

ABSTRACT

Syafiq Pahlevi Kanigoro. 24020121140211. **Isolation and Molecular Identification of PUFA-Producing *Thraustochytrids* from Mangrove Water Samples of Glagah Wangi Beach Demak Using 18S rRNA Gene.** Under the guidance of Nurhayati and Dwi Retno Fatmawati.

Thraustochytrids are heterotrophic marine protists known for producing high-value lipids, particularly polyunsaturated fatty acids (PUFAs). This study aimed to isolate, identify, and evaluate the lipid-producing potential of Thraustochytrids obtained from the mangrove ecosystem of Glagah Wangi Beach, Demak. Isolation was conducted using the pollen baiting method on GYP medium supplemented with antibiotics. Lipid screening was performed through Sudan Black B staining, while molecular identification was carried out using PCR amplification of the 18S rRNA gene with LABY-A and LABY-Y primers, followed by BLAST analysis and phylogenetic reconstruction using MEGA 11. The study successfully obtained four pure isolates (TCS3, TCS4, TCS5, TCS6) with morphological variations in their colonies. Sudan Black B staining revealed that isolate TCS3 had the highest lipid accumulation. Nanodrop analysis showed DNA purity of 2.19, and PCR amplification produced a DNA band of approximately 400 bp. BLAST analysis identified isolate TCS3 with 88.72% similarity to *Thraustochytriaceae* sp., supported by phylogenetic analysis with a bootstrap value of 74%. *Thraustochytriaceae* sp. are capable of producing polyunsaturated fatty acids (PUFAs) such as DHA and EPA, which hold significant value for the food and health industries. Overall, the findings indicate that local Thraustochytrids possess potential to be developed as a biotechnological source of valuable lipids. .

Keywords: *Thraustochytrids*, Lipid, 18S rRNA, mangrove, Demak.