RINGKASAN

Persebaran dan konektivitas populasi *C. mitella* di Indonesia, khususnya di Kepulauan Maluku, belum banyak dilaporkan sehingga perlu dilakukan penelitian untuk mendukung ketersediaan data genetik yang dibutuhkan untuk pengembangan penelitian selanjutnya. Pendekatan secara molekuler diperlukan pada studi konektivitas populasi untuk menghindari bias faktor lingkungan. Salah satu marka molekuler yang biasa digunakan dalam studi genetika populasi hewan adalah sitokrom oksidase I (COI). Oleh karena itu, penelitian ini dilakukan pada populasi *C. mitella* di Kepulauan Maluku menggunakan marka COI dengan tujuan untuk mengetahui (1) keanekaragaman genetik *C. mitella*, (2) ada tidaknya hubungan antara jarak genetik dan jarak geografis populasi *C. mitella*, (3) filogeografi populasi *C. mitella*,

Penelitian dilakukan di Laboratorium Biosistematika dan Evolusi Badan Riset dan Inovasi Nasional selama empat bulan, dari bulan Januari 2023 hingga April 2023. Sampel *C. mitella* diambil secara acak dari enam lokasi yaitu Saparua, Morella, Tawiri, Dermaga Liang, Wakasihu, dan Pantai Akarica Ternate. Secara garis besar, prosedur penelitian terdiri atas ekstraksi DNA genomik menggunakan Qiagen Blood and Tissue Kit serta amplifikasi dan sekuensing marka COI. Marka COI mengunakan sepasang primer universal, yaitu FSrCOI (5'- CAG CAA TAG TTG GGA CAG CTC -3') dan RSrCOI (5'- CTC CGG CTA AAA CTG GTA AGG -3'). Sekuensing marka COI dilakukan dengan metode dideoksi Sanger yang diotomasi dengan pelabelan pada terminator.

Data sekuen amplikon disunting menggunakan Program Bioedit versi 7.0.4.1, yang disertai dengan pemeriksaan manual. Analisis penjajaran sekuen dilakukan menggunakan Program *Clustal W*, yang juga diimplementasikan dalam Bioedit versi 7.0.4.1. Nilai keanekaragaman haplotipe *(h)* dan keanekaragaman nukleotida (π) digunakan sebagai parameter keanekaragaman genetik. Analisis konektivitas populasi *C. mitella* dilakukan menggunakan AMOVA (*Analysis of molecular variance*). Pohon filogenetik dikonstruksi menggunakan analisis Neighbor-Joining, Maximum Likelihood, dan Maximum Evolution menggunakan aplikasi MEGA 5.0. Jaringan haplotipe *C. mitella* dibuat menggunakan piranti lunak Network 5.0.0.3.

Ada tidaknya tekanan seleksi terhadap populasi *C. mitella* dianalisis menggunakan uji netralitas *Fs* Fu dan *D* Tajima. Penghitungan nilai *h* dan π, AMOVA, serta uji netralitas *Fs* Fu dan *D* Tajima dilakukan menggunakan Program *Arlequin* versi 3.5.

Hasil penelitian menunjukkan nilai diversitas haplotipe dari 18 sampel yang di dapat sebesar 1 pada seluruh sampel, kecuali pada sampel Saparua sebesar 0.67. Sementara itu, nilai diversitas nukleotida pada semua sampel lebih kecil dari 0.5. Nilai indeks fiksasi (Fst) tertinggi diperoleh antara Ternate dan Saparua, yakni sebesar 0.31081. Secara keseluruhan didapatkan nilai Fst yang rendah. Pohon filogeni yang terbentuk menunjukkan perbedaan klade populasi Perairan Maluku dari populasi Cina, Taiwan, dan Korea. Berdasarkan hasil penelitian tersebut dapat disimpulkan bahwa terdapat hubungan kekerabatan genetik yang dekat di antara haplotipe-haplotipe yang ditemukan, tidak ada hubungan antara jarak genetik dan jarak geografis pada populasi *C. mitella* di Perairan Maluku karena adanya konektivitas yang tinggi di antara semua lokasi pengambilan sampel, dan populasi *C. mitella* di Perairan Maluku tidak memiliki hubungan evolusi dengan populasi *C. mitella* di Cina, Taiwan, dan Korea.

Kata Kunci: *Capitulum mitella*, kajian genetik, Kepulauan Maluku

SUMMARY

The distribution and connectivity of *C. mitella* populations in Indonesia, especially in the Maluku Islands, have not been frequently reported, so that studies supporting the availability of genetic data required for further development are needed. Molecular approaches are needed in the studies of population connectivity to avoid environmental bias. One of the molecular markers commonly used in the studies of animal population genetics is cytochrome oxidase I (COI). This study on *C. mitella* populations in the Maluku Islands using COI markers aims to assess (1) genetic diversity of *C. mitella*, (2) the existence of relationship between genetic and geographic distance of *C. mitella* populations, (3) population phylogeography of *C. mitella*,

The study has been conducted at the Biosystematics and Evolution Laboratory of the National Research and Innovation Agency for four months, from January 2023 to April 2023. *C. mitella* samples were taken randomly from six locations in the Maluku Islands, namely Saparua, Morella, Tawiri, Liang Harbour, Wakasihu and Ternate Akarica Beach. In outline, the procedures consisted of genomic DNA extraction using the Qiagen Blood and Tissue Kit as well as amplification and sequencing of COI markers. A pair of universal primers were used to amplify COI markers, namely FSrCOI (5'- CAG CAA TAG TTG GGA CAG CTC -3') and RSrCOI (5'- CTC CGG CTA AAA CTG GTA AGG -3'). COI marker sequencing was carried out using the dideoxy Sanger method automated with terminator labeling.

Data on the amplicon sequences were edited using the Bioedit Program version 7.0.4.1, followed by manual checking. Sequence alignment analysis was performed using the Clustal W Program, which was also implemented in the Bioedit version 7.0.4.1. Haplotype diversity (h) and nucleotide diversity (π) values were used as genetic diversity parameters. Analysis on the connectivity of *C. mitella* populations was performed using AMOVA. Phylogenetic trees were constructed using Neighbor-Joining, Maximum-Likelihood and Maximum Evolution analysis. The *C. mitella* haplotype network was created using Network 5.0.0.3 software. The existence of selection pressure on the *C. mitella* populations was analyzed using the

Fs Fu and *D* Tajima neutrality tests. Calculation on *h* and π values, AMOVA, as well as neutrality tests of *Fs* Fu and *D* Tajima was carried out using the Arlequin Program version 3.5.

The results showed haplotype diversity values of 1 for all samples, except for that of the Saparua, which was 0.67. Meanwhile, nucleotide diversity values for all samples were below 0.5. The highest fixation index (Fst) was found between Ternate and Saparua, i.e. 0.31081. Overall, low Fst values were observed. The phylogenetic tree constructed showed the difference between clades of populations in the Moluccas waters and those of China, Taiwan and Korea. Based on the results, it can be concluded that close genetic relationship among haplotypes was observed, no relationship between genetic and geographical distance in the population of *C. mitella* in Maluku waters was obtained, and the population of *C. mitella* in Maluku waters has no evolutionary relationship with those in China, Taiwan and Korea.

Keyword: *Capitulum mitella*, genetic study, Maluku Islands