Original Article

Combined effects of salinity and temperature on survival and growth during the early life cycle of the rock oyster *Saccostrea cucullata* (Born, 1778)

José A. Marcelino[1]*, Adriano Macia[2], Mizeque J. Mafambissa[2], Diego Castejón[3,4], Carlos Andrade[5]

[1] Higher School of Rural Development, Eduardo Mondlane University, 5º Congress Area, Vilanculo City, Inhambane, Mozambique.

[2] Department of Biological Sciences, Eduardo Mondlane University, PO Box 257, Maputo 1100, Mozambique

[3] Mariculture Centre of Calheta, Av. D. Manuel I, nº7, 9370-135 Calheta, Madeira, Portugal

[4] CIIMAR – Interdisciplinary Centre of Marine and Environmental Research - University of Porto, 4450-208 Matosinhos, Portugal.

[5] MARE - Marine and Environmental Sciences Centre/ARNET—Aquatic Research Network, Agência Regional para o Desenvolvimento da Investigação Tecnologia e Inovação (ARDITI), Caminho da Penteada, Edifício Madeira Tecnopolo, 9020-105 Funchal, Portugal

Western Indian Ocean JOURNAL OF Marine Science

Open access

Citation:
Marcelino JA, Macia A, Mafambissa MJ, Castejón D, Andrade C (2023) Combined effects of salinity and temperature on survival and growth during the early life cycle of the rock oyster Saccostrea cucullata (Born, 1778). Western Indian Ocean Journal of Marine Science 22(1): 95-102 [doi: 10.4314/wiojms.v22i1.10]

Received:
December 11, 2022

Accepted:
May 23, 2023

Published:
July 25, 2023

*** Corresponding author:**
jartur@uem.mz

Abstract

Temperature and salinity are among the critical factors affecting the survival and growth of bivalve larvae. The combined effects of temperature and salinity on the embryonic and larval development of the rock oyster *Saccostrea cucullata* (von Born, 1778) in culture conditions were investigated in a laboratory study on Inhaca Island, Mozambique. A factorial experimental design tested three temperatures (24, 30 and 34 °C) and three salinities (30, 35 and 40 parts per thousand) over a seven-day period. Larval survival and growth (in height and length) were assessed by regular sampling by counting and measurement of larvae under an optical microscope equipped with a micrometric scale. Significantly higher larval survival was observed at the combination of 30 °C and 35 salinity. However, the mid-range temperature (30 °C) and highest salinity (40) resulted in faster growth of the larvae. The lowest temperature (24 °C) negatively affected growth regardless of salinity level and survival decreased linearly with increasing salinities. The present results will aid in the understanding of the environmental factors behind the natural recruitment of spat of *S. cucullata* and contribute to the optimization of rearing protocols for the larval culture of this oyster species.

Keywords: temperature, salinity, rock oyster, larvae, growth, survival

Introduction

The rock oyster *Saccostrea cucullata* (Born, 1778) is a widely distributed species in the Indo-Pacific and Eastern Atlantic Oceans and has been introduced into the Eastern Mediterranean Sea (Çevik *et al*., 2001). *S. cucullata* is an intertidal species mainly found in brackish water environments between 0 and 5 m depth, where it is usually attached to rocks, roots and trunks of mangrove trees and on pier piles (Dye *et al*., 1994; Carpenter *et al*., 1997; Poutiers, 1998). The rock oysters are a popular food consumed throughout its distribution area, but the overexploitation of their natural beds is reducing the catches in different countries, including South Africa (Dye, 1989), Kenya (Tack,

1999), Mozambique (Everett, 2008) and India (Tenjing, 2020). In Bangladesh, the decline of the rock oyster beds is leading to the collapse of breakwater reefs (Chowdhury, 2019). It is widely acknowledged that shellfish reefs provide relevant ecosystem services such as coastal protection, nursery grounds to enhance fisheries production, contributing to blue carbon capture, and pollutant removal and detoxification (Nagelkerken *et al*., 2015; Waltham *et al*., 2020).

S. cucculata is also an interesting species for aquaculture purposes due to its high commercial value, tolerance to extreme environmental conditions and fast growth. Most recent research and development efforts on *S. cuccullata* have focused on improving rock oyster natural populations through the development of improved substrates for spat collection and juvenile growth (Racuyal *et al*., 2016; Chowdhury, 2019).

However, the culture requirements for *S. cuccullata* during their early life stages are poorly known, which seems to be a major bottleneck for the development of the culture of this species. Major advances thus far on the larviculture of *S. cuccullata* were the production of embryos (Awati and Rai, 1931), and the description of the embryological development and suitable temperature and salinity conditions for the production of straight-hinge veliger larvae (Kalyanasundaram and Ramamoorthi, 1986). Sukumar and Joseph (1988a, 1988b) provided a general insight on the natural breeding cycle of *S. cuccullata*, including gonadal maturation and the description of the factors influencing the spawning cycle. Recently Thanormjit *et al*. (2020) characterized the gametes and early development of *S. cuccullata*.

Temperature and salinity are major environmental factors that affect the survival and growth of bivalve molluscs during early life stages (Robert *et al*., 1988). Considering the natural occurrence of *S. cucculata* in the intertidal zone, where environmental parameters are likely to reach extreme values, the current research tested a range of water salinity and temperature combinations to evaluate the most suitable conditions for larval culture of the rock oyster.

Materials and methods

Breeding and experimental animals

Adult specimens of *S. cuccullata* were collected manually with a concave iron tool during low spring tide on the rocky shores of Ponta Torres (32° 57 ' S, 26° 4' E) on the island of Inhaca (Maputo, Mozambique). Only specimens larger than 20 mm were selected (Nascimento and Pereira, 1980; Dang *et al*., 2010). During collection, water temperature (26.5 ± 1.0 °C) and salinity (35.5) were measured. The oysters were placed in a waterless plastic bucket and capped with Rachel mesh to minimize sunlight stress and brought to the Inhaca Marine Biology Station (EBMI). At EBMI, the oyster shells were cleaned using a nylon brush and a knife, washed with distilled water and transferred to a 50 L aquarium with natural seawater at constant temperature (25 ± 2 °C) and salinity (40). Aeration was continuous and illumination was based on natural light and photoperiod.

Ten oysters were randomly selected and their valves were opened through the sectioning of the adductor muscle (Legat *et al*., 2017; Santos *et al*., 2020; Thanormjit *et al*., 2020). The oysters were then washed with filtered (1 µm) and UV-treated seawater to remove microorganisms lodged in the inner layer of the valves. The sex of oysters was identified by gonad biopsy using an optical microscope (Olympus model CK40; magnification of 40 x).

The gonadal tissue was cut with a scalpel and the gametes were collected using a Pasteur pipette. They were then transferred to a 100 mL beaker (male gametes) and a 1 L container (female gametes) filled with filtered (1 µm) and UV-treated seawater (Absher *et al*., 2000). The viability of the gametes was examined under the microscope, i.e., the sperm cells were mobile and the shape of the oocytes became spherical 20 min after being exposed to water. In vitro fertilization was performed adding 2.5 mL of the sperm solution in 1 L of oocyte suspension. Through microscopic observations, the necessary adjustments were made to establish a ratio of about 3 to 5 spermatozoa for each female gamete to avoid polyspermy. Of the 10 sacrificed adult oysters, only four females and two males were used to obtain the required number of gametes for use in the experiment. Each female contained on average 5 to 10 million eggs and males about 2 million sperm. For fertilization, gametes (spermatozoa and oocytes) were evaluated through microscopic examination according to Helm (2004): mature gametes were considered all ovules normally pear-shaped when removed for the first time and round-shaped in contact with sea water within 20 minutes; the sperm was considered mature when it was motile. Embryos of approximately 40-45 µm were transferred to a 10 L tank filled with 5 L of filtered (1 µm) and UV-treated seawater with moderate aeration at the density of 204 embryos

mL^{-1}, estimated using an optical microscope by counting the embryos present in 1 ml. The temperature was set at 28 °C with the use of thermostats (Dophin 50W). The embryos remained under these conditions until the appearance of D-larvae 24h after fertilization. Since the time required to reach the D-larvae stage was previously unknown, the stages of embryonic and larval development were observed every five minutes.

Experimental design

Twenty-seven culture tanks each containing 5 L of filtered (1 µm) and UV-treated seawater were prepared about 24 hours after fertilization. Larvae were reared in the combinations of three temperatures (24, 30 and 34°C) and three salinities (30, 35 and 40), each with three replicates. The D-larvae were directly transferred to those experimental treatments in three replicates per combined temperature x salinity. Approximately 25,000 D-larvae were transferred to each culture tank at an initial density of 5 larvae mL^{-1}. Water volume in each tank was maintained at 5 L. The salinities of 35 and 30 were obtained by diluting seawater (salinity 40) with distilled water. Moderate aeration was provided using air stones. Every 48 h, tank bottoms were siphoned (50 % water volume exchange) to ensure good water quality. To avoid the elimination of live larvae as well as to select the largest diameter larvae, 60 and 40 µm overlapping meshes were used (smaller mesh over the larger one). A density of 400 cells mL^{-1} of the microalgae *Isochrysis galbana* was provided daily to the D-larvae. A reduction of 50 % on the number of microalgae was provided in the days when no water was exchanged. Microalgal density was estimated daily with a Neubauer chamber. Growth was measured daily by sampling 30 larvae according to Hillerbrand *et al*. (1999). An optical microscope equipped with an ocular micrometric scale (Olympus CK40) was used to measure height (maximum distance between the dorsal and ventral regions) and length (maximum distance between the anterior and the posterior regions) (Hu *et al*., 1993). The mean (± SD) initial length and height of the larvae were 50.28 ± 0.30 µm and 51.30 ± 0.25 µm, respectively. Survival was estimated as the number of larvae alive at the end of the experiment in relation to the initial number. Transparent larvae, which presumably indicated the absence of tissues, were considered dead (Ponis *et al*., 2003). The trial lasted seven days. The data obtained on length, height and survival of oyster larvae were initially tested for normality and homogeneity of variances by the Kolmogorov-Smirnov (distance) test and the Spearman's test, respectively. The data presented a normal distribution and homogeneous variances ($p<0.05$), therefore they were later submitted to parametric tests. A two-way analysis of variance (ANOVA) was used to verify the influence of temperature and salinity on larval growth and survival. If there were significant differences, the Tuckey test was applied for detailed comparison of the differences (Zar, 2010). A significance level of 5 % ($\alpha=0.05$) was adopted. Statistical treatment was carried out with the aid of the GraphPad Prism V. 8.01.

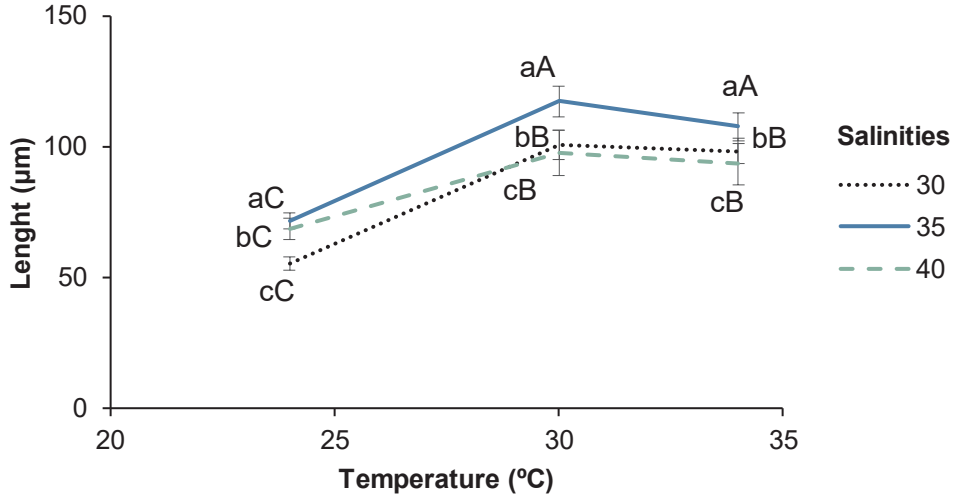

Figure 1. Means (± standard deviation) of length of larvae of the rock oyster, *Saccostrea cuccullata*, reared at combinations of temperature (24, 30 and 34 °C) and salinity (30, 35 and 40) for seven days. Lowercase letters represent comparison between salinities for the same temperature and capital letters represent comparison between different temperatures for the same salinity ($p < 0.05$).

Results

Five minutes after the sperm solution was added to the oocyte suspension, sperm cells were observed surrounding the oocytes. The polar bodies were visible after 20 minutes. The different stages of embryonic development (2, 4, 8, 16 cells, the morula and blastula stages) were all observed within 2 h after fertilization. Trocophora larvae were observed 16 h after fertilization and were characterized by the presence of cilia and circular movements. The D-veliger stage appeared 24 h after fertilization and was characterized by the formation of the D-shaped larval shell.

At the end of the 7 day-long trial, means (± SD) of length (Fig. 1) and height (Fig. 2) of larvae after exposure to the different combinations of salinity and temperature were significantly different with ANOVA $F_{(4,18)}$ = 66.47, p <0.0001 for length and $F_{(4,18)}$ = 380.5, p < 0.0001 for height. The largest larvae (117.30 ± 0.36 µm length and 125.08 ± 0.38 µm height) were observed at 30 °C x 35, while the smaller ones (53.30 ± 0.86 µm length and 54.69 ± 0.40 µm height) were those from treatment 24 °C x 30. The differences in survival rates among treatments were also significant (Fig. 3) with ANOVA $F_{(4,18)}$ = 1309, p <0.0001. The mean (± SD) survival ranged from 26.4 ± 0.4 % in treatment 24 °C x 30 to 35 salinity to 73.2 ± 0.2% in treatment 34 °C x 35 salinity. The highest survival was observed at 34 °C x 35 salinity. All treatments at 24 °C resulted in significantly lower survival rates.

Discussion

The present study suggests that *S. cuccullata* larvae can grow well at 30 °C temperature x 35 salinity to obtain largest larvae, however to obtain higher survival of larvae, they must be grown at 34 °C temperature x 35 salinity. The temperature of 30 °C is well above the average annual temperature of 25 ± 1 °C for the brood stock collection area at Inhaca Island, where (according to Kalk, 1995) the highest average is 27.5 °C and the lowest 21.7 °C. The mean salinity levels at Inhaca are 33 ± 2 (de Boer *et al*., 2000) with a range from 32 to 42 (Pinto, 1996). This confirms the findings of Kalyanasundaram and Ramamoorthi (1986) that *S. cuccullata* larvae tolerate a wide temperature and salinity variation though with different growth and survival trends.

Temperature and salinity are among the main environmental factors affecting the growth (Robert *et al*., 1988; His *et al*., 1989; Manoj Nair and Appukuttan, 2003) and survival of bivalve larvae (Yuan *et al*., 2016; Verween *et al*., 2007; Manoj Nair and Appukuttan, 2003; Robert *et al*., 1988). In this study, embryos and larvae reached all developmental stages defined by Kalyanasundaram and Ramamoorthi (1986). However, due to the distinct culture conditions of the treatments, the life stages were reached at different times.

The highest larval growth was observed at salinity 35, coinciding partly with the results obtained by Kalyanasundaram and Ramamoorthi (1986) which

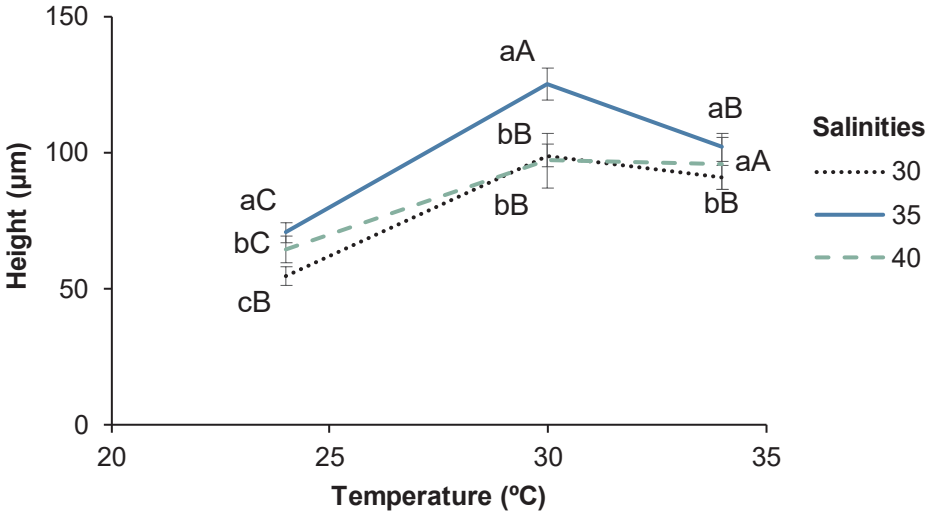

Figure 2. Means (± standard deviation) of height of larvae of the rock oyster, *Saccostrea cuccullata*, reared at combinations of temperature (24, 30 and 34 °C) and salinity (30, 35 and 40) for seven days. Lowercase letters represent comparison between salinities for the same temperature and capital letters represent comparison between different temperatures for the same salinity ($p < 0.05$).

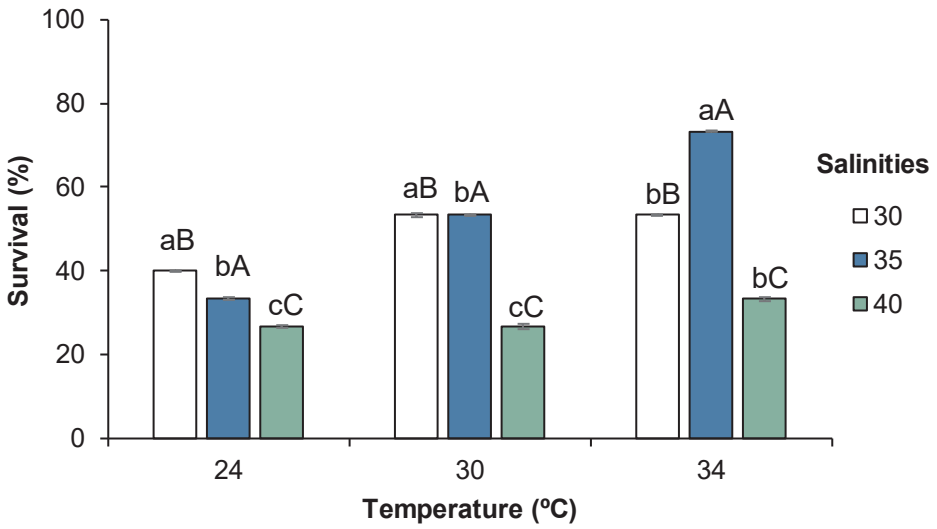

Figure 3. Percentage of survival of rock oyster, *Saccostrea cuccullata* larvae reared at combinations of temperature (24, 30 and 34 °C) and salinity (30, 35 and 40) after seven days. Lowercase letters represent comparison between salinities for the same temperature and capital letters represent comparison between different temperatures for the same salinity ($p < 0.05$).

reported good larval development of *S. cuccullata* at salinities from 20 to 35. In contrast, Sudrajat (1990) determined an optimum salinity of 25 for embryonic development of *S. cuccullata*. In general, oyster larvae tolerate a wide salinity range between 25 and 40 (Lemos *et al*., 1994). However, this study showed that salinity affects the growth of *S. cuccullata* larvae, being the lowest at salinity 30. It may be possible that osmotic disturbances lead to energy allocations that ultimately impair other functions such as growth (Deaton, 2008). Temperature is regarded as the most important environmental factor influencing bivalve culture (Robert *et al*., 1988; Helm *et al*., 2004). Temperature increments accelerate the growth of oyster larvae (Doround *et al*., 1999). The highest growth recorded here at 30 °C supports previous observations by Kalyanasundaram and Ramamoorthi (1986) that larval growth of *S. cuccullata* is higher at 30 °C. Yukihira *et al*. (2000) reported that the influence of temperature on the metabolism and physiological processes of oysters creates an optimal temperature range for each species providing a maximum rate of growth as well as survival. In fact, a similar trend was observed in the present study, where the maximum temperature tested (34 °C) resulted in reduced larval growth.

The greatest survival (73 %) obtained at the combination of salinity 35 x 34 °C, was higher than the one from the microalgae feeding experiments by Martínez-Fernández and Southgate (2007) in *Pinctada margaritifera* with 70.5 %. It was also greater than the 50 % larval survival obtained by Mafambissa (2009) for *Crassostrea rhizophorae*. Other oyster species such as *Crassostrea gigas* have shown higher larval survival rates of up to 87 % (Ponis *et al*., 2003).

The results from this study are in partial agreement with Nell and Holliday (1988) who reported higher larval survival of *S. commercialis* in salinities ranging between 23 and 39. However, the current results differ from those obtained by Coeroli *et al*. (1984) where they reported higher survival of *Saccostrea echinata* larvae at salinities ranging from 25 to 30 and temperatures from 25 to 29 °C. Heral and Deslous-Paoli (1990) demonstrated that *C. gigas* larvae tolerate higher salinities (from 45 to 50) with significant mortality above 50. These differences reflect the genetic variability among different species of oysters and their high adaptability to different environmental conditions. The lowest larvae survival in the present study (26.4 %) was achieved at salinity 40, suggesting this is a threshold salinity level for *S. cucculata* larvae.

All treatments at 24 °C resulted in significantly lower survival rates. This seems to be the lower temperature limit tolerated by the larval stages of this species. Another factor to consider is the abrupt submission of the larvae to low temperatures without any previous acclimation, which may have caused a thermal shock and contributed to a decrease in survival rates. Opposite to what was previously reported by Kent *et al*. (1999), the fertilization method used here was very effective as high survival of larvae was observed in treatments where appropriate environmental

conditions were provided. Together with the optimal water salinity and temperature for larval rearing established here, this may contribute to the further development of the culture of *S. cuccullata*. These findings related to the environmental variable thresholds for *S. cuccullata* larvae are relevant for future studies on recruitment, as well as to evaluate the direct impact of global warming on this species.

Acknowledgements

The authors would like to express their appreciation to Mr Estação Nhaca for the field assistance. This study was funded by Ocean Revolution Mozambique and Fundo Nacional de Investigação (FNI). The cultivation of microalgae was done at the National Fisheries Research Institute. The experiment of larvae cultivation was done in the laboratory of the Inhaca Marine Biology Station and larval measurements were made at the Department of Biology, Eduardo Mondlane University.

References

Absher TM, Vergara EM, Christo SW (2000) Growth and allometry of the Brazilian oyster *Crassostrea brasiliana* (Lamarck, 1819) (Bivalvia: Ostreidae). O*phelia* 53: 105-112 [doi:https://org/10.1080/00785236.2000.10409440]

Awati PR, Rai HS (1931) *Ostrea cucullata* (the Bombay oyster). Indian Zoological Memoir 3: 1-107

Carpenter KE, Krupp F, Jones DA, Zajonz U (1997) Species identification guide for fishery purposes. The living marine resources of Kuwait, Eastern Saudi Arabia, Bahrain, Qatar, and the United Arab Emirates. FAO, Rome. 324 pp

Çevik C, Öztürk B, Buzzuro G (2001) The presence of *Crassostrea virginica* (Gmelin, 1791) and *Saccostrea commercialis* (Iredale and Roughley, 1933) in the Eastern Mediterranean Sea. La Conchiglia 298: 25-28

Coeroli M, de Gailland D, Landret JP, Coatanea D (1984) Recent innovations in cultivation of molluscs in French Polynesia. Aquaculture 39: 45-67

Chowdhury MSN (2019) Ecological engineering with oysters for coastal resilience: Habitat suitability, bioenergetics, and ecosystem services. PhD Thesis, Wageningen University. Wageningen. 198 pp [doi: https://doi.org/10.18174/466205]

Dang C, Xavier de M, Mériame G, Christian P, Noelle B, Nathalie C (2010) The Manila clam population in Arcachon Bay (SW France): Can it be kept sustainable? Journal of Sea Research 63 (2): 108-118 [doi:10.1016/j.seares.2009.11.003]

De Boer WF, Rydberg L, Saide V (2000) Tides, tidal currents and their effects on the intertidal ecosystem of the Southern Bay, Inhaca Island, Mozambique. Hydrobiologia 428: 187-196 [doi: 10.1023/A:1004030605474]

Deaton L (2008) Osmotic and ionic regulation in molluscs. In: Evans DH (ed) Osmotic and ionic regulation: cells and animals. CRC Press, London. pp 107-133 [doi: https://doi.org/10.1201/9780849380525]

Doroudi MS, Southgate PC, Mayer J (1999) The combined effects of salinity and temperature on embryos and larvae of the black-lip pearl oyster, *Pinctada margaritifera* (L.). Aquatic Research 30: 271-277 [doi: 10.1046/j.1365-2109.1999.00324.x.]

Dye AH (1989) Studies on the ecology of *Saccostrea cucullata* (Born, 1778) (Mollusca: Bivalvia) on the east coast of southern Africa. South African Journal of Zoology 24 (2): 110-115 [doi: 10.1080/02541858.1989.11448142]

Dye AH, Schleyer MH, Lambert G, Lasiak TA (1994) Intertidal and subtidal filter-feeders in Southern Africa. Ecological Studies 103: 57-74 [doi: http://dx.doi.org/10.1007/978-3-642-78283-1_4]

Everett BI, van der Elst RP, Schleyer MH (2008) A natural history of the Bazaruto Archipelago, Mozambique. Oceanographic Research Institute, No. 8. 126 pp

Helm MM, Bourne N, Lovatelli A (2004) Hatchery culture of bivalves: A practical manual. FAO Fisheries Technical Paper, Rome. 471 pp

Heral M, Deslous-Paoli JM (1990) Oyster culture in European countries. In: Menzel W (ed) Estuarine and marine bivalve mollusk culture. CRC Press, New York. pp 153-190 [doi: https://archimer.ifremer.fr/doc/00000/3038/]

Hillerbrand H, Durselen CD, Kirschtel D, Pollinger U, Zohary T (1999) Biovolume calculation for pelagic and benthic microalgae. Journal of Phycology 35: 403-424 [doi: https://doi.org/10.1046/j.1529-8817.1999.3520403.x]

His E, Robert R, Dinet A (1989) Combined effects of temperature and salinity on fed and starved larvae of the Mediterranean mussel *Mytilus galloprovincialis* and the Japanese oyster *Crassostrea gigas*. Marine Biology 100 (4): 455-463 [doi: https://doi.org/10.1007/BF00394822]

Hu YP, Fuller CF, Castagna M, Vrijenhoek RC, Lutz RA (1993) Shell morphology and identification of early life history of congeneric species of *Crassostrea* and *Ostrea*. Journal of the Marine Biological Association of the UK 73 (03): 471-496 [doi:10.1017/S0025315400033051]

Kalk M (1995) A natural history of Inhaca Island, Mozambique (3th ed). University Press, Witwatersrand. 395 pp

Kalyanasundaram M, Ramamoorthi K (1986) Temperature and salinity requirements for embryonic development of *Saccostrea cuculata* (Born). Mahasagar-Bulletin of the National Institute of Oceanography 19 (1): 53-55

Kent GN, Maguire GB, Duthie I (1999) Spawning, settlement, and growth of the New Zealand venerid *Ruditapes largillierti* (Philippi, 1849) in culture. New Zealand Journal or Marine and Freshwater Research 33: 55-62 [doi: 10.1080/00288330.1999.9516856]

Legat JFA, Puchnick-Legat A, Gomes CHAM, Suhnel, S, Melo CMR (2017) Effects of salinity on fertilization and larviculture of the mangrove oyster, *Crassostrea gasar* in the laboratory. Aquaculture 468: 545-548 **[doi:** 10.1016/j.aquaculture.2016.11.016]

Lemos MBN, Nascimento IA, de Araújo MMS, Pereira SA, Bahia I, Smith, DH (1994) The combined effects of salinity, temperature, antibiotic and aeration on larval growth and survival of the oyster *Crassostrea rhizophorae*. Journal of Shellfish Research 13: 187-192

Mafambissa MJ (2009) Avaliação dos efeitos de microalgas marinhas nativas sobre o crescimento e sobrevivência de larvas da ostra do mangue *Crassostrea rhizophorae* (Guilding, 1828). Dissertação de Mestrado, Universidade Federal Fluminsense, Brazil. 65 pp

Martínez-Fernández E, Southgate PC (2007) Use of tropical microalgae as food for larvae of the black-lip pearl oyster *Pinctada margaritifera*. Aquaculture 263: 220-226 [doi: 10.1016/j.aquaculture.2006.09.040]

Manoj Nair R, Appukuttan KK (2003) Effect of temperature on the development, growth, survival and settlement of green mussel *Perna viridis* (Linnaeus, 1758). Aquaculture Research 34 (12): 1037–1045 [doi:org/10.1046/j.1365-2109.2003.00906.x]

Nagelkerken I, Sheaves M, Baker R, Connolly RM (2015) The seascape nursery: a novel spatial approach to identify and manage nurseries for coastal marine fauna. Journal of Fish and Fisheries 16: 362-371 [doi:org/10.1111/faf.12057]

Nascimento IA, Pereira SA (1980) Changes in the condition index for mangrove oysters (*Crassostrea rhizophorae*) from Todos os Santos Bay, Brazil. Aquaculture 20: 9-15 [doi:org/10.1016/0044-8486(80)90057-5]

Nell JA, Holliday JE (1988) Effects of salinity on the growth and survival of Sydney rock oyster (*Saccostrea commercialis*) and Pacific oyster (*Crassostrea gigas*) larvae and spat. Aquaculture 68: 39-44 [doi:org/10.1016/0044-8486(88)90289-X]

Pinto I (1996) Comunidades zooplanctônicas das águas da costa ocidental da Ilha da Inhaca. Universidade Eduardo Mondlane. 66 pp

Ponis E, Robert R, Parisi G (2003) Nutritional value of fresh and concentrated algal diets for larval and juvenile Pacific oysters (*Crassostrea gigas*). Aquaculture 221: 491–505 [doi:10.1016/S0044-8486(03)00075-9]

Poutiers JM (1998) Bivalves. Acephala, Lamellibranchia, Pelecypoda. In: Carpenter KE, Niem VH (eds) FAO species identification guide for fishery purposes. The living marine resources of the Western Central Pacific. Volume 1. Seaweeds, corals, bivalves, and gastropods. FAO, Rome. pp 123-362

Racuyal JT, Mabonga DA, Roncesvalles ER (2016) Rock mounds as rock oyster (*Saccostrea cucullata* (von Born, 1778) bed in an intertidal zone. Journal of Academic Research 1 (4): 11-21

Robert R, His E, Dinet A (1988) Combined effects of temperature and salinity on fed starved larvae of the European flat oyster, *Ostrea edulis*. Marine Biology 97: 95-100 [doi: https://doi.org/10.1007/BF00391249]

Sukumar P, Joseph MM (1988a) Annual reproductive cycle of the rock oyster *Saccostrea cucculata* (von Born). Proceedings of the 1st Indian Fisheries Forum. Mangalore, Karnataka: 207-210

Sukumar P, Joseph MM (1988b) Larval development of the rock oyster *Saccostrea cucullata* (von Born). In: M. Mohan Joseph (ed) The First Indian Fisheries Forum, Proceedings. Asian Fisheries Society, Indian Branch, Mangalore: 255-258

Sudrajat A (1990) Studies on the reproductive biology and culture of the rock oyster, *Saccostrea cuculata* (Born) and slipper oyster, *Crassostrea iredalei* (Faustino). PhD Thesis, University College of Swansea. 85 pp

Tack JF (1999) Behavioural aspects of the mangrove oyster *Saccostrea cucullata* (von Born, 1778) explaining its macro and micro distribution along the Kenyan coast. Vrije Universiteit, Brussels. 228 pp

Tenjing SY (2020) Population dynamics of the edible rock oyster *Saccostrea cucullate* (Born, 1778) along the south-west coast of India. Indian Journal of Fisheries 67 (1): 12-22 [doi: 10.21077/ijf.2019.67.1.82239-03]

Thanormjit K, Chueycham S, Phraprasert P, Sukparangsi W, Kingtong S (2020) Gamete characteristics and early development of the hooded oyster *Saccostrea cuccullata* (Born, 1778). Aquaculture Reports 18 (2020): 100473 [doi: org/10.1016/j.aqrep.2020.100473]

Verween A, Vincx M, Degraer S (2007) The effect of temperature and salinity on the survival of *Mytilopsis leucophaeata* larvae (Mollusca, Bivalvia): The search for environmental limits. Journal of Experimental Marine Biology and Ecology 348 (1-2): 111-120 [doi:org/10.1016/j.jembe.2007.04.011]

Waltham NJ, Elliott M, Lee SY, Lovelock C, Duarte CM, Buelow C, Simenstad C, Nagelkerken I, Claassens

L, Wen CKC, Barletta M, Connolly RM, Gillies C, Mitsch WJ, Ogburn MB, Purandare J, Possingham H, Sheaves M (2020) Decade on ecosystem restoration 2021–2030—What chance for success in restoring coastal ecosystems? Frontiers in Marine Science 7: 71 [doi:org/10.3389/fmars.2020.00071]

Yuan WS, Walters LJ, Brodsky SA, Schneider KR, Hoffman EA (2016) Synergistic effects of salinity and temperature on the survival of two nonnative bivalve molluscs, *Perna viridis* (Linnaeus 1758) and *Mytella charruana* (d'Orbigny 1846). Journal of Marine Biology. 14 pp [doi.org/10.1155/2016/9261309]

Yukihira H, Lucas JS, Klumpp DW (2000) Comparative effects of temperature on suspension feeding and energy budgets of the pearl oysters *Pinctada maxima* and *P. margaritifera.* Marine Ecology Progress Series 195: 179-188

Zar JH (2010) Biostatistical analysis (1st ed). Prentice Hall, Upper Saddle River, New Jersey. 944 pp