



Distribution of Pacific bluefin tuna (*Thunnus orientalis*) eggs and larvae in Korean waters

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Introduction

To identify spawning grounds of fish species in Korean waters, extensive surveys of fish eggs and larvae have been conducted since 2017 (Fig. 1). From 2017 to 2020, surveys were carried out over a broad spatial range encompassing Korean waters. Subsequently, from 2021 to 2022, sampling efforts were focused on the waters around Jeju Island and the East Sea, where a high diversity of fish eggs and larvae had been observed. Since 2023, surveys have been conducted from May to July in waters extending from south of Jeju Island to the Kuroshio Current-influenced regions, including Ulleungdo and Dokdo.

Pacific bluefin tuna is a large pelagic species belonging to the family Scombridae. This species generally reaches sexual maturity at 3–6 years of age, has a long lifespan of up to 15 years, and typically attains a fork length of approximately 200 cm, with a maximum length exceeding 300 cm (Fujioka et al., 2015; Froese and Pauly, 2025). Due to its rapid growth, longevity, and wide distribution throughout the Pacific Ocean, *T. orientalis* is one of the most commercially important fish species in the Pacific region (Collette and Nauen, 1983; Tanaka et al., 2007). Previous studies based on histological analyses of gonads have reported that Pacific bluefin tuna spawn from April to June in the waters extending from north of the Philippines to the Ryukyu Islands (Chen et al., 2006; Ashida et al., 2015), and from June to August in the southern region of the East Sea (Okochi et al., 2016). These findings indicate that Pacific bluefin tuna form distinct spawning grounds depending on spawning period and region, and further suggest the potential formation of additional spawning grounds.

The objective of the present study is to identify spawning grounds of Pacific bluefin tuna in Korean waters by examining the spatial and temporal distributions of their eggs and larvae.

Data and Methods

Sampling

Fish eggs and larvae were collected aboard research vessels of the National Institute of Fisheries Science using a Bongo net (mouth diameter: 80 cm; mesh size: 500 μm) and a multiple opening/closing net and environmental sensing system (MOCNESS; mouth area: 1 m^2 ; mesh size: 330 μm). Sampling was conducted by oblique tows across discrete depth layers, including surface–10, 10–20, 20–30, 30–40, 40–60, 60–80, and 80–110 m. Immediately after collection, fish eggs and larvae were fixed in 10% seawater formalin for 10 min and subsequently transferred to 99% ethanol for long-term preservation.

Morphological and molecular identification

Fish eggs were examined under a stereomicroscope (SZH-16; Olympus, Tokyo, Japan) and sorted into size classes, except for anchovy and bullet tuna eggs that could be reliably identified based on morphological characteristics. Larvae were also examined under a stereomicroscope, and species identification was conducted to the lowest possible taxonomic level based on external morphological features following Okiyama (2014).

When larval specimens were damaged during sampling or when morphological characteristics were insufficiently developed for reliable identification, molecular analysis was performed to confirm species identity. Molecular identification of larvae was conducted using the right eye of each specimen (Lee et al., 2019). Fish eggs were grouped according to size, and molecular analysis was performed by randomly selecting five individuals from each size group (Lin et al., 2016). If all five individuals were identified as belonging to the same species, the remaining eggs in that group were assigned to that species. In cases where the five selected individuals were identified as different species, the remaining eggs were reclassified based on morphological characteristics and subjected to repeated molecular analyses. This procedure was repeated until all five selected individuals were confirmed to belong to a single species.

Total genomic DNA was extracted using a GeneAll Exgene Clinic SV DNA Extraction Kit. A partial fragment of the mitochondrial cytochrome oxidase subunit I (COI) gene was amplified using universal primers (Ward et al., 2005). Polymerase chain reaction (PCR) was performed under standard conditions, and the amplified PCR products were sequenced using an ABI PRISM BigDye Terminator v3.1 Ready Reaction Cycle Sequencing Kit (Applied Biosystems Inc., Foster City, CA, USA) on an ABI 3730xl DNA Analyzer (Applied Biosystems Inc.). Obtained sequences were identified by comparison with reference sequences deposited in GenBank (National Center for Biotechnology Information) and the Barcode of Life Data System.

Results and Discussion

Larval Pacific bluefin tuna are morphologically similar to other scombrid species, making species-level identification based solely on external morphology difficult. Therefore, molecular analysis was conducted following morphological identification to ensure accurate species determination. As a result, a 419 bp fragment of the mitochondrial cytochrome oxidase subunit I (COI) gene was obtained. Intraspecific genetic distances within *Thunnus orientalis* ranged from 0 to 0.2%, whereas interspecific distances between

T. orientalis and *Thunnus albacares*, *T. obesus*, and *T. alalunga* ranged from 1.7–1.9%, 2.0–2.7%, and 4.7–5.0%, respectively. These clear genetic differences confirmed that molecular analysis enabled reliable identification of Pacific bluefin tuna larvae (Fig. 2).

Based on Bongo net sampling, Pacific bluefin tuna eggs were first collected in waters near Dokdo in 2021, while larvae were initially recorded from the eastern coastal waters of Jeju Island. Thereafter, both eggs and larvae of Pacific bluefin tuna have been continuously observed in Korean coastal waters through 2025. In particular, compared with the results from 2021, samples collected in 2025 exhibited an expanded spatial distribution and an increasing trend in abundance (Fig. 3).

Although direct interannual comparisons are limited due to differences in sampling stations and survey periods among years, the number of stations where Pacific bluefin tuna eggs were collected increased markedly to 24 station in 2023, compared with only one station in 2022. Similarly, the number of stations where larvae were collected increased to 23 station in 2025, compared with six stations in 2024. In addition, substantial increases were observed in the number of individuals collected per station between 2021 and 2025. Egg density increased from 34 inds./1,000 m³ in 2021 to 5,195 inds./1,000 m³ in 2025, while larval density increased from 15 inds./1,000 m³ to 1,316 inds./1,000 m³. These results suggest that spawning grounds of Pacific bluefin tuna have been consistently formed in the waters around Jeju Island and the East Sea since 2021, with both the spatial extent of spawning grounds and the number of participating individuals increasing over time.

Results from MOCNESS sampling conducted from the surface to 110 m depth revealed that Pacific bluefin tuna eggs were distributed primarily in the surface–10, 20–30, 30–40, and 60–80 m depth layers (Fig. 4). The highest egg density was observed in the surface–10 m layer, reaching 5.0 inds./1,000 m³ (74.9%), whereas substantially lower densities were recorded in the remaining depth layers. Larvae were distributed from the surface to 30 m depth layer, with the highest density observed in the 10–20 m depth layer, reaching 230.0 inds./1,000 m³ (86.0%).

In this study, the first occurrence of Pacific bluefin tuna eggs and larvae in Korean waters was confirmed in 2021, indicating the formation of spawning grounds in the waters around Jeju Island and the East Sea. To more accurately define the spatial extent and timing of these spawning grounds, continued monitoring is planned.

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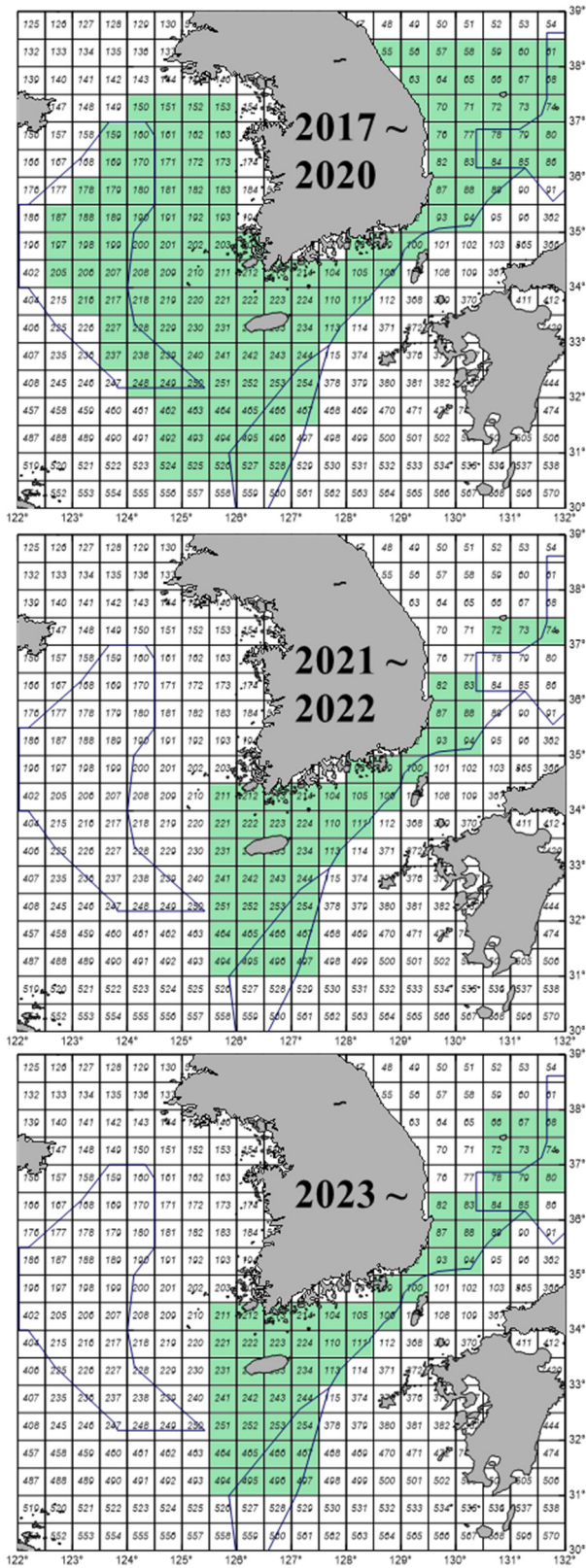


Fig. 1. Schematic overview of the three paradigms based on survey time points (2017-2020, 2021-2022, and 2023 survey station).

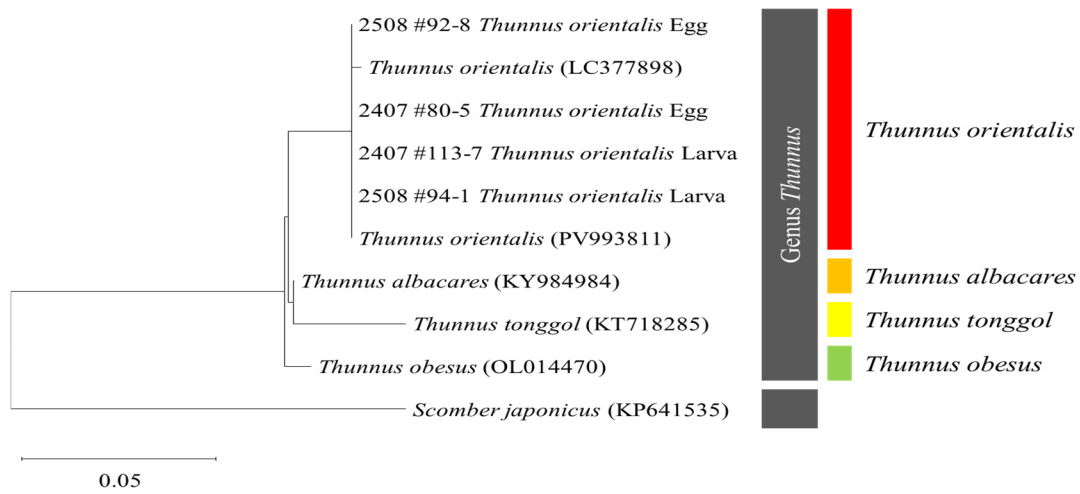


Fig. 2. A neighbor-joining tree based on partial mtDNA COI region using *Thunnus orientalis*, showing the relationships among four species of *Thunnus* and one outgroup (*Scomber japonicus*). Numbers at branches indicate bootstrap probabilities in 10,000 bootstrap replications. Scale bar equals 0.05 of Tamura and Nei's distance (1993) with K2 parameter model. COI, cytochrome c oxidase subunit I.

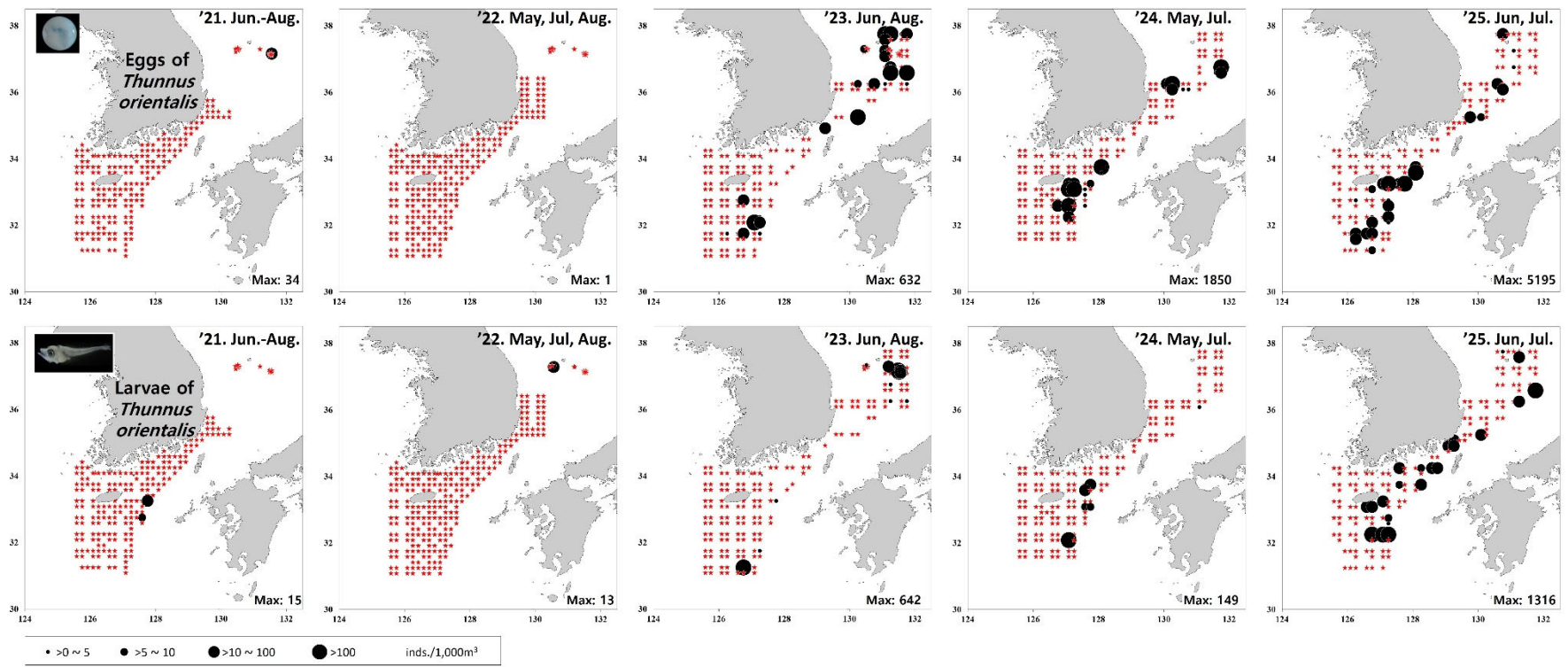


Fig. 3. Spatial distribution of Pacific bluefin tuna (*Thunnus orientalis*) eggs and larvae from 2021 to 2025.

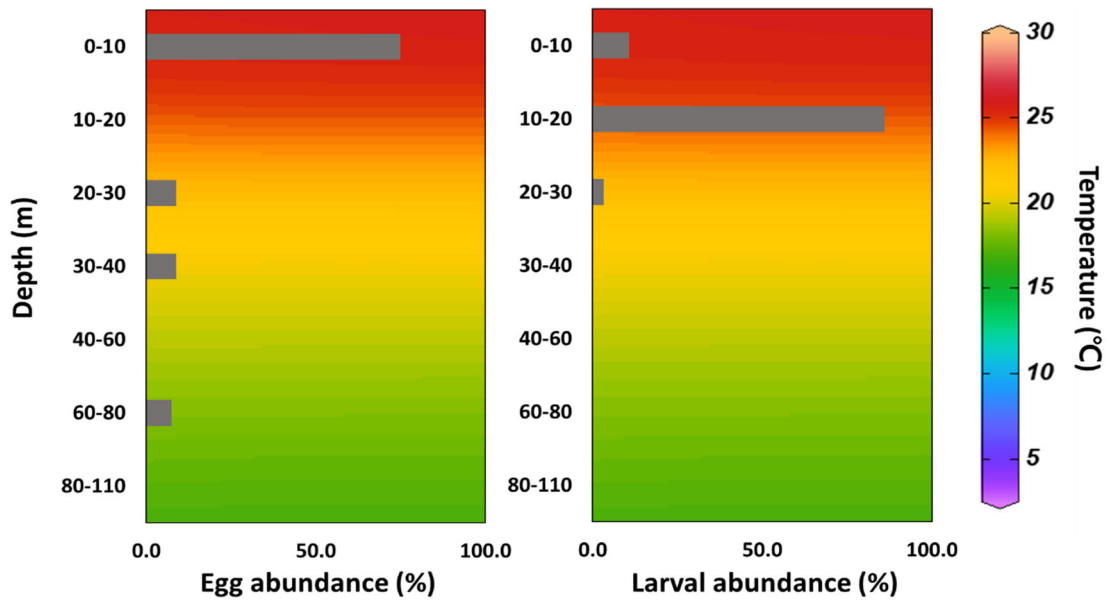


Fig. 4. Relative abundance (%) of Pacific bluefin tuna (*Thunnus orientalis*) eggs and larvae across seven depth layers, collected using MOCNESS (multiple opening/closing net and environmental sensing system) at eight stations in July 2025.