues on policy and strategy development (Region 3). Workshop on Regional Natural Resources Management Potentials and Constraints, Bahir Dar, Ethiopia, 11-13 January 1995. Bureau of Natural Resources and Environmental Protection, Bahir Dar, Ethiopia.

Hanggi F. 1997. Temporal and Spatial Variation of Soil Erosion in the Research Unit Anjeni and Andit Tid: Case Studies of Test Plot Soil Erosion in Northwestern and Central highlands of Ethiopia. SCRP Research Report No. 37. University of Bern, Switzerland in Association with MOA, Ethiopia.

IIRR. 1996. Recording and using Indigenous Knowledge: A manual. International Institute for Rural Reconstruction (IIRR), Silang, Cavite, Philippines.

Lalonde A. 1991. African Indigenous Knowledge and its Relevance to Environment and Development Activities. Environment and Development Consultant R.R. 3, Wakefield, Quebec, Canada.

Mulat Y. 2013. Indigenous knowledge practices in soil conservation at Konso People, South western Ethiopia. J Agric Environ Sci 2 (2): 1-10.

Palm CA, Myers RJK, Nandwa SM. 1997. Combined use of organic and inorganic nutrient sources for soil fertility maintenance and replenishment. In: Buresh RJ, Sanchez PA, Calhoun F (eds) Replenishing soil fertility in Africa. SSSA Spec. Publ. 51. SSSA and ASA, Madison, WI.

Rockström J, Barron J, Brouwer J, Galle S, de Rouw A. 1999. On-farm spatial and temporal variability of soil and water in Pearl Millet cultivation. Soil Sci Soc Am J 63 (5): 1308-1319. DOI: 10.2136/sssaj1999.6351308x.

SCRP [Soil Conservation Research Programme]. 1982. Inception Report. University of Berne, Switzerland in association with United Nations University, Japan.

SCRP [Soil Conservation Research Programme]. 2000. A Database Report from 1981-1992; Series 1: Maybar Research Unit. University of Berne, Switzerland in Association with the Ministry of Agriculture, Ethiopia.

Shibru T. 2010. Land Degradation and Farmers' Perception: The Case of Limo *Woreda*, Hadya Zone of. SNNPR, Ethiopia. [Thesis]. Addis Ababa University, Addis Ababa.

Teklu E, Gezahegn A. 2003. Indigenous Knowledge and Practices for Soil and Water Management in East Wollega, Ethiopia. Conference on International Agricultural Research for Development October 8-10, 2003

Tesfaye M. 1988. Soil conservation experiments on cultivated land Maybar area. SCRP Research Report No. 16. University of Bern, Switzerland in Association with MOA, Ethiopia.

Tsighe Z. 1995. The political economy of land degradation in Ethiopia. Northeast Afr Stud 2 (2): 71-98. DOI: 10.1353/nas.1995.0031.

Weigel G. 1986. The soil of Gununo Area, Sidama Research Unit, Sidama Ethiopia. SCRP. Research Report 8. University of Berne, Switzerland in association with United Nations University, Japan.

BONOROWO WETLANDS
Volume 7, Number 2, December 2017
Pages: 108-116

P-ISSN: 2088-110X
E-ISSN: 2088-2475
DOI: 10.13057/bonorowo/w070207

Phytochemical composition of crude extracts derived from *Vernonia* spp. and its larvicidal activity against *Anopheles gambiae*

BEATRICE TARWISH[1,♥], JOSEPH J. N. NGERANWA[2,♥], JOSPHAT C. MATASYOH[3]
[1]School of Pure and Applied Sciences, Kenyatta University. Nairobi, Kenya.
[2]Department of Biochemistry and Biotechnology, Kenyatta University. Nairobi, Kenya. ♥email: ngeranwa.joseph@ku.ac.ke, ngeranwa@avu.org
[3]Department of Chemistry, Egerton University. Nairobi, Kenya.

Manuscript received: 1 July 2017. Revision accepted: 14 November 2017.

Abstract. *Tarwish B, Ngeranwa JJN, Matasyoh JC. 2017. Larvicidal activity* and *phytochemical composition of crude extracts derived from* Vernonia *spp. against* Anopheles gambiae. *Bonorowo Wet*lands *7: 108-116.* This study aimed to elucidate the larvicidal activity of three species of *Vernonia*, i.e., *Vernonia lasiopus*, *V. auriculifera,* and *V. galamensis,* against the malaria vector *Anopheles gambiae*. Dried samples from leaves and roots of the three plants were sequentially extracted with hexane, chloroform, ethyl acetate, acetone, methanol, and water. Following rotor evaporation of the solvents, the extracts were subjected to phytochemical analysis and larvicidal assays against the third instar larvae of *A. gambiae.* The phytochemical screening revealed the presence of steroids, saponins, flavonoids, terpenoids, and cardiac glycosides in all extracts. Tannins were present in methanol and water extracts of all three plants and acetone extract of *V. lasiopus* roots, *V. auriculifera* root, and *V. galamensis* leaf. Phlobatannins were absent in all extracts. Percent mortality for different concentrations 125, 250, 500, 750, and 1000 ppm of the extracts was calculated and subsequently subjected to probit regression analysis to determine LC_{50} and LC_{90} values. The most active extract recorded was acetone root extract of *V. galamensis* with an LC_{50} of 22.85. *Vernonia auriculifera* and *V. lasiopus* recorded the highest activity in acetone root extract and ethyl acetate root extract with an LC_{50} value of 37.7 and 205.9, respectively. A one-Way ANOVA test was performed to compare the mean mortality of all tested groups. At all exposure hours except at 3 hours, the differences were significant. The Tukey post hoc tests indicated that the mortality rate for *V. galamensis* was substantially different from the mortality rates of *V. lasiopus* and *V. auriculifera* after 24 hours. This study underlines *V. lasiopus*, *V. auriculifera,* and *V. galamensis* as alternative sources of new, cheap, and readily available larvicides to control the mosquito vector *A. gambiae.* Further study is required to determine what secondary metabolites in *Vernonia* extracts are responsible for larvicidal activity.

Keywords: *Anopheles gambiae*, crude extracts, larvicidal activity, phytochemical composition, *Vernonia*

INTRODUCTION

Malaria has been widely known as one of the major diseases in the tropical and subtropical regions of the world. The coincidence of malaria is higher in poorer countries and mainly confined in the tropical areas of Asia, Africa, and Latin America. More than 90% of malaria cases and the majority of death caused by malaria occur in tropical Africa (Bell 2006). According to The Presidential Malaria Initiative (PMI 2014), about 70% of the Kenyan population is at risk for malaria. WHO (2010) announced that the disease contributed to 20% of childhood death. While some diseases such as yellow fever have reasonably been brought under control by vaccination, unfortunately, the effective vaccine for malaria is still awaiting (Matasyoh et al., 2008). The efficacy of Mosquirix as a vaccine for malaria is being tested in phase 3 clinical trials. Early results show partial protection against malaria (Bejon 2013). Malaria is transmitted by mosquitoes carrying parasitic protozoans of the genus *Plasmodium.* The parasites are transmitted to humans by biting an infected female anopheline mosquito. In Africa, *A. gambiae* is the principal mosquito vector of malaria (Bockarie 2005).

One of the World Health Organization (WHO) strategies in combating tropical diseases is to demolish their vectors or intermediate hosts. Consequently, bioassays have become a preferred method to detect activities from traditional medicinal plant extract that could be used to prevent the transmission of four of the most common tropical diseases: malaria, schistosomiasis, dengue hemorrhagic fever, and yellow fever (Diallo 2001).

Continuous and indiscriminate synthetic insecticides have caused toxicity to non-target organisms and environmental pollution (Albuquerque 2007). Insect resistance to conventional insecticides stresses searching for new insecticides (Macedo et al. 1997). Secondary metabolites contribute towards the therapeutic value of plants, and when isolated from plants, they represent not only valuable drugs but also valuable lead molecules (Shrivastava and Patel 2007). Botanical insecticides are promising due to the nature of the compound that is potent, environmentally friendly, readily biodegradable, and inexpensive, too (Kalu et al., 2010).

The discovery of insecticidal activity of phytotoxins present in Asteraceae species has stimulated the search for new plant-derived insecticides, mainly from this plant’s family (Macedo et al., 1997). The genus *Vernonia* is one of the primary sources of bioactive sesquiterpenoids, an important class of compounds due to their diverse biological activities (Magadula and Erasto 2009). *Vernonia* comprises about 1000 species in the family Asteraceae. They have intense purple flowers in South America, North America, Asia, and Africa.

Vernonia lasiopus grows on abandoned farmland, and a decoction of the leaves is locally used to cure stomach aches (Kokwaro 2009). *Vernonia auriculifera* grows on very fertile soils. It is abundant in the wet montane forest and beside lakes or streams (Oyen 2010). Local people use a concoction of the root to treat joint pains. *Vernonia galamensis* grows in highlands on sites where rainwater collects. It is native to East Africa and is produced in many parts of Ethiopia as an industrial oilseed. This investigation aimed to determine whether the three *Vernonia* species growing in Kenya, namely *V. lasiopus, V. auriculifera*, and *V. galamensis*, possess larvicidal activity against *A. gambiae* as a possible measure to control malaria.

The objectives of the study were (i) To determine the phytochemicals in organic extracts of *V. lasiopus, V. auriculifera,* and *V. galamensis* leaves and roots; (ii) To evaluate larvicidal activity of crude extracts from *V. lasiopus*, *V. auriculifera*, and *V. galamensis* against *A. gambiae*; (iii) To determine the LC_{50} and LC_{90} values of the crude extracts from *V. lasiopus*, *V. auriculifera* and *V. galamensis* against *A. gambiae*; (iv) To determine the effective concentration of the leaf and root extracts of *V. lasiopus*, *V. auriculifera* and *V. galamensis* against *A. gambiae*.

MATERIALS AND METHODS

Collection and Identification of plant materials

Vernonia lasiopus, V. auriculifera, and *V. galamensis* were identified from the Botanical garden of Egerton University in Njoro, Kenya, by a taxonomist from the Department of Botany in the University. Voucher specimens *V. lasiopus* (BT 1), *V. auriculifera* (BT 2), and *V. galamensis* (BT 3) were deposited at the Department of Biological Sciences, Egerton University, Kenya. Fresh aerial parts of the plants and roots were collected.

Preparation of plant extracts

The roots and fresh aerial parts of plant materials of *V. lasiopus, V. auriculifera,* and *V. galamensis* were dried separately under shade to achieve a constant weight and then ground to a fine powder. About 500 g of each powder was extracted sequentially with hexane 1.5 L, chloroform 1.5 L, ethyl acetate 1.5 L, acetone 1.5 L, methanol 1.5 L, and water 1.5 L after soaking the sample in each solvent for three days. The extracts were run through a filter paper to obtain a homogenous solution. A thin layer of activated charcoal was added to the extracts from the aerial part of the plant samples to adsorb chlorophyll before filtering. The filtrates were then concentrated using a rotary evaporator to recover the solvent. The extracts were allowed to dry into a semi-solid state and subjected to phytochemical tests and larvicidal assays.

Phytochemical analysis

The extracts were analyzed for the presence of tannins, phlobatanins, saponins, flavonoids, steroids, terpenoids, and cardiac glycosides using standard procedures. A characteristic color change identified them.

Test for tannins. Two drops of 0.1% ferric chloride were added to 2ml of the extract. A brownish-green or blue, black coloration indicated a positive test (Trease and Evans 2002).

Test for phlobatannins. One milliliter of 1% aqueous HCl was added to 2 ml of the extract and boiled. Deposition of a red precipitate indicated a positive test (Trease and Evans 2002).

Test for saponins. Before filtration, the extract (1 g) was boiled with 5 ml of distilled water. Approximately 3 ml of distilled water was further added to the filtrate and shaken vigorously for 5 minutes. Frothing, which persisted in warming, was taken as evidence for the presence of saponins (Sofowora 1993).

Test for flavonoids. Extract (1 g) was dissolved in ethanol, warmed up, and filtered. Three pieces of magnesium chips were added to the filtrate, followed by two drops of concentrated HCl. A pink, orange, or red to purple coloration indicates the presence of flavonoids (Trease and Evans 2002).

Test for steroids. Two milliliters each of acetic anhydride and H_2SO_4 was added to about 2 ml of extract. A change of color indicated the positive test from violet to blue or green (Sofowora 1993).

Test for terpenoids (Salkowski test). Extract (1 g) was dissolved in ethanol. One milliliter of acetic anhydride was added, followed by concentrated H_2SO_4. A change in color from pink to violet showed the presence of terpenoids (Sofowora 1993).

Test for cardiac glycosides (Keller-Killiani test). Extract (5 ml) was tested with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. One milliliter of concentrated H_2SO_4 was added slowly (underlay). The positive test was shown by a brown ring of the interface, which indicates the deoxy-sugar characteristic of cardenolides. The positive test was also suggested by the appearance of a violet ring below the brown ring. In contrast, in the acetic acid layer, a greenish ring formed gradually throughout the thin layer (Trease and Evans 2002).

Anopheles gambiae larval culture

Larvae of *A. gambiae* from the third instar were reared in the insectaries at the Kenya Medical Research Institute (KEMRI), Centre for Disease Control (CDC), Kisumu, Kenya, using a protocol for mosquito rearing obtained from (Das et al. 2007) with slight modifications. Male and female adult mosquitoes were kept in an insectary room for 4-5 days and were fed on 10% sucrose solution. Mating occurred during this period. Four to five-day-old female mosquitoes were enclosed in a cage where a shaved rabbit was provided for the mosquitoes to acquire a blood meal by sucking the rabbit for about one hour. These females laid eggs two days after feeding on blood. A small filter paper wrapped in a conical shape was put in a bit of beaker containing water, ensuring the moistness of the filter paper. The beaker was kept inside the cage overnight, providing a place for the mosquitoes to lay eggs. The filter paper carrying the mosquito eggs was placed in a plastic tray with 300 ml of spring river water. The eggs were kept at 26 ±3 ºC to be allowed to hatch to larvae within two to three days. Growing larvae were fed on tetramine fish food until they reached the third instar larvae. During this time, water on the tray was replaced every day, and a pinch of tetramine fish food was added to the water. On the 8th day (5-day old larvae), the larvae population was diluted from one tray to 10 trays. A pinch of tetramine fish food was added to each tray. The third instar larvae emerged on day ten and were ready for bioassay experiments.

Larvicidal assays

The extracts were solubilized in Dimethyl-sulphoxide (DMSO) purchased from Lobarchemi and diluted with water to give 2000 ppm of stock solution with DMSO kept at a concentration of 1%. Serial dilution of different concentrations (125, 250, 500, 750, and 1000 ppm) was prepared from the stock solution. The bioassay tests were carried out according to a standard WHO procedure (1981) with slight modifications. The bioassays occurred at the Kenya Medical Research Institute (KEMRI), Centre for Disease Control (CDC), Kisumu, Kenya. The insect larvae were reared in plastic and enamel trays spring river water. The larvae were maintained, and all experiments were carried out at 26 ±3ºC, with the humidity range between 70 to 75%. The bioassays were done with third instar larvae of *A. gambiae* and examined in triplicate using 20 larvae for each replicate assay. The larvae were laid up in 50 ml disposable plastic cups containing 15 ml of the test solution and fed on tetramine fish feed during all testing periods. Larvae were considered dead if they were unresponsive, even when gently prodded. The dead larvae were counted at 3-hour intervals for 24 hours. The dead larvae in the three replicates were combined and expressed as the percentage mortality for each concentration. The negative control was 1% DMSO in spring river water, while the positive control was 100 ppm pyrethrum-based larvicide, pylarvex.

Data analysis

There was a significant difference between the means of *the groups (V. lasiopus, V. auriculifera,* and *V. galamensis*) as determined by a one-way ANOVA. Nevertheless, a one-way ANOVA is an omnibus test statistic that can only tell that somewhere between the groups, a difference exists and cannot tell which specific groups were significantly different from each other. For this reason, the post hoc tests were run to confirm where the differences occurred between groups. Post hoc tests are executed when an overall significant difference in group means has already been established. Furthermore, Tukey's honestly significant difference (HSD) test is used as a post hoc test because it is less conservative than the Scheffe test (that means that you have a higher probability of detecting differences if they exist with Tukey's HSD test). The average mortality data of the larvae were also subjected to probit regression analysis based on the previous method (Finney 1971) to estimate LC_{50} and LC_{90} values and associated 95% confidence limits. Probit analysis is a regression analysis used to analyze binomial response variables. It transforms the sigmoid dose-response curve to a straight line which can then be analyzed by regression through least squares or maximum likelihood. This analysis is commonly used in toxicology to determine the relative toxicology of chemicals to living organisms. The method is done by testing the response of an organism under various concentrations of each of the substances in question and then comparing the concentrations at which one encounters a reaction. The response is always binomial, and the relationship between the response and the various levels is still sigmoid. Probit analysis acts as a transformation from sigmoid to linear and then runs a regression on the link. The researcher can utilize the output of the probit analysis to compare the amount of the chemicals needed to create the same response in each of the various chemicals. Many endpoints are used to analyze the differing toxicities of compounds, but the LC_{50} (Liquid) and the LD_{50} (Solid) are the most widely used outcomes of the current dose-response experiments. The LC_{50}/LD_{50} defines the concentration (LC_{50}) and the dose (LD_{50}) at which 50% of the population responds. Statistical Package for the Social Sciences (SPSS) software version 19 (IBM) was used for data analysis.

RESULTS AND DISCUSSION

Phytochemical compounds in *Vernonia* extracts

Both leaves and roots from three *Vernonia* plant extracts contained almost similar compounds. The results obtained from phytochemical tests are given in (Table 1). The data for the phytochemical screening of *V. lasiopus, V. auriculifera,* and *V. galamensis* extracts (Table 1) revealed the presence of steroids, flavonoids, saponins, terpenoids, and cardiac glycosides. Tannins were present in the methanol and water extracts of the three plants and acetone extracts of *V. lasiopus* root, *V. auriculifera* root, and *V. galamensis* leaf. However, phlobatannins were absent in all extracts.

Results of larvicidal activity

The larvicidal activity of hexane, chloroform, ethyl acetate, acetone, methanol, and water extracts of the leaves and roots of *V. auriculifera, V. lasiopus,* and *V. galamensis* revealed that all the tested extracts were active against the larvae of *A. gambia*e except the water extracts. Data of the larvicidal activity of the extracts against *A. gambiae* are presented in (Table 2). In the negative control, no mortality was detected of 1% DMSO in spring river water.

A one-Way ANOVA test was done to compare the means of the groups (*V. lasiopus, V. auriculifera, V. galamensis*). The results are recorded in (Table 3). The differences were significantly different at all hours except at 3 hours as determined by one-way ANOVA (Table 3). For each significant test, Post-hoc Tukey's HSD tests were done to determine which two pairs differed significantly, and the results were shown in the tables below.

Table 1. Phytochemical compounds present in *Vernonia* plant extracts from leaf and root using different solvents

Fraction	**Species**	**Leaf**						**Root**					
		Hexane	**EtOAc**	**$CHCl_3$**	**Acetone**	**MeOH**	**Water**	**Hexane**	**EtOA**	**$CHCl_3$**	**Acetone**	**MeOH**	**Water**
Tannins	*V. lasiopus*	-	-	-	-	+	+	-	-	-	+	+	+
	V. auriculifera	-	-	-	-	+	+	-	-	-	+	+	+
	V. galamensis	-	-	-	+	+	+	-	-	-	-	+	+
Phlobatanins	*V. lasiopus*	-	-	-	-	-	-	-	-	-	-	-	-
	V. auriculifera	-	-	-	-	-	-	-	-	-	-	-	-
	V. galamensis	-	-	-	-	-	-	-	-	-	-	-	-
Saponin	*V. lasiopus*	+	+	+	+	+	+	+	+	+	+	+	+
	V. auriculifera	+	+	+	+	+	+	+	+	+	+	+	+
	V. galamensis	+	+	+	+	+	+	+	+	+	+	+	+
Flavonoids	*V. lasiopus*	+	+	+	+	+	+	+	+	+	+	+	+
	V. auriculifera	+	+	+	+	+	+	+	+	+	+	+	+
	V. galamensis	+	+	+	+	+	+	+	+	+	+	+	+
Steroids	*V. lasiopus*	+	+	+	+	+	+	+	+	+	+	+	+
	V. auriculifera	+	+	+	+	+	+	+	+	+	+	+	+
	V. galamensis	+	+	+	+	+	+	+	+	+	+	+	+
Terpenoids	*V. lasiopus*	+	+	+	+	+	+	+	+	+	+	+	+
	V. auriculifera	+	+	+	+	+	+	+	+	+	+	+	+
	V. galamensis	+	+	+	+	+	+	+	+	+	+	+	+
Cardiac glycosides	*V. lasiopus*	+	+	+	+	+	+	+	+	+	+	+	+
	V. auriculifera	+	+	+	+	+	+	+	+	+	+	+	+
	V. galamensis	+	+	+	+	+	+	+	+	+	+	+	+

Note: + present, - absent

The Tukey post hoc tests indicated that the mortality rate for *V. galamensis* (mean = 1.47) was significantly different from the rate of the mortality of *V. lasiopus* (mean = 0.66) and *V. auriculifera* (mean = 0.79). However, there is no significant difference between *V. lasiopus* (mean = 0.66) and *V. auriculifera* (mean = 0.79). This pattern was maintained at all hours, with the final recording made at 24 hours, as shown in (Table 4).

On a similar note, the Tukey post hoc tests showed that *V. galamensis* mortality rate (mean = 4.08) was significantly different from that of *V. lasiopus* (mean = 2.16) and *V. auriculifera* (mean = 2.41).

Additionally, based on Tukey post hoc tests, the mortality rate for *V. galamensis* (mean = 6.62) was significantly different from the mortality rates of *V. lasiopus* (mean = 4.02) and *V. auriculifera* (mean = 4.23).

The Tukey post hoc tests suggested that the mortality rate for *V. galamensis* (mean = 7.73) was significantly different from that of *V. lasiopus* (mean = 5.29) and *V. auriculifera* (mean = 5.57).

The Tukey post hoc tests gave the following results, as shown in (Table 5).

The Tukey post hoc tests demonstrated that the mortality rate for *V. lasiopus* (mean = 0.50) and *V. auriculifera* (mean = 0.19) were significantly different from the mortality rate for *V.galamensis*(mean = 1.23). This pattern was replicated for hours 9, 12,15, and 18, as shown in (Table 5) after which the trend changed.

The Tukey post hoc tests showed that the mortality rate for *V. galamensis* (mean = 4.84) was significantly different from the counterparts of *V. auriculifera* (mean = 1.91). Yet, the mortality rate for *V. lasiopus* (mean = 3.40) is neither significantly different from that of *V. galamensis* nor *V. auriculifera.* The same pattern was maintained at hour 24, as shown in (Table 5).

When all the root extracts were analyzed, The Tukey post hoc tests indicated significant differences between the means of groups (*V. lasiopus, V. auriculifera,* and *V. galamensis*) as recorded in (Table 6).

The Tukey post hoc result showed that the mortality rate between *V. lasiopus* (mean = 0.82) and *V. galamensis* (mean = 1.71) was significantly different. However, the mortality rate for *V. auriculifera* (mean = 1.40), on the other hand, was not substantially different from neither *V. lasiopus* nor *V. galamensis*. This pattern was replicated for hours 9, 12, 15, and 18, as shown in (Table 6).

When extracts of various solvents were analyzed, acetone extracts of root and leaf showed significant differences between the means of V. lasiopus, V. auriculifera, and V. galamensis (Table 7).

When acetone was used for roots extraction, the Tukey post hoc tests indicated that the mortality rate for the three plants was significantly different from each other. The differences were found to be significantly different between any two plants, *V. galamensis* (mean = 19.47), *V. auriculifera* (mean = 16.87), and *V. lasiopus* (mean = 5.29).

At hour 24, the Tukey post hoc tests indicated the mortality rate of acetone extract of the leaves for *V. lasiopus* (mean = 6.60) and *V. auriculifera* (mean = 6.07) was not statistically different. Yet, the two means were statistically different from the mean of *V. galamensis* (mean = 17.53).

The larvicidal activity of *V. lasiopus*, *V. auriculifera*, and *V. galamensis* (Table 8) was determined by probit analysis as estimates of lethal concentration that kills 50% (LC_{50}) and 90% (LC_{90}) of the exposed larvae respectively and their associated 95% confidence interval.

Table 2. Mean mortality and the standard deviation of *A. gambiae larvae* exposed to root and leaf extracts of *V. Auriculifera, V. galamensis,* and *V. lasiopus*

Plant	Solvent		3hrs	6hrs	9hrs	12hrs	15hrs	18hrs	21hrs	24hrs
V. auriculifera	Acetone	Mean	.20	1.93	3.67	5.67	7.37	9.13	10.93	11.47
		N	30	30	30	30	30	30	30	30
		SD	.484	2.067	3.231	4.205	5.623	5.698	6.416	6.329
	Chloroform	Mean	.03	1.57	2.73	4.33	6.47	6.67	6.77	7.73
		N	30	30	30	30	30	30	30	30
		SD	.183	2.738	4.362	6.456	8.476	8.535	8.553	8.558
	Ethyl acetate	Mean	.70	1.23	2.17	3.73	4.97	7.20	8.47	8.80
		N	30	30	30	30	30	30	30	30
		SD	3.650	1.870	3.217	5.119	6.708	8.540	9.012	9.118
	Hexane	Mean	.00	.00	.00	.00	.00	.00	.00	.00
		N	30	30	30	30	30	30	30	30
		SD	.000	.000	.000	.000	.000	.000	.000	.000
	Methanol	Mean	.00	.03	.17	.73	1.70	2.37	3.23	3.77
		N	30	30	30	30	30	30	30	30
		SD	.000	.183	.531	.907	1.643	2.141	3.491	3.520
	Water	Mean	.00	.00	.00	.00	.00	.00	.00	.00
		N	30	30	30	30	30	30	30	30
		SD	.000	.000	.000	.000	.000	.000	.000	.000
	Total	Mean	.16	.79	1.46	2.41	3.42	4.23	4.90	5.29
		N	180	180	180	180	180	180	180	180
		SD	1.505	1.771	2.943	4.371	5.786	6.535	7.140	7.284
V. galamensis	Acetone	Mean	.43	5.87	9.27	13.53	17.20	17.97	18.27	18.50
		N	30	30	30	30	30	30	30	30
		SD	.626	3.421	2.690	3.256	3.755	3.189	2.947	2.862
	Chloroform	Mean	.10	1.23	2.67	4.23	6.50	6.77	6.83	7.60
		N	30	30	30	30	30	30	30	30
		SD	.403	1.888	3.642	5.734	8.936	8.943	8.906	8.609
	Ethyl acetate	Mean	.00	1.60	3.03	4.77	5.87	10.20	11.20	12.27
		N	30	30	30	30	30	30	30	30
		SD	.000	1.993	3.368	4.939	6.196	8.252	8.062	7.939
	Hexane	Mean	.13	.13	.47	1.40	2.23	2.93	3.77	4.27
		N	30	30	30	30	30	30	30	30
		SD	.507	.507	1.008	2.094	3.287	4.201	4.925	5.298
	Methanol	Mean	.00	.00	.00	.53	1.30	1.87	2.80	3.77
		N	30	30	30	30	30	30	30	30
		SD	.000	.000	.000	1.634	2.867	3.655	4.874	5.418
	Water	Mean	.00	.00	.00	.00	.00	.00	.00	.00
		N	30	30	30	30	30	30	30	30
		SD	.000	.000	.000	.000	.000	.000	.000	.000
	Total	Mean	.11	1.47	2.57	4.08	5.52	6.62	7.14	7.73
		N	180	180	180	180	180	180	180	180
		SD	.394	2.727	3.979	5.771	7.581	8.234	8.349	8.390
V. lasiopus	Acetone	Mean	.17	.50	1.17	1.97	2.60	3.47	4.40	4.93
		N	30	30	30	30	30	30	30	30
		SD	.461	.900	1.642	2.109	2.513	2.763	3.654	4.076
	Chloroform	Mean	.07	2.33	3.87	5.83	7.00	7.37	7.70	8.30
		N	30	30	30	30	30	30	30	30
		SD	.254	4.020	5.097	6.894	7.991	8.122	8.322	8.730
	Ethyl acetate	Mean	.00	.93	1.97	3.07	4.53	9.27	10.73	11.30
		N	30	30	30	30	30	30	30	30
		SD	.000	1.413	2.341	3.704	5.643	8.337	8.725	8.864
	Hexane	Mean	.00	.00	.27	.87	1.10	1.53	1.90	2.67
		N	30	30	30	30	30	30	30	30
		SD	.000	.000	.980	2.047	2.383	2.825	3.166	3.968
	Methanol	Mean	.00	.20	.40	1.23	1.63	2.47	5.33	6.20
		N	30	30	30	30	30	30	30	30
		SD	.000	.664	1.102	2.112	1.921	3.003	6.375	6.435
	Water	Mean	.00	.00	.00	.00	.00	.00	.00	.00
		N	30	30	30	30	30	30	30	30
		SD	.000	.000	.000	.000	.000	.000	.000	.000
	Total	Mean	.04	.66	1.28	2.16	2.81	4.02	5.01	5.57
		N	180	180	180	180	180	180	180	180
		SD	.221	1.952	2.769	3.957	4.852	6.052	6.822	7.108

Table 3. One-Way ANOVA test results showing the level of significance at a 3-hour interval

Hour	3	6	9	12	15	18	21	24
$F(2,537)$	0.758	7.102	8.267	8.614	9.510	7.687	5.167	5.544
p-value	0.469	0.001	0.001	0.000	0.000	0.001	0.006	0.004

Table 4. Total mean mortality rates of all extracts after

Plant name	N	Subset for alpha = 0.05	
		1	2
6 hours			
V. auriculifera	90	0.66	
V. lasiopus	90	0.79	
V. galamensis	90		1.47
12 hours			
V. auriculifera	90	2.16	
V. lasiopus	90	2.41	
V. galamensis	90		4.08
18 hours			
V. auriculifera	90	4.02	
V. lasiopus	90	4.23	
V. galamensis	90		6.62
24 hours			
V. auriculifera	90	5.29	
V. lasiopus	90	5.57	
V. galamensis	90		7.73

Table 5. Total mean mortality rates of all leaf extracts of *Vernonia* spp.

Plant name	N	Subset for alpha = 0.05	
		1	2
6 hours			
V. auriculifera	90	0.19	
V. lasiopus	90	0.50	
V. galamensis	90		1.23
9 hours			
V. auriculifera	90	0.36	
V. lasiopus	90	0.87	
V. galamensis	90		1.81
12 hours			
V. auriculifera	90	0.64	
V. lasiopus	90	1.32	
V. galamensis	90		2.80
15 hours			
V. auriculifera	90	0.99	
V. lasiopus	90	1.59	
V. galamensis	90		3.64
18 hours			
V. auriculifera	90	1.69	
V. lasiopus	90	2.54	
V. galamensis	90		4.28
21 hours			
V. auriculifera	90	1.91	
V. lasiopus	90	3.40	
V. galamensis	90		4.84
24 hours			
V. auriculifera	90	2.42	
V. lasiopus	90	3.90	
V. galamensis	90		5.58

Table 6. Total mean mortality rates of all root extracts

Plant name	N	Subset for alpha = 0.05	
		1	2
6 hours			
V. auriculifera	90	0.82	
V. lasiopus	90	1.40	
V. galamensis	90		1.71
9 hours			
V. auriculifera	90	1.69	
V. lasiopus	90	2.56	
V. galamensis	90		3.33
12 hours			
V. auriculifera	90	3.00	
V. lasiopus	90	4.18	
V. galamensis	90		5.36
15 hours			
V. lasiopus	90	4.03	
V. auriculifera	90	5.84	
V. galamensis	90		7.39
18 hours			
V. lasiopus	90	5.49	
V. auriculifera	90	6.77	
V. galamensis	90		8.97

Table 7. Total mean mortality rates of all root and leaf extracts of acetone after 24 hours

Plant name	N	Subset for alpha = 0.05		
		1	2	3
Root extracts				
V. lasiopus	15	3.27		
V. auriculifera	15		16.87	
V. galamensis	15			19.47
Leaf extracts				
V. auriculifera	15	6.07		
V. lasiopus	15	6.60		
V. galamensis	15		17.53	

Table 9. Effective concentration of *V. auriculifera, V. galamensis*, and *V. lasiopus* extracts against *A. gambiae*

Plant	Part	Effective concentration
V. auriculifera	Leaf	None
V. auriculifera	Root	1000 ppm
V. galamensis	Leaf	None
V. galamensis	Root	500 ppm and above
V. lasiopus	Leaf	1000 ppm
V. lasiopus	Root	1000ppm

Table 8. Estimates of LC_{50} and LC_{90} values of *V. Lasiopus, V. auriculifera,* and *V. galamensis* extracts from the root and leaf with different solvents against *A. gambiae* and their associated 95% confidence interval

Solvent	Part	LC_{50} (ppm)and 95% confidence interval	LC_{90} (ppm)and95% confidence interval
V. lasiopus			
Hexane	Leaf	1730.4 (753.7-2707.2)	2306.9 (520.1-4093.6)
	Root	966.6 (790.0-1143.2)	1469.6 (1039.3-1899.9)
Chloroform	Leaf	872.9 (820.2-925.6)	997.9 (918.2-1077.6)
	Root	309.9 (273.8-346.0)	405.4 (355.3-455.0)
Ethyl acetate	Leaf	622.9 (574.6-671.3)	748.3 (685.1-811.4)
	Root	205.9 (179.2-232.6)	276.63 (239.4-313.8)
Acetone	Leaf	1108 (554.9-1661.0)	3744.7 (1813.2-7308.1)
	Root	1285.5 (907.6-1663.5)	2012.2 (1087.6-2936.8)
Methanol	Leaf	2247.1(376.9-4117.3)	7176.4 (-2807.6-17161)
	Root	577.6 (517.9-637.3)	767.94 (677.17-858.71)
Water	Leaf	0.0	0.0
	Root	0.0	0.0
V. auriculifera			
Hexane	Leaf	0.0	0.0
	Root	0.0	0.0
Chloroform	Leaf	1095.3 (905.5-1285.1)	1466.6 (1066.1-1867.0)
	Root	314.4 (278.7-350.1)	406.0 (357.4-454.6)
Ethyl acetate	Leaf	2246.1(355.1-4116.3)	7174.0 (-2807-17160)
	Root	198.0 (175.4-220.6)	250.0 (219.6-280.3)
Acetone	Leaf	1730.3 (752.7-2706.2)	2306.9 (752.7-4093.6)
	Root	37.7 (15.0-60.4)	148.4 (81.1-215.8)
Methanol	Leaf	1494.0 (873.1-2114.9)	2713.4 (944.3-4482.5)
	Root	1824.7 (634.0-3015.5)	4836.2 (4129.3-10085)
Water	Leaf	0.0	0.0
	Root	0.0	0.0
V. galamensis			
Hexane	Leaf	959.5 (848.5-1070.4)	1227.1(1005.0-1449.1)
	Root	814.0 (746.2-881.8)	1005.6 (889.7-1121.6)
Chloroform	Leaf	1339.39 (943.3-1844.5)	2040.5 (1043.0-3038.0)
	Root	293.4 (267.4-319.5)	338.1(300.5-375.7)
Ethyl Acetate	Leaf	735.0 (661.0-809.1)	972.5 (839.4-1105.7)
	Root	68.2 (33.06-103.4)	114.1(375.0-153.1)
Acetone	Leaf	62.6 (28.7-96.5)	139.2 (88.7-189.7)
	Root	22.8 (5.82-39.8)	45.3 (17.9-72.7)
Methanol	Leaf	105587(-365501-576676)	2623534 (-15360000-20609746)
	Root	15368(-22676-53413)	129930(-370638-630498)
Water	Leaf	0.0	0.0
	Root	0.0	0.0

The most active extracts recorded were *V. galamensis* acetone root extract, acetone leaf extract, and ethyl acetate root extract with LC_{50}values of 22.85, 62.61, and 68.24, respectively. Ethyl acetate root extracts were the most active extract of *V. lasiopus,* with an LC_{50}value of 205.9. *Vernonia lasiopus* recorded an LC_{50} value of 37.7 in acetone root extract as its most active extract.

Since effective concentration was assumed to be any concentration that kills at least 50% of the exposed larva of *A. gambiae*, Table 9 shows the sufficient concentration for *V. auriculifera* root extracts and *V. lasiopus* root and leaf extracts were 1000 ppm. In contrast, the effective concentration for *V. galamensis* root extracts was 500 ppm and above. Extracts of *V. galamensis* and *V. auriculifera* leaf extracts did not have an effective concentration of 125 ppm to 1000 ppm.

Discussion

Phytochemical screening of *V. lasiopus, V. auriculifera,* and *V. galamensis* extracts revealed the presence of steroids, saponins, flavonoids, terpenoids, and cardiac glycosides. Tannins could be extracted with methanol from the three plants and acetone from *V. lasiopus* root, *V. auriculifera* root, and *V. galamensis* leaf. However, phlobatannins were absent in all extracts (Table 1). This study suggested extracts from the three *Vernonia* plants possess larvicidal activity against *Anopheles gambiae*. Their activity was significantly different at all hours except at 3 hours exposure as determined by one-way ANOVA. The activity of crude plant extracts is attributed to the complex mixture of active compounds. We suggest that the bioactivity of the extracts observed here could be due to the phytochemicals detected, which have been reported in an earlier study to be responsible for mosquito larvicidal activity. Most studies have reported active insecticidal compounds like saponins, tannins, terpenoids, flavonoids, and steroids. Several plants of the Asteraceae family are said to have mosquito larvicidal activity due to the presence of several flavonoids (Srivastava et al., 2008). Steroid compounds are responsible for mosquito larval toxicity (Chowdhury et al., 2008). Saponin is well known as a potential mosquito larvicidal compound. It works by interacting with the cuticle membrane of the larva, ultimately disconnecting the membrane, which is the most plausible reason for larval death (Bayavan 2008; Chowdhury et al. 2008).

The difference in the activity of the extracts could be attributed to the different concentrations of phytochemicals in the extracts. The bioactivities demonstrated by the various extracts may also be attributed to the uneven distribution of chemical constituents within these extracts. When used in their crude form, the phytochemicals could have exhibited synergistic or additive effects (Mohamed et al., 2010). Secondary compounds of plants may jointly or independently have activity against mosquito targets from their ovicidal, pupicidal, activity against the adult, and inhibition of growth activity (Borah et al., 2010).

The primary target of insecticides includes the nervous system, endocrine, and metabolic processes. Metabolic processes are inhibited by disrupting the cell membrane and mechanical suffocation. Inhibition of metabolic pathways such as electron transport and oxidative phosphorylation disrupts energy production leading to death. Other insecticides, e.g., organophosphates, cause overstimulation of the nervous system leading to paralysis and death. The chemicals might achieve this effect by inhibiting cholinesterase activity or acting as sodium channel modulators (Brown 2006). Insect growth regulators (IGRs) attack the insect's endocrine system, which produces the hormones needed for growth and development into the adult form. Insects poisoned with IGRs cannot molt or reproduce, and they eventually die since hormones play various roles in molting (Elsheikha and Khan 2011). Disturbance of or interference with any of these hormones suppressed the molting process. Different insecticides target insect growth and development by interfering with hormones and others by preventing the production of a structural component of the exoskeleton. Some insecticides called Chitin Synthesis Inhibitors (CSI) inhibit the production of chitin. An insect poisoned with a CSI cannot produce chitin and thus cannot molt. Since molting must occur for the insect to reach the adult stage, a CSI poisoned insect cannot reproduce (Brown 2006). The mortality obtained in all the extracts may be due to any of these effects brought by the presence of active chemical compounds.

The larvicidal activity was observed at 3-hour intervals for 24 hours. No mortality was seen in the controls. Extracts from hexane, chloroform, ethyl acetate, acetone, and methanol showed a moderate toxic effect on *A. gambiae* after 24 hours of exposure. Ethyl acetate, chloroform, and acetone exhibited more significant activity than the least polar hexane extract and the more polar extract methanol. Water extracts as the most polar solvent showed no activity. Higher activity in ethyl acetate, chloroform, and acetone compared to hexane and methanol may be due to the differences in the type of active constituents present in the extracts (Jeyaseelan et al., 2012).

Lack of activity in water extracted samples could have resulted from the hydrolytic effect of water—hydrolysis of particularly saponins. A known larvicidal compound yields saprogenic and corresponding glycosides with loss of activity. Saponin also hydrolyzes with time, and so the longer the duration of extraction of saponin, the higher the hydrolysis. On the other hand, tannins, flavonoids, terpenes, and some saponins are most difficult to be dissolved in water but more easily extractable with methanol (Sakagami et al., 2012). According to Olugbenga et al. (2012), methanol is a better solvent for saponin than water. Several tannins, including hydrolyzable geranin, are also unstable in water. Organic plant extracts were found to give more consistent antimicrobial activity than water extracts. Also, water-soluble flavonoids, mostly anthocyanins, have no significant antimicrobial activity, and water-soluble phenolics are only crucial as antioxidant compounds (Das et al., 2010).

The Tukey post hoc tests indicated that after 24 h, the mortality rate for *V. galamensis* (mean = 7.73) was significantly different from that of *V. lasiopus* (mean = 5.29) and *V. auriculifera* (mean = 5.57). It is evident that *V. galamensis* showed the highest total mean mortality making it the most promising plant. The results were also confirmed by the results of the probit analysis that indicated that *V. galamensis* recorded low LC_{50} values in some extracts, which included acetone root extract (22.85), acetone leaf extract (62.61), and ethyl acetate root extract (68.24). *Vernonia auriculifera* and *V. lasiopus* recorded the highest activities in acetone root extract, and ethyl acetate root extracts with an LC_{50} value of 37.7 and 205.9, respectively.

Results in (Table 9) confirm that root extracts were more potent than the leaf extracts. The effective concentration for *V. auriculifera* root extracts and *V. lasiopus* root extracts were 1000 ppm. On the other hand, the active concentration for *V. galamensis* root extracts was 500 ppm and above. *Vernonia lasiopus* leaf extracts recorded a sufficient concentration of 1000 ppm. However, *V. galamensis* and *V. auriculifera* leaf extracts did not have effective levels ranging between 125 ppm and 1000 ppm.

The larval stage is regarded as the most vulnerable stage to attack mosquitoes as they are concentrated in small areas. One of the approaches for controlling malaria transmission is interrupting the mosquito life cycle at the larval stage (Srivastava et al., 2008). The finding obtained in this study confirms the potential of extracts from V. galamensis, V. lasiopus, and V. auriculifera to control the larval population of *A. gambiae.*

Conclusions

The leaves and roots of *V. lasiopus*, *V. auriculifera*, and *V. galamensis* contain active insecticidal compounds, including steroids, flavonoids, cardiac glycosides, terpenoids, tannins, and saponins. All of the three species possess larvicidal activity against *A. gambiae*, with *V. galamensis* exhibiting the highest total mean mortality (mean = 7.73), making it the most potent plant compared to *V. lasiopus* (mean = 5.29) and *V. auriculifera* (mean = 5.57) after 24 hours. The result was also confirmed by the data of the probit analysis, which indicated that *V. galamensis* recorded low LC50 values in some extracts that included acetone root extract, acetone leaf extract, and ethyl acetate root extract. *Vernonia auriculifera* and *V. lasiopus* recorded the highest activities in acetone root extract, and ethyl acetate root extracts with an LC50 value of 37.7 and 205.9, respectively. The finding from this study indicates that the root extracts were more potent than the leaf extracts. The effective concentration for *V. auriculifera* root extracts and *V. lasiopus* root extracts were 1000 ppm. Meanwhile, the effective concentration for *V. galamensis* root extracts was from 500 ppm and above. *Vernonia* lasiopus leaf extracts recorded an adequate concentration of 1000 ppm. However, from the concentration tested in this study that falls between 125 ppm and 1000 ppm, neither *V. galamensis* nor *V. auriculifera* leaf extracts showed effectiveness against *A. gambiae*. *Vernonia lasiopus*, *V. auriculifera*, and *V. galamensis* can be used to develop newer, more selective, and biodegradable larvicide as an alternative to the rather expensive and environmentally hazardous inorganic insecticides. Further work to determine what secondary metabolites in *Vernonia* extracts are responsible for the larvicidal activity is of future interest.

REFERENCES

Albuquerque MRJR, Costa SMO, Bandeira PN, Santiago GMP, Andrede-Neto M, Silveiria ER, Pessoa ODL. 2007. Nematicidal, larvicidal activities of *Pectis oligocephala* and *Pectis apodocephala* Baker. Anais da Academia Brasileira de Ciencias 79 (2): 209-213. DOI: 10.1590/S0001-37652007000200003.

Bayavan A, Rahuman AA, Kamaraj C, Kannappan G. 2008. Larvicidal activity of a saponin from Achyranthes aspera against *Aedes aegypti* and *Culex quinquefasciatus*. Parasitol Res 103: 1223-1229. DOI: 10.1007/s00436-008-0962-z.

Bell D, Wongsrichanalai C, Barnwell JW. 2006. Ensuring quality and access for malaria diagnosis: how can it be achieved? Nat Rev Microbiol 4 (9): S7-S20. DOI: 10.1038/nrmicro1525.

Benjon P, White MT, Olotu A, Bojang K, Lusingu JPA, Salim N, Otsyula N, Agnandji ST, Asante SO, Abdulla S, Ghani AC. 2013. Efficacy of RTS, S malaria vaccines: Individual-participant pooled analysis of phase 2 data. Lancet Infect Dis 13: 319-327. DOI: 10.1016/S1473-3099(13)70005-7.

Bockarie JM. 2005. Biology of Disease Vectors. 2nd ed. Am J Trop Med Hygiene 73: 1167. DOI: 10.4269/ajtmh.2005.73.1167.

Borah R, Kalita MC, Kar A, Talukdar AK. 2010. Larvicidal efficacy of *Toddalia asiatica* (Linn.) Lam against two mosquito vectors, *Aedes aegypti* and *Culex quinquefasciatus*. Afr J Biotechnol 9 (17): 2527-2530.

Brown AE. 2006. Mode of action of insecticides and related pest control chemicals for production agriculture, ornamentals and turf. Pestic Educ Assess Programs 301: 405-3913.

Chowdhury N, Ghosh A, Chandra G. 2008. Mosquito larvicidal activities of *Solanum villosum* berry extract against the dengue vector *Stegomyia aegypti*. BMC Compl Altern Med 8 (1): 1-8. DOI: 10.1186/1472-6882-8-10. DOI: 10.1186/1472-6882-8-10.

Das K, Tiwari RKS, Shrivastava DK. 2010. Techniques for evaluation of medicinal plant products as antimicrobial agent: Current methods and future trends. J Med Plants Res 4 (2): 104-111.

Das S, Garver L, Dimopoulos G. 2007. Protocol for mosquitoe rearing (*A. gambiae*). J Visualised Exp 5: 221. DOI: 10.3791/221.

Diallo D, Marston A, Terreaux C, Touré Y, Paulsen BS, Hostettmann K. 2001. Screening of Malian medicinal plants for antifungal, larvicidal, molluscicidal, antioxidant, and radical scavenging activities. Phytotherapy Res 15 (5): 401-406. DOI: 10.1002/ptr.738.

Elsheikha HM, Khan NA. 2011. Essentials of Veterinary Parasitology. Caister Academic Press, Poole, UK.

Finney DJ. 1971. Probit Analysis. 3rd ed. Cambridge University Press, London, UK.

Jeyaseelan EC, Vinuja T, Pathmanathan K, Jeyadevan J.P. 2012. Control of plant pathogenic fungi using organic solvent extracts of leaf, flower, and fruit of *Lawsonia inermis* L. Intl J Pharmaceut Biol Arch 3 (4): 783-788.

Kalu IG, Ofoegbu U, Eroegbusi J, Nwachukwu CU, Ibeh B. 2010. Larvicidal activities of ethanol extracts of *Allium sativum* (garlic bulb) against the filarial vector, *Culex quinquefasciantus*. J Med Plants Res 4 (6): 496-498.

Kokwaro JO. 2009. Medicinal Plants of East Africa. 3rd ed. University of Nairobi Press, Nairobi.

Macedo ME, Consoli RAGB, Grandi SM, Dos Anjos M .G, Oliera BA, Mendes AN, Queiroz OR, Zani LC. 1997. Screening of Asteraceae (Compositae) plant extracts for larvicidal activity against *Aedes fluviatilis* (Dipthera: Culicidae). Memórias do Instituto Oswald Cruz. Rio de Janeiro 92: 565-570. DOI: 10.1590/S0074-02761997000400024.

Magadula JJ, Erasto P. 2009. Bioactive natural products derived from the East African flora. Nat Prod Rep 26 (12): 1535-1554. DOI: 10.1039/b906089h.

Matasyoh JC, Wathuka EM, Kariuki ST, Chepkorir R, Kavulani J. 2008. Aloe plant extracts as alternative larvicides for mosquito control. Afr J Biotechnol 7 (7): 912-915.

Mohamed LT, El Nur BS, Abdelrahman MN. 2010. The antibacterial, antiviral activities, and phytochemical screening of some Sudanese medicinal plants. EurAsia J Biosci 4: 8-16. DOI: 10.5053/ejobios.2010.4.0.2.

Olugbenga AM, Musa AH, Olwatoyin AH. 2012. Toxicological activity of crude saponin extract of *Ficus platyphylla*. Asian J Pharmaceut Clin Res 5 (1): 30-33.

Oyen LPA. 2010. *Vernonia auriculifera* Hiern. In: Brink M, Achigan-Dako EG (eds) Prota 16: Fibres. PROTA, Wageningen, Netherlands.

PMI. 2014. Kenya Malarial Operational Plan FY 2014. President’s Malaria Initiative, Government of Kenya and USA, Washington, DC.

Sakagami H, Kushida T, Makino T, Hatano T, Shirataki Y, Matsuta T, Matsuo Y, Mimaki Y. 2012. Functional Analysis of Natural Polyphenols and Saponins as alternative Medicine. In: Bhattacharya A (ed) A Compendium of Essays on Alternative Therapy. InTech, Rijeka, Croatia. DOI: 10.5772/30449.

Sofowora A. 1993. Medicinal Plants and Traditional Medicine in Africa. Spectrum Books Ltd., Ibadan, Nigeria.

Srivastava A, Bartarya R, Tonk S, Srivastava SS, Kumari M. 2008. Larvicidal activity of an Indigenous plant, *Centratherum anthelmintcum*. J Environ Biol 29 (5): 669-672.

Srivastava N, Patel T. 2007. *Clerodendrum* and healthcare: An overview-part II phytochemistry and biotechnology. Med Aromat Plant Sci Biotechnol 1 (2): 209-223.

Trease GE, Evans WC. 2002. Pharmacolognsy. 15th ed. Saunders Publishers, London, UK.

WHO. 1981. Instructions for determining susceptibility of resistance of mosquito larvae to insecticides. WHO/VBC-81. World Health Organization, Geneva.

WHO. 2010. Malaria Fact sheet No. 94, April 2010. World Health Organization, Geneva.