Preliminary taxonomic survey and molecular documentation of jellyfish species (Cnidaria: Scyphozoa and Cubozoa) in Malaysia

Mohammed Rizman-Idid¹, Abu Bakar Farrah-Azwa², Ving Ching Chong²

¹ Institute of Ocean and Earth Sciences, University of Malaya, Malaysia
² Institute of Biological Sciences, University of Malaya, Malaysia

Scientific enquiries into jellyfish blooms and associated problems in Malaysia are often deterred by the lack of taxonomical and ecological studies. To date, only two scyphozoan jellyfish species have been documented from field surveys in Malaysian waters, whereas other four Malaysian scyphozoan and two cubozoan jellyfish species have been mentioned in toxicological studies, all of which their species identity were not verified. Thus, this study aimed to document and to resolve the identification of jellyfish in Malaysia using morphology and molecular DNA sequencing of COI, 16S and ITS1 regions. Jellyfish specimens were collected from 2008 to 2010 in the Straits of Malacca, South-China Sea and the Sulu-Sulawesi Sea. Ten scyphozoan and two cubozoan species were recorded, which included eight species from the order Rhizostomeae (Rhizostomatidae, Lobonematidae, Mastigiidae, Catostylidae and Cepheididae), two species from Semaestomeae (Pelagiidae and Cyaneidae) and two species from class Cubozoa; one from order Carybdeida (family Carukidae) and another from order Chiropodida (family Chiropsalmidae). The COI phylogenetic tree of Cubozoan and Scyphozoan species from the Atlantic and Pacific region showed distinct clustering of six Malaysian jellyfish species. However, most of the deeper divergences and relationships between the families were unresolved, which were also observed in the 16S and ITS1 phylogenetic trees. The Malaysian edible species Lobonemoides robustus, Rhopilema hispidum and Rhopilema esculentum were proven to belong to Rhizostomeae, whereas other scyphozoans showed phylogenetic affinities to Semaestomeae and Kolpophorae. Chrysaora and Cyanea appeared non-monophyletic, however their paraphyly was not confirmed. Although this study has provided much needed baseline information on the barcoding of Malaysian jellyfish species, there is still a general lack of jellyfish sequences in GenBank to facilitate better species confirmation.

Mohammed Rizman-Idid
Institute of Ocean and Earth Sciences, University of Malaya
Malaysia
rizman@um.edu.my
Jellyfish Studies in Malaysia

- Very few records of jellyfish in Malaysia (Straits of Malacca and South China Sea).

- Jellyfish species have been overlooked in the country's marine biodiversity checklists and surveys and the inability to identify them.

- Almost nothing is known about the diversity, biology and ecology of jellyfish in Malaysia. Even edible jellyfish were poorly documented (Daud, 1998 & Rumpet, 1991).
Jellyfish Studies in Malaysia

- Inability to identify species.
- Morphological variations among and within species.
- Their seasonal presence, which is often unpredictable and highly variable in numbers, confounds jellyfish research.

Major Concerns about Jellyfish in Malaysia

- Large blooms of jellyfish of occur seasonally, blocking power station systems and clogging fishing nets.
- Biofouling of fish cage nets by hydrozoans in mangroves has been observed, with the possibility that jellyfish abundance is increasing.
- Sporadic cases of human mortality from jellyfish stings, resulting in closure of tourist beaches.
Unverified Jellyfish Identifications

- Some edible jellyfish were documented as *Rhopilema esculenta* (Rumpet, 1991) and *Lobonema smithi* (Daud, 1998).

- Toxicological studies (Othman and Burnett 1990; Tan et al. 1993; Azila and Othman 1993):
  - Scyphozoan – *Catostylus mosaicus, Lychnorhiza lucerna, Chrysaora quinquecirrha* and *Chrysaora hysoscella*
  - Cubozoan – *Carybdea rastoni* and *Chironex fleckeri*

- Unfortunately, the species identifications of all these studies remain doubtful without satisfactory taxonomic scrutiny and verification.

Objectives

- This preliminary study aims to document and identify the jellyfish species in Malaysian waters based on their morphological characteristics and molecular DNA sequences.

- Molecular identification of species using phylogenetic approaches was based on DNA sequences of partial cytochrome oxidase I (COI), 16S and internal transcribed spacer (ITS1) regions.
MATERIALS AND METHODS

- Sampling off the coast of Malaysia (13 sites - Straits of Malacca, South China Sea, Sulu-Sulawesi Sea) from June 2008 to October 2010.
- Molecular genetic markers = COI (DNA barcoding), 16S, ITS.

Mastigia sp.  Chrysaora chinensis  Cephea cephea

Klang Strait, Selangor, Malaysia

Jellyfish Sampling

20 bag nets (in 3 transects) set across the pathway of tidal stream that comes in and out the strait.
RESULTS
8 species from the order Rhizostomeae (Rhizostomatidae, Lobonematidae, Mastigiidae, Catostylidae and Cepheidae), 2 species from Semaestomeae (Pelagiidae and Cyaneidae) and two species from class Cubozoa: 1 from order Carybdeida (family Carukiidae), 1 order Chirodropida (family Chiropsalmidae).

12 jellyfish species

Cubozoa (box jellyfish)

Morbakka sp.
Class: Cubozoa
Family: Carukiidae
Genus: Morbakka (Gershwin 2008)

Chiropsoides buitendjiki van der Horst, 1907
Class: Cubozoa (Werner 1975)
Family: Chirodropidae (Haeckel 1882)
Genus: Chiropsoides
Scyphozoa

*Acromitus flagellatus* Maas, 1903
Class: Scyphozoa
Family: *Catostylidae* (Gegenbaur 1857)
Genus: *Acromitus* (Light, 1914)

*Catostylus townsendi* Mayer, 1915
Class: Scyphozoa
Family: *Catostylidae* (Gegenbaur 1857)
Genus: *Catostylus* (Mayer 1915)

Scyphozoa

*Cephea cephea* Forskål, 1775
Class: Scyphozoa
Family: *Cepheidae* (L. Agassiz 1862)
Genus: *Cephea* (Péron and Lesueur 1809)

*Chrysaora chinensis* Linne, 1766
Class: Scyphozoa
Family: *Pelagiidae* (Gegenbaur 1856)
Genus: *Chrysaora* (Linne 1766)
**Scyphozoa**

![Image of Cyanea sp. Linne, 1758](image1)

Cyanea sp. Linne, 1758  
Class: Scyphozoa  
Family: Cyaneidae  
Genus: Cyanea (Péron and Lesueur, 1809)

![Image of Lobonemoides robustus Stiasny, 1920](image2)

Lobonemoides robustus Stiasny, 1920  
Class: Scyphozoa  
Family: Lobonematidae  
Genus: Lobonemoides (Light 1914)  
(Edible jellyfish)

**Scyphozoa**

![Image of Mastigias sp. Lesson, 1830](image3)

Mastigias sp. Lesson, 1830  
Class: Scyphozoa  
Family: Mastigiidae (Stiasny 1921)  
Genus: Mastigias (L. Agassiz 1862)

![Image of Phyllorhiza punctata von Lendenfeld, 1884](image4)

Phyllorhiza punctata von Lendenfeld, 1884  
Class: Scyphozoa  
Family: Mastigiidae (Stiasny 1921)  
Genus: Phyllorhiza (L. Agassiz 1862)
The success of PCR and DNA sequencing varied among the species and the targeted region used.

The COI - six Malaysian jellyfish species are monophyletic and show affinity to four major clades. Strong support at species nodes, but unresolved at deeper branches.
Malaysian Morbakka sp. sequences are strongly related to *Carukia barnesi*, suggesting it belongs to family Carukiidae.

"rabbit ear"- like rhopalar horns

Affinity of *P. punctata, A. flagellatus, R. esculentum and R. hispidum* to Rhizostomeae clade

Close relatedness between *R. esculentum* and *R. hispidum*. The Malaysian *R. esculentum* clusters with *R. esculentum* from China - average sequence diversity of 4.6% (+0.5).

Affinity of (*Chrysaora quinquecirrha, C. chinensis* and *Cyanea sp.*) to Semaestomeae clade

Malaysian *C. chinensis* is genetically closer to the Malaysian *Cyanea sp.* rather than reference *C. quinquecirrha* (average sequence diversity of 9.5% ±0.5).
Partial ITS1 (Bayesian tree)

These species are associated with five scyphozoan clades when rooted with the Cubozoa sequences (reference Chironex fleckeri and Malaysian C. butendijki).

Non-monophyly of the genus Cyanea?

Chrysaora appears paraphyletic?

Chrysaora chinensis from Malaysia and reference Chrysaora fulgida show close relationship with Pelagia noctiluca

The reference Chrysaora sp. related unexpectedly to the Japanese C. nozakii.

Concatenated COI and ITS1 (Bayesian tree)

Concatenated COI and ITS1 tree failed to establish the monophyly of Cyanea and Chrysaora.

The Australian Cyanea species remains separate from the Malaysian Cyanea sp. and Cyanea nozakii. Only Chrysaora sp. and Chrysaora hysoscella were available from GenBank for the total evidence molecular analysis.

Paraphyly of Chrysaora and Cyanea remains inconclusive.
Conclusion

This study has provided the much needed baseline information on the diversity of Malaysian jellyfish species which have been substantiated by partial cytochrome oxidase I (COI), 16S and internal transcribed spacer (ITS1) sequences.

A total of 12 putative species of jellyfish were identified, which encompassed 12 genera.

Although the present study could not conclude the paraphyly of Cyanea, the genetically distinct Malaysian Cyanea sp. may indicate the possibility of a new or cryptic species.

Many jellyfish species are therefore yet to be recorded and described while awaiting taxonomic identification.

Concerted global action and collaboration in jellyfish studies, including global initiatives to document jellyfish information.

Other jellyfish species

Versuriga anadyomene

Chironex sp.

Lychnorhiza malayensis
Other jellyfish species

Physalia sp.? (Siphonophore)

Cassiopea sp. Aequorea sp. (Hydrozoa)

Crambione mastigophora

Other jellyfish species

Cassiopea sp. Aequorea sp. (Hydrozoa)
Future Studies
(For Potential Collaborations)

- Regional Jellyfish Systematics & Taxonomy workshops – many species are yet to be described from Indo-Pacific/SEA.
- Jellyfish response to a changing ocean environment.
- Jellyfish culture – obtaining & maintaining jellyfish polyps
- Biochemistry/Biotechnology – bioactive compounds of marine fungi associated with jellyfish.

Acknowledgement

- This project was funded by Fundamental Research Grant Scheme (FP001/2008C) and University Malaya Research Grant (RG104/11SUS).
- Institute of Biological Sciences, University of Malaya
- Institute of Ocean and Earth Sciences, University of Malaya

Muchas gracias
rizman@um.edu.my