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Population abundance and growth parameters of an exotic bivalve species, *Anadara kagoshimensis*, in the Southwestern Black Sea

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Abstract: Blood cockle (*Anadara kagoshimensis*) is an Indo-Pacific species that later entered the Black Sea. The abundance of *A. kagoshimensis*, which is not subjected to commercial fishing, is important in terms of food competition with other bivalvia species. Baby clam (*Chamelea gallina*) together with the *A. kagoshimensis* are dominant bivalve species found in the sandy and muddy areas off the coastal waters of the Black Sea. In this study, specimens of *A. kagoshimensis* have been recognized by morphological analysis and also confirmed by molecular characterization. Furthermore, the abundance and growth parameters of *A. kagoshimensis* were investigated in the Southwestern Black Sea. Blood cockles were sampled between February 2011 and December 2012, seasonally. According to the Von Bertalanffy Growth Parameters (VBGP) the results were $L_{\infty} = 81.96$ mm, $K = 0.32$ year⁻¹, $t_0 = -0.22$ year, and nonseasonal $L_{\infty} = 84.32$ mm, $K = 0.31$ year⁻¹, $t_0 = -0.21$, $WP = 0.65$, $t_s = 0.15$. The growth pattern showed the slope $[b] = 2.96-3.01$ in 2011 and 2012. The stock size was estimated according to two different years in 5 different subareas (Cide, İnebolu, Türkel, Ayancık, and Sarıkum) and by 4 different strata (0–5 m, 5–10 m, 10–15 m, and 15–20 m). Considering subareas, the *A. kagoshimensis* population in all subareas increased significantly in a single year. Compared to other regions, İnebolu was the main highly distributed area of the *A. kagoshimensis*, and also the estimated stock size was the highest in the region. Due to food competition with other commercial species (mainly *Chamelea gallina*) *A. kagoshimensis* is an ecology important species for the Southern Black Sea habitats. It is aimed to make contributions to Good Environmental Status (GES) and fisheries management in the region.

Key words: *Anadara kagoshimensis*, Black Sea, abundance, biomass, growth parameters

1. Introduction

The blood cockle or ark clam (*Anadara kagoshimensis* (Tokunaga, 1906)), which belongs to the family Arcidae, has been introduced into the Black Sea in the ballast water in the 1960s (Zolotarev and Zolotarev, 1987; Şahin et al., 1999; Şahin et al., 2006). *A. kagoshimensis* is included in the 100 alien invasive species for the Mediterranean Sea and Black Sea (Streftaris and Zenetos, 2006). *A. kagoshimensis* are spread in Indo-West Pacific, Mediterranean, and the Black Sea. It was first encountered in the Black Sea in 1968 around the coasts of the Caucasus (Şahin et al., 2006). In later years, this species was distributed in all coastal areas in the Black Sea (Băncilă et al., 2022). Together with the species that entered the Black Sea ecosystem, later on, it caused important changes in this ecosystem (Zolotarev, 1996). *A. kagoshimensis* is generally encountered in areas between 0 and 30 m in soft-muddy, sandy-muddy, and

crusted-sandy habitats (Şahin et al., 2009). The spawning season is from June to September and individuals with a size of 20 mm are considered to have reached maturity on the Southern Black Sea coast (Şahin et al., 2006). There are commercially important species of blood cockle in the Far-East countries; however, there are no economically important species in the Black Sea (Acarlı et al., 2012; Mirzai et al., 2015).

Chamelea gallina and *A. kagoshimensis* are two dominant bivalvia species in the South Black Sea. These two species are the most important prey of alien gastropod species, *Rapana venosa* (Savini and Occhipinti-ambrogi, 2006). The competition with striped venus clam (*C. gallina*) for nutrients, as well as the impact on other noneconomic bivalvia species within the ecosystem, is important. Although it is not a commercial species, it is one of the important bycatch species in baby clam (Dalgıç

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and Ceylan, 2012) and rapa whelk fisheries (Erik et al., 2020) in the Black Sea. Especially, baby clam fishing is not selective for blood cockle (Dalgıç and Ceylan, 2012). Therefore, fisheries' pressure has an effect on these stocks.

It is important to constantly monitor the biological and ecological relationships of the blood cockle (Aydın et al., 2014). The largest body of studies on blood cockles in the South Black Sea has focused on biology and population parameters (Şahin, 1995; Şahin et al., 1999; Şahin, 2006; Şahin et al., 2009; Aydın et al., 2014). In this study, abundance and growth parameters of *A. kagoshimensis*, exotic species which are distributed in the Southwestern Black Sea, were tried to be determined.

2. Materials and methods

2.1. Sampling and survey area

Surveys were done between July 2011 and 2012. In addition, seasonal samplings were made for growth parameters in each subarea. The study covered the areas between Sarıkum (Sinop) and Cide (Kastamonu) in the western Black Sea (Latitude: 41°54'31.68" - 42°6'28.96" N, Longitude: 32°44'44.95" - 34°55'50.26 E). The survey area was divided into five subareas, based on the region's structure. The subregions are Sarıkum, Ayancık, Türkeli, İnebolu, and Cide. The coastal length between these areas is approximately 90 nautical miles. The study area covers up to 20 m in depth. The research area was divided into four stratum according to depth: Stratum 1 (0–5 m), Stratum 2 (5–10 m), Stratum 3 (10–15 m), and Stratum 4 (15–20 m) (Figure 1). The hydraulic dredge method was used because it is the ideal sampling gear for the estimation of bivalvia biomass. The mouth opening of the hydraulic dredge was 350 cm and the length was 300 cm. Samples collected from the hydraulic dredge were placed directly into the collection box without

sieving (Figure 2). The length, height, and width of the *A. kagoshimensis* specimens were measured with digital calipers (0.01 mm). Once the dredge was lowered into the sea and reached the bottom, the boat moved at a speed of approximately 2.0 knots and another engine started to inject water up to 3 bar pressure into the dredge with the help of a nose. Each haul took 2 min. Following hydraulic dredge sampling at sea, 10 kg of subsamples were obtained. Subsamples were identified and weighed (0.01 g) at the Sinop University Fishery Faculty laboratory. Acquired data were processed into the relevant form. In this study, data was collected from 174 stations selected to represent the continental shelf of the western region of Turkey.

2.2. Estimation of *A. kagoshimensis* biomass

The *A. kagoshimensis* catch per unit area (km²) was calculated by taking standard dredge shots. The total stock size for the entire study area was calculated. The formula proposed by Sparre and Venema (1992) was used to determine the area covered by the hydraulic dredge in 1 h towards calculation of the size of the swept area. The formulas for average biomass and total biomass per unit area are given below:

$$Ca = Cw/a \quad b = (Cw/a)/X_1,$$

where Cw is the weight of *A. kagoshimensis* in one sampling, a is the area scanned in one haul (km²), Ca is the catch per unit area (kg km⁻²) in one haul, b is the biomass per unit area (kg km⁻²), and X_1 is the coefficient of catch. The hydraulic dredge coefficient of fishing (X_1) has been accepted as "1". The biomass

$$B_i = \frac{A_i}{X_1} * \frac{1}{n} * \sum_{i=1}^n Ca(i) = \frac{A_i}{X_1} * \overline{Ca}$$

The stock size was estimated according to the distributed areas of *A. kagoshimensis* (Table 1).

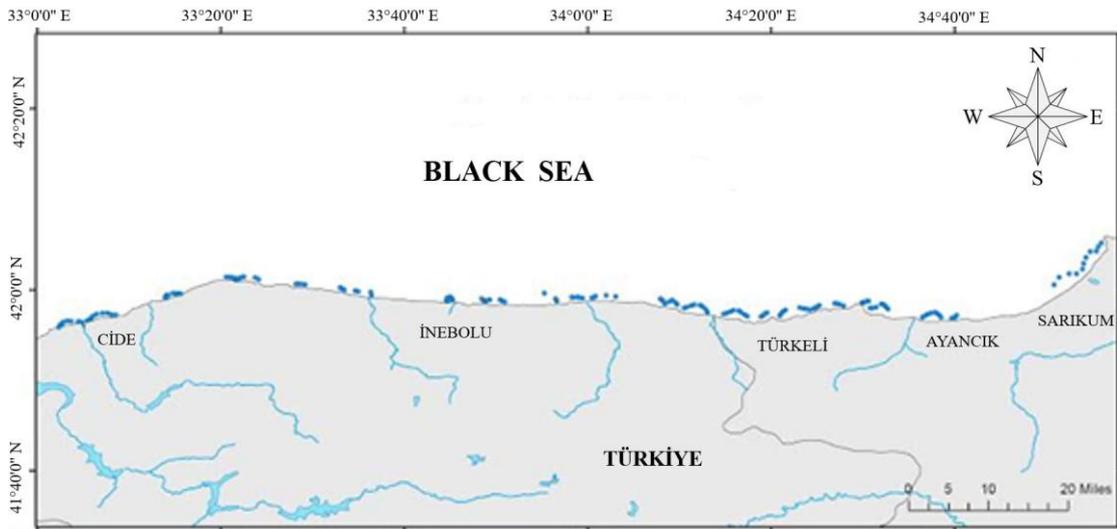


Figure 1. Map showing the study areas and sampling stations in the Southwestern Black Sea



Figure 2. Sampling gear (hydraulic dredge).

Table 1. Area covered by *A. kagoshimensis* beds according to subareas and number of hauls.

| Subarea | Depth (m) | Number of hauls | Area (km ²) |
|---------|-----------|-----------------|-------------------------|
| Cide | 15–20 | 13 | 13.75 |
| | 10–15 | 13 | 13.09 |
| | 5–10 | 12 | 17.29 |
| | 0–5 | 11 | 8.94 |
| İnebolu | 15–20 | 15 | 24.24 |
| | 10–15 | 12 | 25.42 |
| | 5–10 | 9 | 56.65 |
| | 0–5 | 5 | 21.5 |
| Türkeli | 15–20 | 12 | 35.44 |
| | 10–15 | 13 | 28.26 |
| | 5–10 | 10 | 16.15 |
| | 0–5 | 11 | 12.97 |
| Ayancık | 15–20 | 6 | 15.44 |
| | 10–15 | 6 | 10.96 |
| | 5–10 | 8 | 7.22 |
| | 0–5 | 6 | 6.62 |
| Sarıkum | 15–20 | 3 | 2.25 |
| | 10–15 | 3 | 1.91 |
| | 5–10 | 3 | 1.16 |
| | 0–5 | 3 | 0.82 |

$$a = D * hr * X_2,$$

where a is swept area, X_2 is a fraction expressing the width of the area swept by the net divided by the length of the head rope, hr, which is equal to width of path swept by the trawl, the “wing spread”, D is the distance covered, and h is the length of the head-rope. The towing distance was estimated in units of km² (1 n.mil = 1.852 km), by:

$$D = 60 * \sqrt{(Lat1 - Lat 2)^2 + (Lon1 - Lon2)^2 * \cos^2(0.5 * (Lat1 + Lat2))}$$

Lat1 is latitude at the start of the haul (degrees), Lat2 is the latitude at the end of the haul (degrees), Lon1 is the longitude at the start of the haul (degrees), and Lon2 is the longitude at the end of the haul (degrees). The stock size was estimated according to the area distribution of *A. kagoshimensis* (Table 1). Ocean Data View (ODV, version 5.3) program was used to map the biomass distribution. For this, advanced DIVA gridding software (Schlitzer, 2015) was used. A three-way analysis of variance was performed using the SPSS software package (version 16) to detect differences between years, subareas, and depths. The significance level was accepted as $p < 0.05$ for all statistical analyses (Zar, 1984). Data were analyzed in R (R Core Team, 2020).

2.3. Growth

Length–weight relationships of *A. kagoshimensis* were examined using the equation below (Le Cren, 1951):

$$W = aSL^2 \text{ or } \log W = \log(a) + b(\log SL)$$

where a and b are regression constants, a is intercept, b is slope, W is total body weight (g), and SL is total length (mm). In order to determine the growth pattern, a and b values were found by subjecting the length and weight to regression analysis. With the found ‘b’ value, it was found that the growth is isometric ($H_0, b = 3$) or allometric with the formula $ts = (b-3) / S_b$ (Sokal and Rohlf, 1987) where ts denotes the Student’s t-test value, b represents slope, and S_b signifies standard error of “b” value. For the calculation of shell length–height and height–width relationships, the formula (Arneri et al., 1998) was used;

$$H(W) = a + b(SL)$$

where H is height (mm), W is width (mm), and SL is total length (mm).

The von Bertalanffy growth parameters (VBGP) equation estimated length as a function of age and is used when growth has a nonseasonal pattern. Length classes were taken as 2 mm.

$$L_t = L_{\infty} [1 - e^{-K(t-t_0)}]$$

For seasonal growth, the Hoenig and Hanumara (1982) version of the VBG equation was used.

$$L_t = L_{\infty} [1 - e^{-K(t-t_0) + (C(K/2\pi)\sin 2\pi(t-ts) - (C(K/2\pi)\sin 2\pi(t_0-ts))}]$$

where L_t is the length at age t, L_{∞} is the asymptotic length to which the blood cockle grows, K is the growth-rate parameter, C is the relative amplitude ($0 \leq C \leq 1$) of the seasonal oscillations, t_0 is the theoretical age when

the SL is zero (years), and t_s is the phase of the seasonal oscillations ($-0.5 \leq t_s \leq 0.5$), which denotes the time of the year that corresponds to the start of the convex segment of sinusoidal oscillation. The time of the year when growth is slowest, known as the winter point (WP), was calculated as ($WP = t_s + 0.5$).

On the basis of seasonal sampling, frequency was 7 for time series datasets (1 mm SL size classes). Electronic length frequency analysis (ELEFAN) used the procedure in the length–frequency distribution analysis (LFDA) program (Kirkwood et al., 2001). The length was predicted as a function of age according to the von Bertalanffy growth equation (VBGP). This equation is used when a nonseasonal growth pattern is observed. This study conducted by Hoenig and Hanumara (1990) found the Hoenig and Hanamara (1982) model used for seasonal growth data. Maximization has been done on the small area ($0.1 < K < 0.5 \text{ year}^{-1}$ and $60 < L_{\infty} < 90 \text{ mm}$) in order to obtain the highest score function possible. The growth performance index was compared using different growth values reported in the literature according to Munro and Pauly (1983) formula:

$$\Phi = 2\log(L_{\infty}) + \log(K)$$

The maximum lifespan (A_{95}) was calculated as the inverse of the VBGP, where we considered the maximum SL as 95% of the L_{∞} (Taylor, 1958):

$$A_{95} = t_0 + (2.996 / K)$$

The Beverton–Holt was calculated mortality rate (Z) LFDA vers. 5.0 (Kirkwood et al., 2001).

$$Z = K + [(L_{\infty} - L) / (L - L')]]$$

L' is the length when *A. kagoshimensis* were first recruited and L is the mean length of all cockles longer than L' . The natural mortality rate was estimated using the empirical relationship defined by Pauly (1980):

$\log M = -0.0066 + 0.279 \log L_{\infty} + 0.6543K + 0.4634 \log T$, where T is mean annual seawater temperature (16 °C) and L_{∞} is the asymptotic shell length (mm) that *A. kagoshimensis* can reach. This empirical equation assumes that the length is measured as L is mm (Gayani et al., 2005).

Blood cockle’s abundance and biomass were held in July, which is also the breeding season. Hydraulic dredge was used because it is the ideal sampling tool in bivalves biomass estimation. In periods other than the summer period, it is aimed to take only biological data. The mouth opening of the hydraulic dredge was 350 cm and the length was 300 cm (Figure 2). Samples collected from the hydraulic dredge were placed directly into the collection box without sieving. Length, height, and width of the blood cockle specimens were measured with digital calipers (0.01 mm). Once the dredge lowered into the sea and reached the bottom, the boat moved at a speed of approximately 2 knots and another engine started to inject water up to 3 bar pressure into the dredge with the help of a nose. Each operation took 2 min.

2.4. DNA isolation and amplification

DNA was extracted from muscle tissue of six specimens stored at 96% ethanol by QIAamp DNA isolation kit according to the manufacturer's instructions. A partial region of Cytochrome c oxidase subunit I (COI) gene was amplified by primer set of Matsumoto (2003) (5'-ATY GGN GGN TTY GGN AAY TG-3' and 5'-ATN GCR AAN ACN GCN CCY AT-3').

The PCR reactions were performed in a total volume of 20 µL containing 1.5 µL of DNA, 10 µL of Taq PCR Master Mix (QIAGEN), 7.5 µL of nuclease-free water, and 0.5 µL of each primer. Amplification was conducted using the following protocol: initial denaturation at 95 °C for 3 min, followed by 38 cycles of 30 s at 95 °C, 30 s at 48 °C, and 45 s at 72 °C, and a final extension at 72 °C for 7 min. PCR products were visualized by electrophoresis in 1.5% (w / v) agarose gel with 1 × TBE (Tris-Borate-EDTA) buffer containing Et-Br and DNA fragment length was estimated with the migration of 100-bp DNA ladder (Bio Basic). PCR products were sequenced using the BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems) in ABI 3500 Genetic analyzer, according to the manufacturer's instructions.

2.5. Sequence analysis

Three *A. kagoshimensis* specimens out of six were successfully sequenced and the results were manually edited using BioEdit Sequence Alignment Editor software (v. 7.1.11) (Hall, 1999). Edited sequences were compared to published sequences in Genbank (<http://www.ncbi.nlm.nih.gov/genbank/>) and the Barcode of Life Data System (BOLD; <http://www.boldsystems.org>) by BLAST algorithm (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>).

In order to support taxonomic identification, a phylogenetic tree was constructed using sequences from the present study and COI gene sequences of *A. kagoshimensis* (Accession no. KJ490941, KJ490940, HQ258851), *A. kagoshimensis* (Accession no. HQ258858, AB076937), and also *Arca avellana* (Accession no. HM180483) as outgroup from GenBank. Multiple sequences were aligned by CLUSTAL W using BIOEDIT software (v. 7.1.11) (Thompson et al., 1994). The phylogenetic tree was conducted using UPGMA method by maximum composite likelihood model with a gamma-distributed parameter in MEGA-X software (Sneath and Sokal, 1973; Kimura et al., 2018). The confidence of tree topology was tested by bootstrap analysis with 1000 replicates (Felsenstein, 1985).

3. Results

3.1. Evaluation of stock biomass in the region

Results showed that *A. kagoshimensis* stock size increased 3 times from 2011 to 2012 and reached 5800 t in total during the sampling period. In 2011, the highest stock size was estimated in İnebolu (1634 t), while the lowest was in Sarıkum (24.4 t). Estimated biomass values showed significant differences between the years and subarea, but no significant differences were found among depths for overall results ($F(12, 270) = 0.728, p = 0.724$), (Tables 2 and 3; Figures 3–5).

3.2. Length, height, width-weight relationships

Tables 4 and 5 shows the descriptive statistics and estimated parameters of length weight relationship (LWRs) a and b, 95% confidence limits for a and b, and coefficient of determination (R^2) for the *A. kagoshimensis* in the Southwestern Black Sea.

Table 2. Estimation biomass of *A. kagoshimensis*.

| Year | Subarea | Biomass (t) | Biomass confidence interval (t) |
|--------------|---------|-------------|---------------------------------|
| 2011 | Cide | 468.6 | 1152.8 |
| | İnebolu | 1090.6 | 1634.3 |
| | Türkeli | 388.7 | 619.7 |
| | Ayancık | 11.3 | 25.2 |
| | Sarıkum | 5.7 | 24.4 |
| Total | | 1964.9 | 3456.4 |
| 2012 | Cide | 1595.5 | 1404.4 |
| | İnebolu | 3090 | 3672.9 |
| | Türkeli | 754 | 864.9 |
| | Ayancık | 229.7 | 420.4 |
| | Sarıkum | 131.4 | 454 |
| Total | | 5800.7 | 6816.6 |

Table 3. Results of three-way ANOVA testing for biomass (kg km^{-2}) with years, subarea, and depth as fixed factors: (df: degrees of freedom, SS: sum of squares, MS: mean square, F: F-value, significance level (* $p < 0.05$; ** $p < 0.01$)).

| Source | SS | df | MS | F | Significance level |
|---------------------|------------------|-----|----------------|--------|--------------------|
| Intercept | 26263429549.542 | 1 | 26263429549.54 | 24.077 | ** |
| Year | 5941384015.047 | 1 | 5941384015.04 | 5.447 | * |
| Subarea | 12899684917.722 | 4 | 3224921229.43 | 2.956 | * |
| Depth | 2107945235.982 | 3 | 702648411.994 | .644 | |
| Year*Subarea | 1203709613.799 | 4 | 300927403.450 | .276 | |
| Year* Depth | 6286768809.179 | 3 | 2095589603.06 | 1.921 | |
| Subarea* Depth | 5261344220.151 | 12 | 438445351.679 | .402 | |
| Year*Subarea* Depth | 9524053701.678 | 12 | 793671141.806 | .728 | |
| Error | 294522132219.864 | 270 | 1090822711.92 | | |

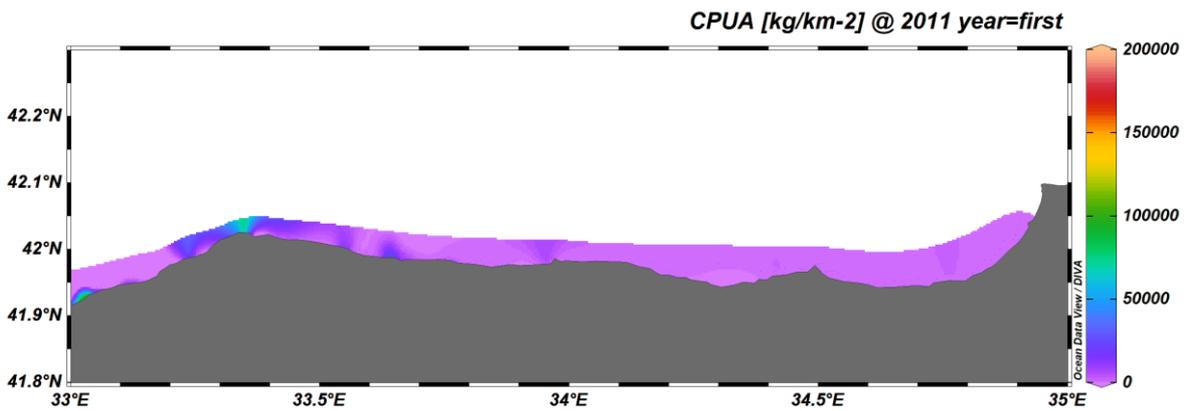


Figure 3. Distribution of biomass CPUA (kg km^{-2}) *A. kagoshimensis* in 2011.

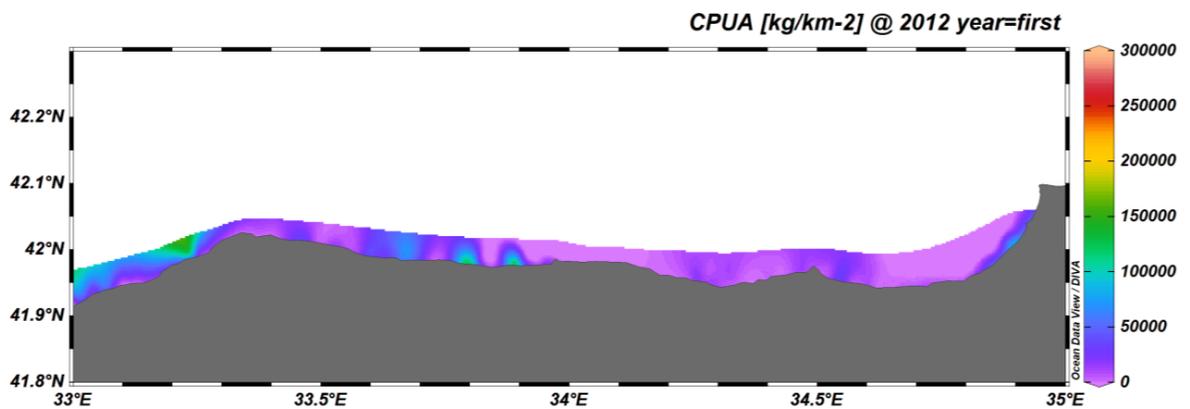


Figure 4. Distribution of biomass *A. kagoshimensis* CPUA (kg km^{-2}) in 2012.

The length of sampled *A. kagoshimensis* specimens ranged from 10 mm to 60 mm. The average length of *A. kagoshimensis* in 2011 was 34.44 ± 0.38 mm and the average weight was 16.76 ± 0.4 g. In 2012, the average length was 34.35 ± 0.33 mm and the average weight was 15.18 ± 0.38 g (Table 4).

The length–weight relationship of *A. kagoshimensis* was found to be $TW = 0.0003SL^{2.9642}$ in 2011 and $TW = 0.0003SL^{3.0171}$ in 2012 (Figure 6). *A. kagoshimensis* were found to have negative allometric growth curve slope ($b \neq 3$), ($p < 0.05$).

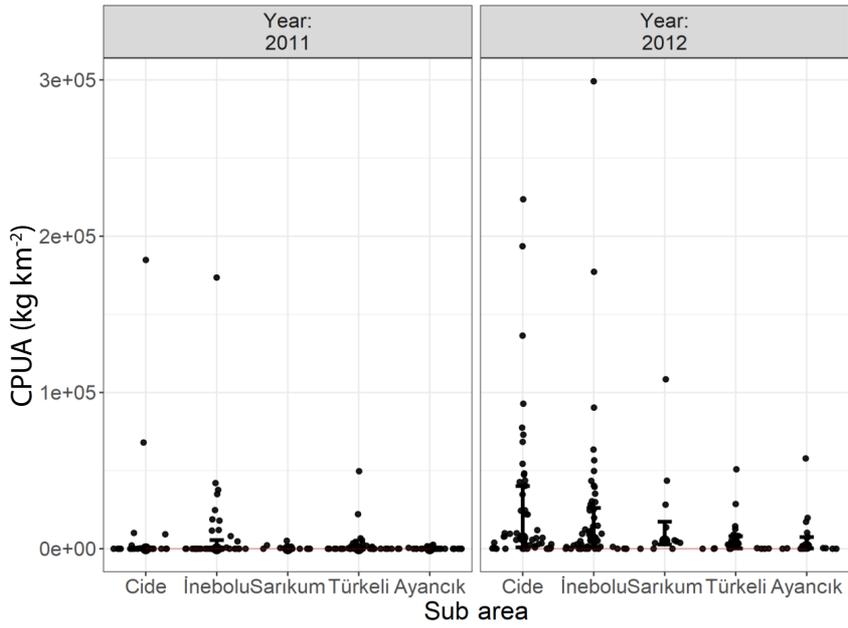


Figure 5. Change of per haul biomass by subarea and year.

Table 4. Length–weight distribution of *A. kagoshimensis*.

| Year | Parameters | n | Min–max | Mean (SE) |
|------|-------------------|------|-------------|--------------|
| 2011 | Shell length (mm) | 1130 | 10.67–60 | 34.44(0.38) |
| | Shell height (mm) | 1130 | 8.8–48.5 | 27.1(0.29) |
| | Shell width (mm) | 1130 | 5.94–48.03 | 23.01(0.28) |
| | Total weight (g) | 1130 | 0.33–57.07 | 16.76(0.4) |
| 2012 | Shell length (mm) | 783 | 11.09–60.39 | 34.35(0.33) |
| | Shell height (mm) | 783 | 8.07–45.3 | 27.06(0.27) |
| | Shell width (mm) | 783 | 5.48–40.71 | 23.2(0.26) |
| | Total weight (g) | 783 | 0.38–57.5 | 15.18(0.38) |

SE: Standart error, n: number

Table 5. The LWR reported in the literature for the same genus species.

| Species | Region | a | b | CI _b | R ² | References |
|--------------------------|-------------------|--------|------|-----------------|----------------|---------------------|
| <i>A. granosa</i> | India | 0.2450 | 3.29 | | | Broom, 1985 |
| <i>A. granosa</i> | Malaysia | 0.0013 | 2.26 | | | Narasimham, 1988 |
| <i>A. cornea</i> | East Black Sea | 0.0033 | 2.58 | | | Şahin, 1995 |
| <i>A. cornea</i> | East Black Sea | 0.0015 | 2.61 | | | Şahin, 1999 |
| <i>A. inaequilvalvis</i> | East Black Sea | 0.0002 | 3.05 | | | Şahin, 2009 |
| <i>A. inaequilvalvis</i> | Aegean Sea | 0.197 | 3.09 | | | Acarlı et al., 2012 |
| <i>A. inaequilvalvis</i> | Eastern Black Sea | 0.2452 | 3.15 | | 0.98 | Aydın et al., 2014 |
| <i>A. kagoshimensis</i> | Western Black Sea | 0.0003 | 2.96 | 2.93–2.98 | 0.96 | This study |
| <i>A. kagoshimensis</i> | Western Black Sea | 0.0003 | 3.01 | 2.97–3.06 | 0.98 | This study |

CI: Confidence interval

The height–weight and width–weight relationships were $TW = 0.0006SH^{2.9708}$, $TW = 0.0046SW^{2.5147}$, respectively, in 2011; In 2012, $TW = 0.0009SH^{2.8814}$, $TW = 0.0035SW^{2.5985}$.

3.3. Growth parameters

L_{∞} of seasonal and nonseasonal VBGP was 81.89 mm and 84.32 mm, $K = 0.32$ and 0.31 (year^{-1}) and $t_0 = -0.19$ and -0.21 (year), respectively. The growth parameter estimates are shown in Figure 7, and the LFDA growth curves are in Figures 8 and 9.

3.4. Genetic characterization

Total DNA was successfully isolated from 6 individuals, but *cox-1* gene region was successfully amplified only for three specimens. Reliable, high-quality sequences were deposited in Genbank under accession numbers OK091154 - OK091156. Blast and BOLD search showed COI gene partial sequences from specimens were similar to *A. kagoshimensis* sequences in Genbank with more than 99% identity (Figure 10).

A total of 7 sequences were retrieved from NCBI nucleotide database and a phylogenetic tree was

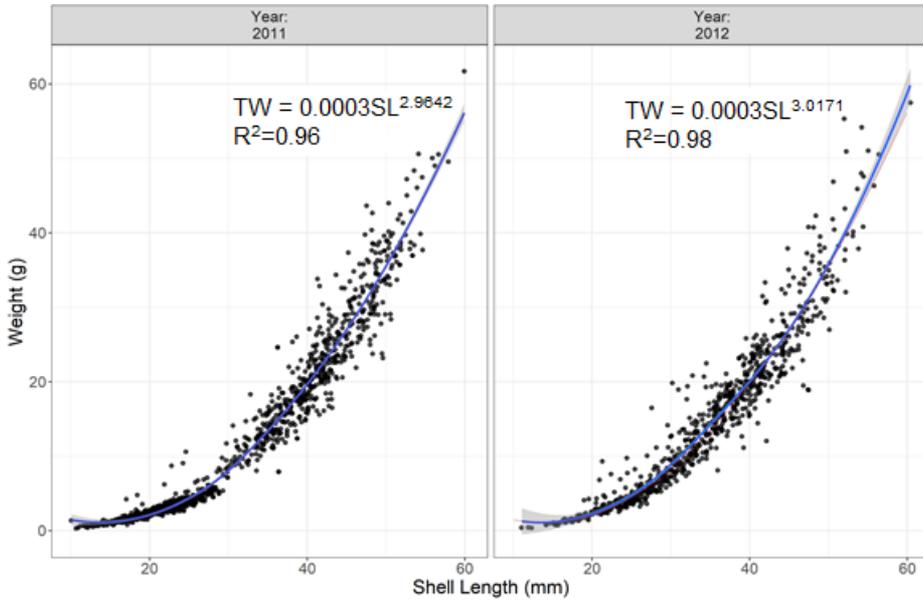


Figure 6. Shell length–weight relationship of *A. kagoshimensis* by years.

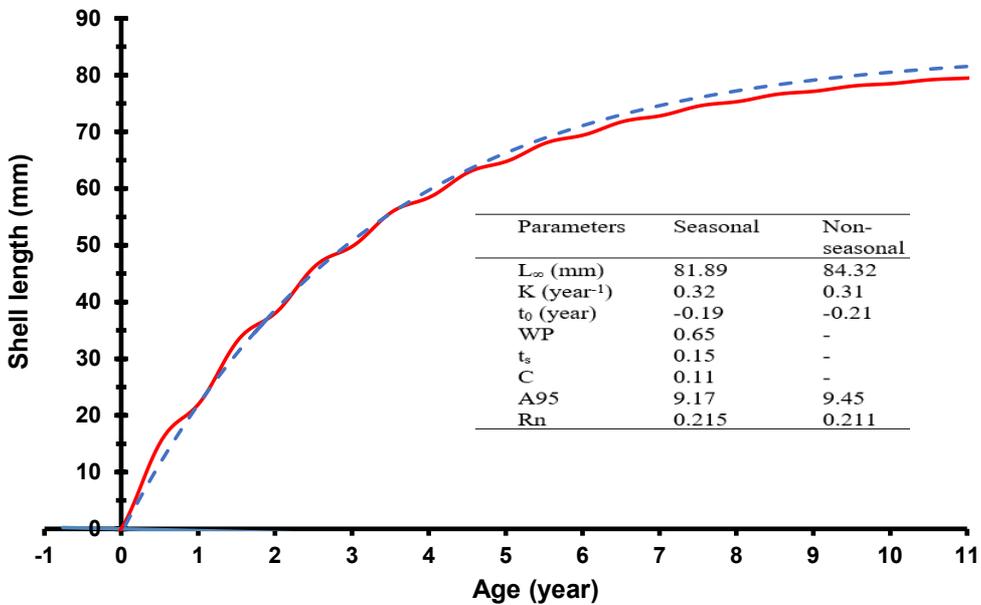


Figure 7. Seasonal (red line) and non-seasonal (blue line) von Bertalanffy growth curves of *A. kagoshimensis*

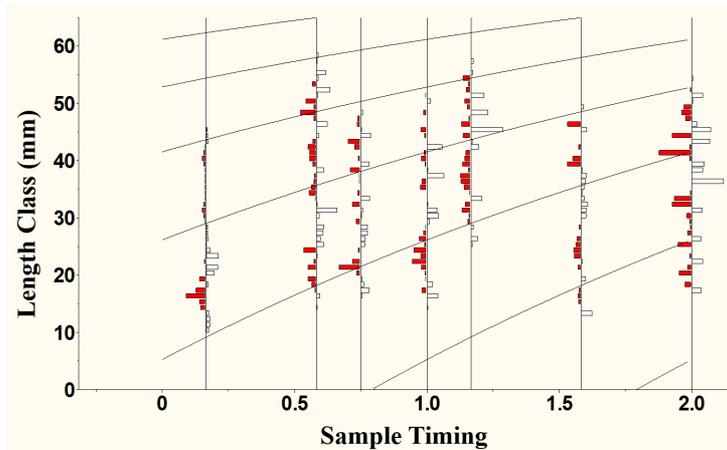


Figure 8. Seasonal VBGP obtained from LFDA (shell length 2 mm size classes frequency distributions; February, May, July, December 2011, February, July, December 2012).

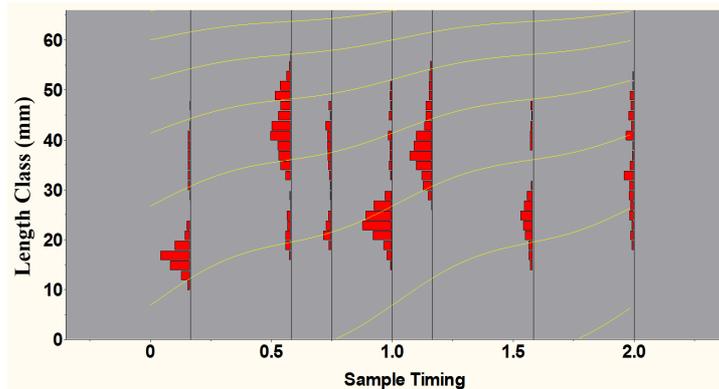


Figure 9. Nonseasonal VBGP obtain from LFDA (shell length 2 mm size classes frequency distributions; February, May, July, December 2011, February, July, December 2012)

constructed with sequences of specimens. The phylogenetic tree showed sequences of specimens made in a group with *A. kagoshimensis* sequences from Genbank with 100% bootstrap value. This group formed a clade with *A. kagoshimensis* and finally *A. avellana* bound to this clade as an outgroup.

4. Discussion

The findings reported biomass and VBGP in this study represent the first data for *A. kagoshimensis* from the South Western Black Sea basin. Studies on the abundance of *A. kagoshimensis* in the Black Sea are limited. Marinov (1990) reported *A. inaequilvalvis* 100 ind/m², and the biomass, over 1 kg/m² on the Bulgaria coasts. In the study conducted on the Romanian coast, it is stated that the biomass value of *A. inaequilvalvis* is between 0.3 and 218.8 g/m². In the same study, biomass was found as 11.6 g/m², 28.6 g/m², and 34.8 g/m² in the evaluation made by

dividing into 3 layers (Abaza et al., 2010). North–Eastern Black Sea has noted *A. kagoshimensis* (2–32 ind/m²) with low biomass values (0.4–51 g /m²) in 2012 (Kolyuchkina et al., 2018). Another study found 0.2 specimens/m² in Kazacha Bay in the Black Sea (Bondarev, 2020). Another study in the same area about baby clam commercial fishing estimated blood cockle by-catch as kg/min. It was given as 0.0065 kg/min in summer, 0.419 kg/minute in winter, and 0.895 kg/min in spring. As can be seen from these results, the amount of mussel beds is higher than the other bivalvia species. Baby clam is an exception to this bivalvia species because it is a dominant and commercial species in the Black Sea. On the other hand, it is stated that the amount of *R. venosa* stocks as predators sharing the same habitat with *A. kagoshimensis* is 1062 t and the amount of *C. gallina* stocks in food competition is 62,000 t (Dağtekin et al., 2016). Thus, the increase in stocks of this species in 1 year shows that the species is not under such pressure.

A. kagoshimensis populations are capable of reaching very high densities. Certainly, as it is known, recruitment involves a very complex and unknown process in itself. Recruitment is related to climate and weather conditions, immediate changes immediately after the reproduction period in the hydrodynamics of the region, reproductive potential, suitability of the substrate, natural predations on adult and juvenile individuals, and competition with other species in the food chain for bivalvia (Osman and Whitlatch, 1995; Eckman, 1996; Apte et al., 2010).

In the study conducted in the South Eastern Black Sea, the average length of blood clam was reported as 39.28 ± 0.261 mm, and the average weight was 24.84 ± 0.372 g. The min–max length in the South Eastern Black Sea was measured as 5–85 mm (Şahin, 1999). Another study by Şahin et al. (1999) found the min–max length of 20–69 mm. On the Romania coasts, another blood cockle species of *Anadara tuberculosa*, mean weight was found to be 43.48 mm (Stern-pirlot and Wolff, 2006). Şahin et al. (2009) reported 39.28 ± 0.261 mm and 24.84 ± 0.37 g. Aydın et al. (2014) reported min–max length of 4.5–71.8 mm and an average length of 37.7 ± 0.02 mm. Bodarev (2020) reported of *A. kagoshimensis* varied between 11.8 and 45.8 mm at a mean shell length of 26.2 mm. In the present study was found an average length of 34.44 ± 0.38 mm and 34.35 ± 0.33 mm average weight of 16.76 ± 0.4 g and 15.18 ± 0.38 g in 2011 and 2012, respectively.

The literature reported *b* values of the species belonging to the *Anadara* genus in the range of 2.26–3.29 (Table 6). The results of this study were found to fall within these ranges. Şahin (1999) found $TW = 0.002 L^{3.058}$ in the South Eastern Black Sea. Acarli et al. (2012) estimated $TW = 0.197 L^{3.095}$ in the Aegean Sea. Aydın et al. (2012) reported $TW = 0.245 L^{3.158}$ in the South Eastern Black Sea. The growth performance index is different from the values found in studies on the family Arcidae (Table 6). There is a similarity with Şahin al.'s study (1999) carried out in the Eastern Black Sea. Generally, bivalvia species have irregular recruitment of juveniles. In the ecosystem, there are large annual spatial variations due to environmental factors occasionally known often not known. These have irregular growth rates (Fritz et al., 2022). They could show large variations from place to place in growth due to food availability for suspension feeders. They can have irregular distribution patterns in the horizontal plane. Moreover, large variations were seen from place to place in abundance occasionally due to sediment type, depth preference, or extremes in water temperatures. Particularly, bivalvia species can be irregular in vertical planes (Jacobs et al., 2014). It is understood in our study results that there are changes due to similar factors. *A. kagoshimensis* can withstand the low summer oxygen content in bottom layers easier than the native, autochthonous species of mussels. This has resulted in an increase of its biomass and replacement of the dominant native species *C. gallina*.

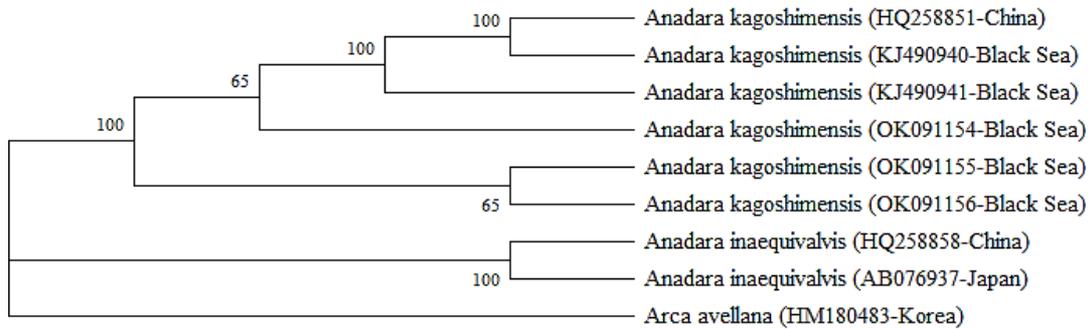


Figure 10. UPGMA phylogenetic tree based on COI gene sequences of two *Anadara* species and *Arca avellana* retrieved from Genbank and also three new sequences obtained from this study (OK091154-OK091156). Numbers by the nodes show bootstrap support probabilities.

Table 6. VBGP and mortality rates of *Anadara* genus in the different areas.

| Species | Region | L_{∞} | K | t_0 | A95 | Z | M | ϕ | References |
|-------------------------|----------------------------|--------------|------|-------|------|------|------|--------|--------------------------|
| <i>A. rhomsa</i> | India | 90.2 | 0.46 | | | | | 3.57 | Narasimhan, 1988 |
| <i>A. cornea</i> | Turkey (East Black Sea) | 75.2 | 0.37 | -0.10 | | | | 3.32 | Şahin et al., 1999 |
| <i>A. inaequivalvis</i> | Turkey (East Black Sea) | 89.38 | 0.23 | -0.45 | | | | 3.26 | Şahin et al., 2009 |
| <i>A. kagoshimensis</i> | Turkey (Western Black Sea) | 81.89 | 0.32 | -0.19 | 9.17 | 1.35 | 1.29 | 3.33 | This study (seasonal) |
| <i>A. kagoshimensis</i> | Turkey (Western Black Sea) | 84.32 | 0.31 | -0.21 | 9.45 | 1.38 | 1.29 | 3.34 | This study (nonseasonal) |

In addition to morphological characterisation, molecular identification confirmed that the ark clam species invading Black Sea is *A. kagoshimensis*. However, only three specimens' sequence were recovered, which might be related to mucopolysaccharide levels which often reduce PCR efficiency (Layton et al., 2014). More research is needed to enhance DNA extraction and/or PCR amplification protocols for marine molluscs. Phylogenetic tree based on these sequences highlighted the relationship of specimens with *A. kagoshimensis* and a clear separation of them from other ark clam species. The results of this study comply with the study of Krapal et al. (2014) and Bañón et al. (2015) in terms of species identification and the suitability of cox - I barcodes as molecular taxonomy markers.

5. Conclusion

Alien important ecological relationships between *A. kagoshimensis* and other species such as *C. gallina* and *R. venosa*. It is a threat to the species with which it competes, especially *C. gallina*, which is a commercial species because there is no commercial fishing of this species. Although there is an advantage in the prey position of *R. venosa*, it would be useful to consider this species as a commercial species. In some of the studies conducted in the Black Sea, genetic analyses were not performed. Therefore, it is not clear whether there are problems in species identification in these studies. It is important

to make the species identification genetically as well as morphological features. Otherwise, problems may arise in the type definition. Species are being regarded as a different Descriptor (i.e., D2) of GES in order to attain Good Environmental Status (GES) for marine ecosystems throughout Europe by 2020 (Băncilă et al., 2022). There are indications that invasive species will gradually increase. *A. kagoshimensis* is included in the 100 alien invasive species for the Mediterranean Sea and Black Sea. *A. kagoshimensis* increase Indo-West Pacific, Mediterranean, and the Black Sea. There is a need for measures to reduce the effect in this direction. The exotic species *A. kagoshimensis* needs further investigation and management plan into the Black Sea. *Anadara kagoshimensis* is one of the bivalvia species that is the least affected by bad environmental conditions compared to other species (Soldatov et al., 2018). This makes it strong in the ecosystem.

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Conflict of interest

The authors declare that they have no known competing financial or nonfinancial, professional, or personal conflicts that could have appeared to influence the work reported in this paper.

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