

Research Article



Assessment of tidal and lunar influence on larval diversity and distribution along the CVCAs of tropical Kali estuary, Karwar, West coast of India

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Abstract

Larval ingress, diversity and distributions were studied for a period of 13 months along the critically vulnerable areas of the tropical Kali estuary, Karwar, West coast of India. Planktonic and hydrological samplings were carried out following the tidal regime and lunar periodicity at Zone (1) (categorized as euhaline <30 PSU to polyhaline 18.0 to 30 PSU in condition), subsequently, Zone (2) (mesohaline 5.0 to 18.0 PSU to oligohaline 0.5 to 5.0 PSU). A total of 26 different larval forms were recorded, representing 10 taxa, of which crustaceans are dominant. The predominant larval forms documented during the study period were Copepod nauplii, Barnacle nauplii, Eggs & fish larvae and Crab zoea, respectively. Larval forms, viz., *Planula*, *Cercaria*, *Rhabditiform*, *Alima* (*Squilla*), *Bipinnaria*, *Ophiopluteus* and *Doliolaria* distributions was restricted to euhaline zone due to tidal ingress. The distribution of Bivalve, Crab and Shrimp larvae along the mesohaline zone represents larval recruitment in their preferred habitat. The larval distribution showed typical seasonal variation governed by hydrological parameters and tidal regimes. Multivariate RDA analysis of larval distributions attributed to salinity and water temperature. The Shannon-Wiener diversity index was higher (2.92) during the post-monsoon season in particular to full moon of the flood tide. Overall, the Kali Estuary represents a suitable habitat for juvenile nurture and larval ingress enhances the estuarine productivity.

Keywords: Larvae, Tides, Diversity, Distribution, Kali Estuary

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Introduction

Tropical semi-enclosed estuaries ranked among the world's most productive and dynamic ecosystems (Sreekanth *et al.*, 2017). These brackish water wetland ecosystems are the most challenging habitat and are the gateway for migratory species to complete their life cycle. Moreover, mangrove canopy along the mudflats of estuaries enhances biological productivity subsequently, offers an essential complex structural habitat for juvenile nurture. Also, the estuarine environment provides ample food resources for the early life stages of organisms (Holt and Holt, 2000). Most of the marine faunal life cycle accomplishes in these shallow sheltered zones as the tidal cycle bridges their ingress and exit. In particular specific ecological-based larval migration evokes mainly through the flood tide transport, which includes olfactory cues, currents, salinity, temperature and turbulence associated with tidal amplitude and phase between shelf region to estuarine waters (Forward and Tankersley, 2001). Similarly, the limiting factors for spatial species recruitment and distribution across the estuary are salinity, temperature, pH, dissolved oxygen, turbidity and nutrients (Darboe, 2002; Perumal *et al.*, 2009; Badsu *et al.*, 2010; Cloern *et al.*, 2017; Tanmoy and Mandal, 2020). Moreover, Queiroga and Blanton (2004), state that successful estuarine species recruitment depends upon the behavioral strategies employed by larvae. The estuarine-dependent robust

resilient larvae grow to the adulthood along the favorable salinity gradient, significantly enhance the regional biodiversity and associated fishery.

Among the crustaceans, molluscan and pisces larval ingress holds a key position from the commercial perspective (Katsanevakis *et al.*, 2008; Kunze *et al.*, 2013; Dias *et al.*, 2020; Tarimo *et al.*, 2022). Thus, larval ingress totally depends on the hydrodynamics and environmental signals to selectively tidal and meteorologically driven currents back into the estuaries and avoid being flushed back out when these currents reverse (Buskey, 2018). Variability in the delivery of larvae from offshore to the nursery ground up to the upstream freshwater zone will certainly impact the population dynamics, trophic structure and associated fisheries.

Larvae are the post-embryonic stages of the life cycle, which morphologically differ from the adults and might possess independent locomotion. Many marine species represent a bipartite life cycle characterized by a dispersive larval phase and a sedentary adult phase. Thus, quantifying larval forms and their dispersal is a goal of ecology and conservation (Kritzer and Sale, 2010). The spatial and temporal distribution of planktonic larval studies carried out across the globe are restricted to species/group viz., penaeid prawn larval ingress (Staple and Vance, 1985; Goswami and Usha, 1992), brachyuran crab larval distribution (Garrison, 1999) and fish larvae (Rao, 1970; Hare *et al.*, 2005).

In general, larval diversity and ingress documentation was lacking. Against this backdrop, an attempt has been made to document the overall larval ingress, distribution, seasonal abundance, and wetland utility to forecast the nursery role of the estuarine ecosystem along the Kali estuary, Karwar, West coast of India.

Materials and methods

Study area

The Kali estuary is a typical, mesotidal well-mixed tropical estuarine ecosystem of the West coast of India ($14^{\circ} 50' 19.19''$ N - $74^{\circ} 07' 13.80''$ E). The river originates at the plateau of the Western Ghats, flows westward for about 184km, and layouts into the Arabian Sea, Karwar. The estuary has a narrow opening followed by a width of 1.5km in the mid-region with an average water column of 10m. Kali estuary is a tropical monsoon-influenced ecosystem in which the southwest monsoon (June-September) average rainfall recorded was 2760mm (IMD- Indian Meteorological Department), of which June and July were the wettest months. Tides in this tropical coast are mixed semi-diurnal with a large variation in the tidal range of 1.5 to 2.5m. The mangrove canopy of 606.47ha offers a network of sheltered zones with innumerable ecological services (Mesta *et al.*, 2016) along the Kali estuary. Being a resilient habitat, this wetland ecosystem was declared as a critically vulnerable coastal area (CVCA) by Indian Coastal Zone Management Authorities (CRZ, 2011).

Sampling stations were set along the estuarine stream based on the salinity gradient and active fishing pockets. Based on the salinity gradient, Zone 1 (Lat $14^{\circ} 50' 33''$ N; Long $074^{\circ} 08' 11''$ E) was the main channel of the estuary which categorized as euhaline (<30 PSU) to polyhaline (18.0 to 30 PSU) in nature. Zone 2 (Lat: $14^{\circ} 50' 18''$ N Long: $074^{\circ} 10' 00''$ E) is a backwater wetland zone of the estuary categorized as mesohaline (5.0 to 18.0 PSU) to oligohaline (0.5 to 5.0 PSU), besides densely covered mangrove vegetation (Fig. 1). Kali estuary supports the livelihood of fishermen as traditional fishing activity prevails throughout the year.

Field sampling methods and laboratory analysis

Chemical and biological samples were collected at monthly intervals following the tidal (flood tide and ebb tide) and lunar phases (full moon and new moon) for a period of 13 months (January 2018 to January 2019) at two fixed study zones. Field samplings were carried out during the daylight of flood tide and ebb tide, wherein fortnightly samplings were carried out during the nocturnal rising lunar tide. Sampling regimes were followed on a specific duration of mid-flood tide and subsequent ebb tide. Larval samples were collected through a plankton net 50 μ M mesh size, (0.5m mouth diameter) hauled horizontally for a period of 10 min at uniform speed through a motorized boat from surface water up to the depth of 5m.

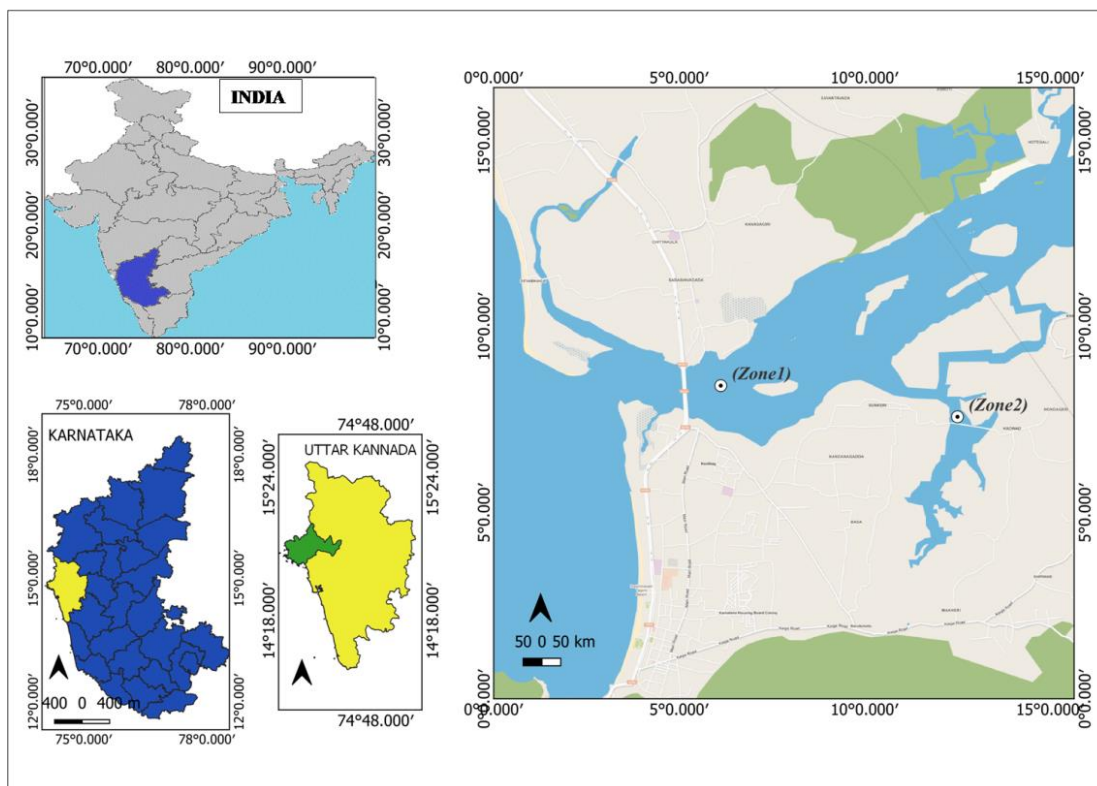


Figure 1: Map showing study area and location of sampling zones along Kali Estuary, Karwar, West coast of India.

A flowmeter attached to the plankton net registered the volume of water filtered (Archambault and Bourget, 1999; Goncalves *et al.*, 2010). Samples collected were transferred to labeled bottles and preserved in 5% neutralized formalin. The known volume of the sample was concentrated to the desired volume (10mL). Samples were homogenized by vigorous random stirring and a triplicate sub-sample of 1ml each was transferred to the Sedgwick counting chamber for enumeration. The mean of sub-samples is calculated and multiplied by the known volume of the sample. Larval samples were identified based on relevant literature (Young *et al.*, 2002; Suthers and Rissik, 2009; Conway, 2012a, 2012b; Weiss, 2017) through a compound microscope fitted with a

camera at a magnification of 4X, 10X and 40X. In parallel to larval sampling, water parameters, viz., pH, salinity (PSU), water temperature ($^{\circ}\text{C}$) and dissolved oxygen (mg/L) were recorded in-situ through a hand-held multiprobe water analyzing kit (SD 400 Oxi L-Lovibond).

Statistical analysis

Environmental variables analyzed for tidal and lunar phases were grouped seasonally, Pre-monsoon (PRE), Monsoon (MON), and Post-monsoon (POM) seasons. The seasonally categorized environmental variables were subjected to analysis of variance (ANOVA) with IBM SPSS (Statistical package for the social science ver. 22) software to find the variance of means among seasons and studied zones

followed by Pearson's correlation coefficient to assess the interrelationship. Prior to the data analysis, all data sets were checked for normality using the Kolmogorov-Smirnov test and homogeneity of variances using the Levene test (Sokal and Rohlf, 1995). Further, to test the variance between seasonal larval distributions, a non-parametric Kruskal-Wallis test (two or more groups) was performed to derive the significance between the seasons, followed by a post-hoc Mann-Whitney's U-test (two groups). Univariate diversity indices for larval community viz., the Shannon-Wiener (H) diversity index (Shannon, 1948) was computed (Tidal and Lunar phase) through the PAST software (PAleontological Statistics Ver. 4.07b).

In order to elucidate the relationship of larval distribution with environmental parameters a Detrended correspondence analysis (DCA) was

carried out. The DCA gradient plot did not exceed the deviation of 3.0, thus multivariate Redundancy Data Analysis (RDA) was employed to discover the relationship between larval distribution and environmental factors. RDA was plotted to determine the correlations between tidal-larval diversity with environmental parameters, similarly, lunar-larval diversity and environmental parameters, followed by seasonal-larval diversity and environmental parameters. Only dominant larvae were considered and rare larvae were omitted subsequently, statistical significance was evaluated based on the Monte Carlo Permutation Test (MCPT) with CANOCO 4.5 software (Leps and Smilauer, 2003).

Results

Environmental clauses

The Physicochemical parameters studied during the study period are represented in (Table 1 and Fig. 2).

Table 1: Zone wise environmental parameters with seasonal significance difference.

Water parameters	PRE	MON	POM	mean±SD	Seasonal significance	
					F	p
pH	7.7±.21	7.7±.30	7.7±.2	7.7±.24	0.647	0.526
Dissolved oxygen (mg/L)	4.9±.87	6.0±.71	5.1±.7	5.38±.90	16.427	0.000
Salinity (PSU)	27.1±6.8	13.3±10.9	26.7±7.6	22.7±10.61	27.952	0.000
Water temperature (°C)	27.9±1.7	25.5±2.03	27.4±2.25	27.03±2.26	12.155	0.000

Significance level at 5%

The physicochemical characteristics studied for the tides and lunar phases are categorized primarily into three conventional seasons; southwest monsoon (June-September), post-monsoon (October-January), and pre-monsoon (February-May), respectively. Kali estuary is majorly influenced by

river run-off where salinity markedly varies seasonally. The mean salinity values were higher during pre and post-monsoon (27.17 ± 6.8 and 26.7 ± 7.6 PSU) seasons wherein, salinity sharply decreased during the southwest monsoon (13.3 ± 10.9 PSU).

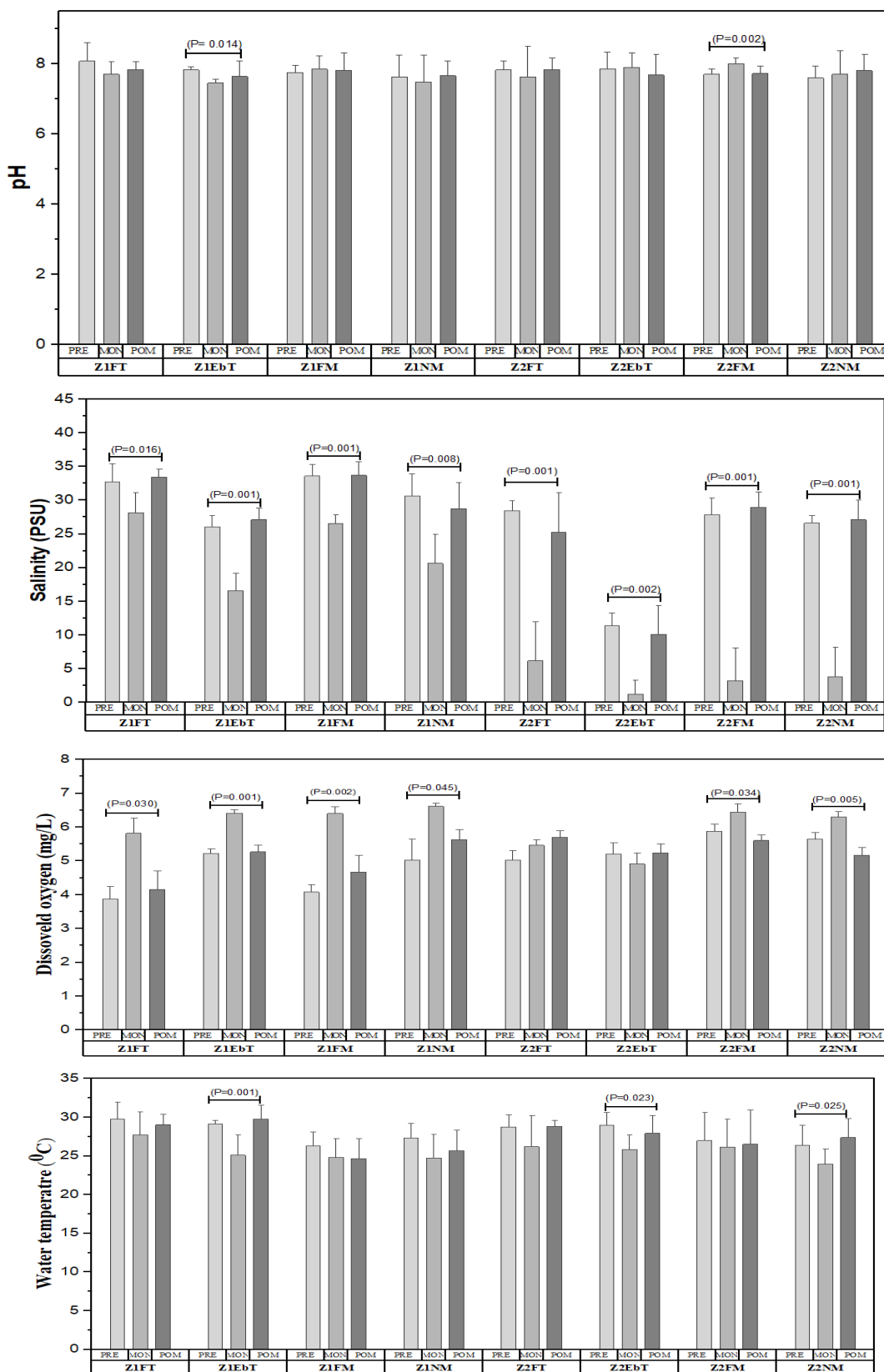


Figure 2: Spatio-temporal variations of environmental parameters with significance difference.

The water temperature followed the seasonal cycle with a mean of $27.03 \pm 2.26^\circ\text{C}$, high-temperature values represented during the pre and post-monsoon season (27.9 ± 1.79 and $27.4 \pm 2.25^\circ\text{C}$). The pH value ranged from 7.1 to 8.3 with a mean of 7.7 ± 0.24 , wherein, dissolved oxygen varied from 3.12 to 6.93 mg/L with a mean of 5.38 ± 0.90 mg/L during the study period. Overall, seasonal significant differences ($p < 0.05$) were observed in the water temperature and salinity, followed by dissolved oxygen (Table 1). Moreover, the physicochemical parameters of the tidal regime and lunar phase also represented seasonal significant variation between the studied zones (Fig. 2). A significant variation ($p = 0.016$) of salinity and dissolved oxygen ($p = 0.030$) was observed during flood tide at Zone (1), whereas, salinity ($p = 0.001$) was significant at Zone (2). The significant variation during the ebb tide was represented by pH ($p = 0.014$), salinity ($p = 0.001$), dissolved oxygen ($p = 0.001$) and water temperature ($p = 0.001$) at Zone (1), whereas, salinity ($p = 0.002$) and water temperature ($p = 0.023$) were significant at Zone (2). Similarly, the lunar phase showed seasonal significance for salinity ($p = 0.001$) and dissolved oxygen ($p = 0.002$) at Zone (1), whereas, pH ($p = 0.002$), salinity ($p = 0.001$) and dissolved oxygen ($p = 0.034$) at Zone (2) during the full moon phase. During the new moon phase, salinity ($p = 0.008$) and dissolved oxygen ($p = 0.045$) were statistically significant at Zone (1), wherein, salinity

($p = 0.001$), dissolved oxygen ($p = 0.005$), and water temperature (0.025) at Zone (2) (Fig. 2). The person's correlation coefficient showed a strong positive correlation between the water temperature and salinity ($r = 0.889$, $p = 0.037$) during the study period.

Percentage composition of larvae distribution

A total of 26 larval forms of 10 taxa, namely Cnidaria, Platyhelminthes, Nematoda, Annelida, Mollusca, Arthropoda, Echinodermata, Bryozoa, Brachiopoda and Ichthyoplankton, were recorded in this study. Arthropods and crustaceans were the dominant group, with Copepod nauplii (15.30%), Crab zoea (8.50%), Barnacle nauplii (8.30%) followed by Shrimp zoea (8.20%), respectively. The Egg & fish larvae (10.3%), Bivalve veliger (6.90%) and Gastropod veliger (6.10%) are the other major predominant groups (Table 4). During the study period, larval forms such as Polychaete larvae, Barnacle nauplii, Barnacle cypris, Shrimp nauplii, Shrimp zoea, Shrimp mysis, Shrimp post-larvae, Crab zoea, Copepod nauplii, Bivalve veliger, Gastropod veliger, *Cyphonautes* and Egg & fish larvae were the representatives of both the studied zones (Tables 2 to 4).

The ingress of larval forms was markedly influenced by the tidal regimes. During the flood tide, *Ephyra*, *Bipinnaria* and *Ophiopluteus* were recorded throughout the year along the euhaline zone, wherein *Planula*, *Cercaria*, *Rhabditiform*, *Alima*

(Squilla), and *Doliolaria* larvae were recorded during the pre-monsoon and post-monsoon seasons.

Table 2: Tidal and lunar seasonal percentage (%) composition of larvae at zone 1.

Larval forms	ZONE 2											
	Flood tide			Ebb tide			Full moon			New moon		
	P	M	PO	P	M	PO	P	M	PO	P	M	PO
Cnidaria												
<i>Ephyra (Ep)</i>	3.0	x	2.5	1.2	x	1.0	5.2	1.9	2.5	3.2	x	3.4
<i>Planula (Pla)</i>	x	x	0.1	x	x	x	0.2	x	0.4	0.2	x	0.2
<i>Actinula (Act)</i>	0.6	x	4.1	4.9	x	4.6	3.7	4.3	1.4	2.8	x	2.8
Platyhelminthes												
<i>Cercaria (Cer)</i>	x	x	x	x	x	x	0.3	x	0.3	X	x	x
Nematoda												
<i>Rhabditiform (Rha)</i>	0.3	x	0.1	x	x	x	0.3	x	0.2	X	x	x
Annelida												
Polychaete (Pol)	4.2	3.3	4.9	3.4	2.5	4.8	5.7	1.8	6.7	7.5	1.3	6.5
Arthropoda												
<i>Alima (Al)</i>	x	x	x	x	x	x	x	x	0.1	X	x	x
Barnacle nauplii (BNau)	7.9	6.1	9.3	7.5	1.3	10.7	9.8	3.7	7.5	9.2	2.6	8.9
Barnacle cypris (BCyp)	10.8	5.9	5.4	5.4	8.2	6.8	6.9	4.5	7.1	8.1	2.0	6.6
Shrimp nauplii (NauSr)	2.1	0.6	1.1	0.4	1.0	0.8	1.1	1.0	2.0	1.3	1.0	1.4
Shrimp zoea (ZoSr)	3.6	5.7	10.6	10.4	8.3	9.0	8.6	6.3	7.2	3.5	11.6	9.7
Shrimp mysis (MySr)	5.3	4.6	2.5	4.7	13.5	5.8	2.1	9.6	3.2	5.2	3.6	2.3
Shrimp Post-larvae (PLSr)	0.8	1.0	2.1	3.1	3.3	1.2	4.3	6.1	4.9	4.0	2.9	4.6
Ephausiid nauplii (EpNau)	1.3	x	0.4	0.4	x	0.3	2.0	0.3	4.5	3.5	1.5	3.2
Crab zoea (CrZo)	7.7	5.8	9.0	6.9	3.2	10	6.5	2.4	6.8	6.6	6.8	5.8
Megalopa (Meg)	0.1	0.8	0.6	0.2	x	0.5	0.1	0.2	0.6	0.3	0.7	0.5
Crab Post-larvae (PICr)	0.4	0.2	0.5	0.5	0.7	0.6	0.5	x	1.2	1.2	x	0.4
Copepod nauplii (NaCop)	17.9	31.5	12.1	17.9	36.2	13.4	16.3	11.9	11.1	16.9	32.4	10.8
Mollusca												
Bivalve veliger (BiVvel)	5.3	9.4	7.1	7.5	3.6	7.3	2.9	14.1	5.6	3.7	13.7	5.9
Gastropod veliger (GaVel)	7.8	8.4	4.5	6.8	3.1	9.2	1.6	13.6	4.3	4.1	12.8	5.0
Echinodermata												
<i>Bipinnaria (Bip)</i>	0.6	x	0.3	x	x	x	0.5	0.8	0.6	0.1	x	0.1
<i>Ophiopluteus (Oph)</i>	0.5	x	1.1	0.8	x	0.2	4.2	1.1	3.7	1.2	0.5	1.3
<i>Doliolaria (Dol)</i>	0.6	x	0.5	0.3	x	0.3	0.8	x	0.9	0.5	x	0.5
Bryozoa												
<i>Cyphonautes (Cyp)</i>	3.9	3.4	4.8	0.7	0.6	0.4	4.8	0.6	2.5	1.7	1.5	3.0
Brachiopoda												
<i>Lingula (Lin)</i>	2.6	5.8	3.3	2.5	x	2.7	6.2	8.5	5.2	6.2	0.8	6.8

Table 3 continued:

<i>Doliolaria</i> (Dol)	x	x	x	x	x	x	x	x	x	x	x	x	
Bryozoa													
<i>Cyphonautes</i> (Cyp)	x	x	0.1	x	x	x	x	x	x	x	x	x	
Brachiopoda													
<i>Lingula</i> (Lin)	6.9	1.4	2.0	1.0	0.8	3.4	8.0	2.5	5.3	3.9	x	3.0	
Ichthyoplankton													
Egg&Fish larvae (Eg&F)	14.7	14.8	9.5	17.6	10.3	12.5	12.6	10.7	9.5	12.6	18.6	7.8	
TOTAL	16	15	18	15	13	15	16	12	17	15	8	17	

Note: 'x' indicates absence of larvae, P-Pre-monsoon, M-monsoon, Po-Post-monsoon.

Among the lunar phases, the new moon represented fewer larval forms than the full moon.

Larval abundance showed significant seasonal variation ($p < 0.001$) between the studied zones through the Kruskal-Wallis test (Table 5). Furthermore, Mann-Whitney's U test signifies the significance between pre-monsoon and monsoon seasons (PRE and MON),

followed by monsoon and post-monsoon seasons (MON and POM), respectively ($p < 0.05$). There were no significant variations ($p > 0.05$) between the pre-monsoon and post-monsoon seasons (PRE and POM) for the tidal cycle. However, a significant difference was observed during the full moon phase at Zone (1) (Table 6).

Table 4: Mean, SD and larval percentage (%) composition (zone 1 and zone 2).

Larval forms	Mean (ind. m ⁻³)	SD (ind. m ⁻³)	Percentage (%)
Cnidaria			
<i>Ep</i>	492.9	921.99	1.5
<i>Pla</i>	28.6	124.09	0.1
<i>Act</i>	720.71	1121.59	2.2
Platyhelminthes			
<i>Cer</i>	21.09	139.2	0.1
Nematoda			
<i>Rha</i>	17.47	110.82	0.1
Annelida			
<i>Pol</i>	1904.8	2106.12	5.7
Arthropoda			
<i>Al</i>	2.08	21.18	0
<i>BNau</i>	2737.11	2631.58	8.3
<i>BCyp</i>	2243.87	2139.60	6.8
<i>NauSr</i>	258.81	530.69	0.8
<i>ZoSr</i>	2714.11	2632.42	8.20
<i>MySr</i>	1580.81	1700.77	4.80
<i>PlSr</i>	1325.51	1637.11	4.00
<i>EpNau</i>	606.85	1334.91	1.80
<i>CrZo</i>	2828.63	2604.32	8.50
<i>Meg</i>	110.26	247.22	0.30

Table 4 continued:			
<i>PICr</i>	233.35	510.72	0.70
<i>NaCop</i>	5054.89	3728.3	15.30
Mollusca			
<i>BiVvel</i>	2271.77	2273.74	6.90
<i>GaVel</i>	2014.35	1997.96	6.10
Echinodermata			
<i>Bip</i>	61.63	210.50	0.20
<i>Oph</i>	344.02	999.40	1.00
<i>Dol</i>	100.44	251.32	0.30
Bryozoa			
<i>Cyp</i>	511.66	1323.72	1.50
Brachiopoda			
<i>Lin</i>	1483.96	2042.28	4.5
Ichthyoplankton			
<i>Eg&F</i>	3463.13	2882.09	10.3

The total mean larval abundance during the full moon phase was (2059±2809 ind. m⁻³), of which post- monsoon season represented maximum mean abundance of (3297±3502 ind. m⁻³) (Table 5 and Fig. 3).

Table 5: Total mean larval abundance (mean±SD) between seasons (Kruskal-Wallis test).

	PRE	MON	POM	Total mean abundance (ind. m ⁻³)	df	P
	mean±SD	mean±SD	mean±SD	mean±SD		
Zone1 FT	996±1402	557±1203	1880±2666	1201±2020	2	<0.001
Zone2 FT	870±1241	327±649	1720±2816	1030±1995	2	<0.001
Zone1 EbT	844±1313	337±809	1704±2424	1019±1819	2	<0.001
Zone2 EbT	831±1380	203±555	1167±1883	767±1481	2	<0.001
Zone1 FM	1761±2140	812±1514	3297±3502	2059±2809	2	<0.001
Zone2 FM	1356±1729	239±957	2369±3023	1402±2339	2	<0.001
Zone1 NM	1462±1732	411±950	2720±3060	1623±2388	2	<0.001
Zone2 NM	1223±1572	113±389	1776±2522	1094±1928	2	<0.001

Table 6: Total mean larval abundance (mean±SD) with significance difference between two seasons (Mann-Whitney's U test).

ZONES	PRE*MON	PRE*MON	PRE*POM	PRE*MON
	Z	Asymp. Sig.(2-tailed)	Z	Asymp. Sig.(2-tailed)
Z1FT	3.496	0.001*	-2.048	0.122
Z2FT	2.869	0.012*	-1.011	0.937
Z1EbT	3.266	0.003*	-2.365	0.054
Z2EbT	3.357	0.002*	-0.789	1.000
Z1FM	3.861	0.001*	-2.874	0.012*
Z2FM	4.549	0.001*	-1.325	0.555
Z1NM	5.334	0.001*	-2.283	0.067
Z2NM	6.040	0.001*	-0.234	1.000

*Implies the result is significant at $p < 0.001$, Z is absolute average value, PRE-pre-monsoon; MON-monsoon; POM- post-monsoon.

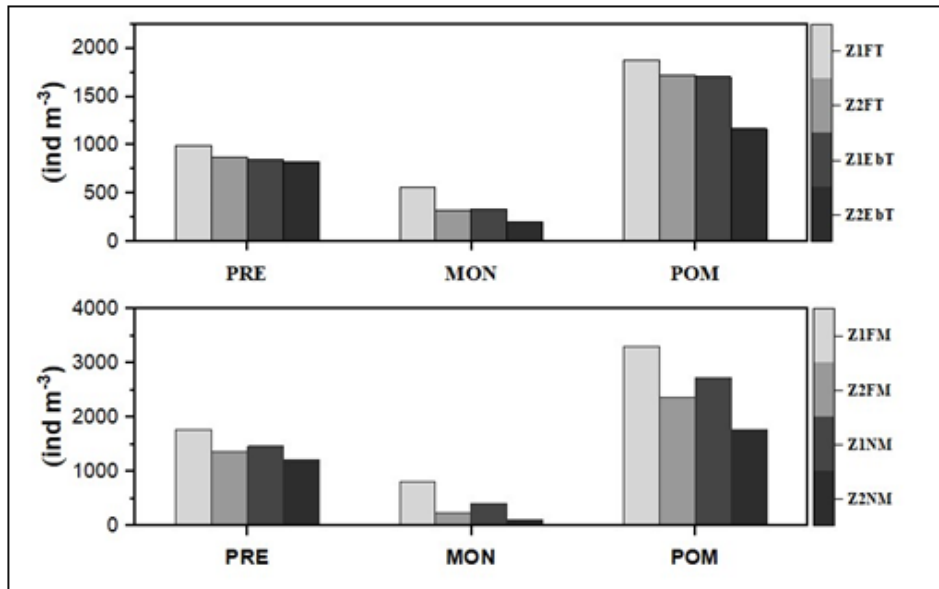


Figure 3: Total mean abundance of larval distribution across zones according to seasonal, tidal and lunar cycle.

Larval diversity indices

The Shannon Wiener (H) diversity index (mean=2.44, SD= 0.31) ranged from 1.64 to 2.92. During the study period, the Shannon wiener index increased at Zone (1) of the full moon during post-monsoon season (26 larvae

and $H=2.92$) (Fig. 4). However, diversity index enormously varied from studied zones explaining Zone (1) with highest diversity (post-monsoon season) and Zone (2) with minimum diversity predominantly in monsoon season.

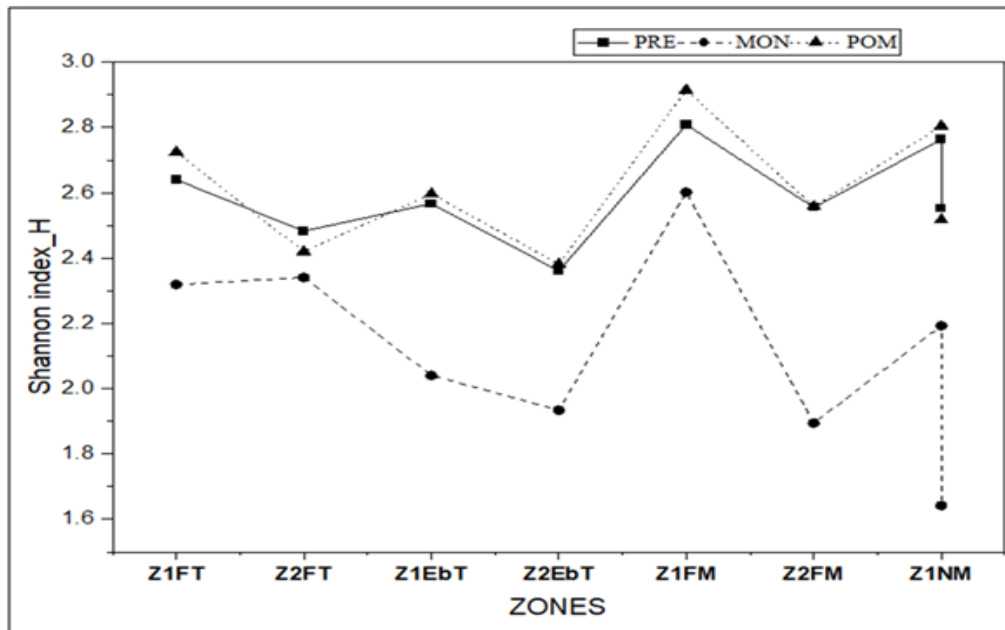


Figure 4: Shannon Wiener (H) diversity indices.

Redundancy data analysis (RDA) for larval distribution

The results of multivariate Redundancy Data Analysis (RDA) combining physicochemical variables with dominant larval communities were depicted in triplots (Figs. 5 to 7). The larval-environmental relations for the first two axes varied from 66.3 to 22.6% and 47.7 to 46.9% for the tide and lunar phases, respectively (Figs. 5 and 6). Among the environmental factors, the Monte Carlo permutations test showed that salinity, water temperature, dissolved oxygen and pH were significant in explaining the relationship between the larval and tidal cycle ($F=13.521$, $P=0.002$) contrarily,

there was no statistically significant variation ($F=3.93$, $P=0.68$) observed for larvae and lunar phase.

The RDA (Fig. 5) explains a strong positive correlation of larvae like *Planula*, *Rhabditiform*, *Bipinnaria*, *Cyphonautes*, *Megalopa*, *Ophiopluteus*, *Doliolaria*, *Ephyra*, Shrimp nauplii and *Lingula* with salinity and pH during the flood tide at Zone (1). Further, the larval distributions, viz., Bivalve veliger, Barnacle nauplii, Barnacle cypris, Actinula, Gastropod veliger, Egg & fish and Crab post-larvae were positively correlated to water temperature during the ebb tide.

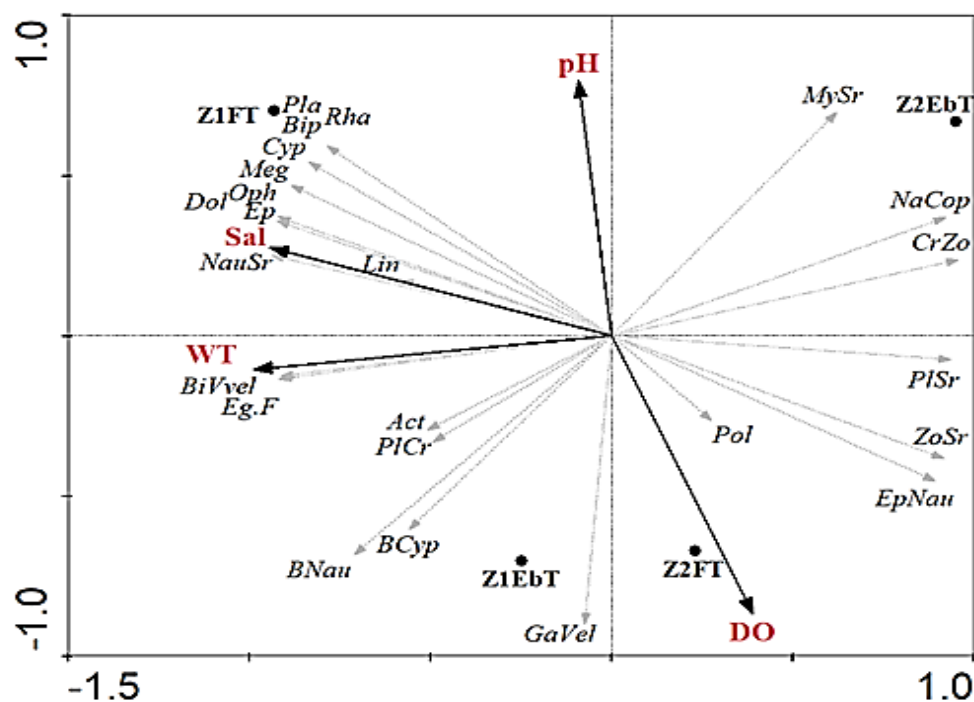


Figure 5: The RDA ordination triplot for larvae with respect to environmental parameters and tidal cycle. Environmental parameters (continuous line) and larvae (dotted lines) are shown by arrows, tides with zones are displayed by filled dots. (Abbreviations: WT- Water temperature ($^{\circ}\text{C}$), DO- Dissolved oxygen (mg/L), Sal- Salinity; Z1FT- Zone 1 Flood tide, Z1EbT – Zone 1 ebb tide, Z2FT- Zone 2 Flood tide, Z2EbT- Zone 2 ebb tide; Larval abbreviation presented in Table 2).

Similarly, the RDA plot for the lunar phase explains the positive correlation for larvae such as *Megalopa*, *Rhabditiform*, *Bipinnaria*, *Ophiopluteus*, *Cercaria*, *Alima*, *Doliolaria*, *Cyphonautes*, *Planula*, Shrimp nauplii, *Actinula* and *Ephyra* with salinity during the full moon and new moon phases at Zone (1)

(Fig. 6).

The RDA eigenvalues for the larval-environmental relationship for the first two axes between seasons were found to vary from 54.5 to 14.2%, registering maximum larvae during the post-monsoon and pre-monsoon seasons (Fig. 7).

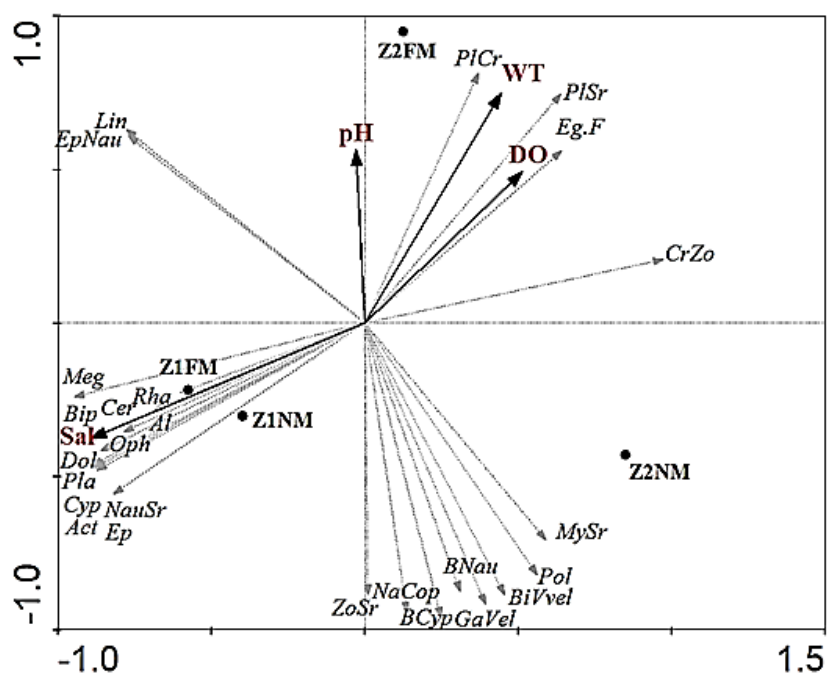


Figure 6: The RDA ordination triplot for larvae with respect to environmental parameters and the lunar cycle. Environmental parameters (continuous line) and larvae (dotted lines) are shown by arrows, lunar phase with zones is displayed by filled dots. (Abbreviations: WT- Water temperature ($^{\circ}\text{C}$), DO- Dissolved oxygen (mg/L), Sal- Salinity; Z1FM- Zone1 Full moon, Z1NM- Zone1 new moon, Z2FM -Zone2 Full moon, Z2NM- Zone2 new moon; Larval abbreviation presented in Table 2).

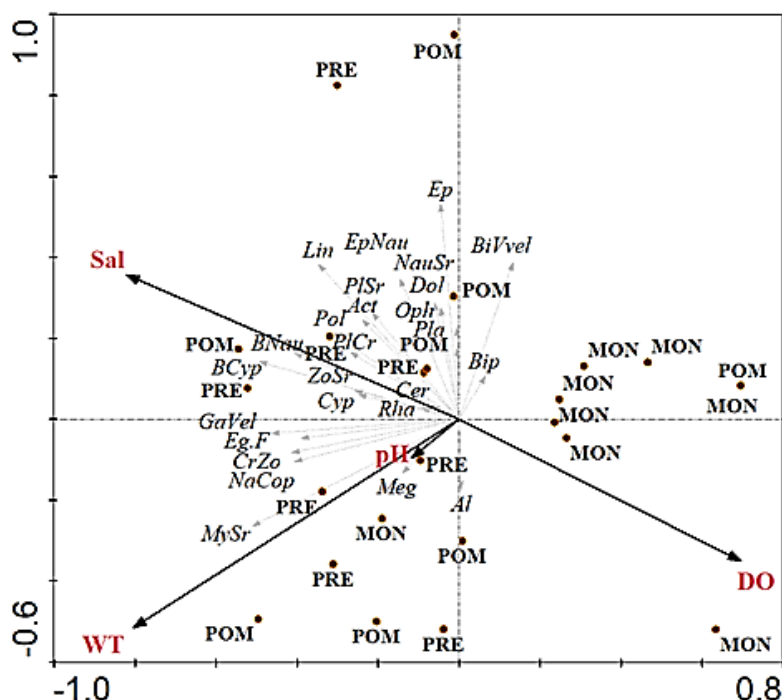


Figure 7: The RDA ordination triplot for larvae with respect to environmental parameters and seasons. Environmental parameters (continuous line) and larvae (dotted lines) are displayed by arrows, filled dots represent seasons (Abbreviations: WT- Water temperature ($^{\circ}$ C), DO- Dissolved oxygen), Sal- Salinity; PRE- Pre-monsoon, MON- Monsoon, POM- Post-monsoon; Larval abbreviation presented in Table 2).

Discussion

Tropical estuaries are biogeochemical hotspots with higher primary production, organic matter and nursery grounds (Baran, 2000). Being the transitional zone between freshwater and marine environment, hydrological parameters vary significantly. In the present study, significant seasonal variations of hydrological parameters were observed, of which water temperature, salinity and dissolved oxygen markedly fluctuated along the study zones. These changes are mainly attributed to regular tidal fluctuations and seasonal changes in the freshwater flow (Costa *et al.*, 2009a). Further, water parameters are generally stable during non-rainy seasons due to well-

mixed tidal salinity, pronounced solar radiation and reduced turbidity along the tropical estuary (Rajaneesh and Mitbavakar, 2013). In the present study, water pH was consistent with persistent alkaline conditions throughout the study at both zones. Dissolved oxygen is the most vital parameter of water quality for biological productivity, which was ambient during the study. Dissolved oxygen levels are affected by rising temperature and salinity during non-rainy seasons (Saravanakumar *et al.*, 2008) and similar trends were observed during the study. The present study infers that salinity is one of the most important parameters influencing the distribution of larvae across the Kali estuary ($p < 0.05$). Seasonal salinity

variations were pronounced during the monsoon season compared to pre and post-monsoon, mainly due to the southwest monsoon and river runoff (Bhat, 1988; VishnuRadhan, 2015; Vineetha *et al.*, 2015; Kumbhar *et al.*, 2020).

The relationship between the distribution of larvae and hydrological parameters has been studied in several estuaries around the globe. Salinity, water temperature, pH, chlorophyll a, and turbidity are the most important parameters which influence the distribution and abundance of larvae (Mouny and Dauvin, 2002). The geo-climatic larval distribution states that salinity mainly regulates larval distribution and abundance along tropical estuaries (Costa *et al.*, 2009b; Atique *et al.*, 2016), wherein, water temperature is responsible across temperate estuaries (Villate, 1994). Moreover, studies showed that the salinity gradient restricts the spatial distribution of planktonic communities across the estuaries due to the explicit tolerance range (Vezi *et al.*, 2019; Rose *et al.*, 2019).

In this study, 26 larval forms were recorded and their distributions were higher at Zone (1), particularly during the post-monsoon season due to the rising tides (Srichandan *et al.*, 2021). Subsequently, Zone (2) is considered a mesohaline zone with 18 larval forms which may be considered as estuarine recruiters. The larval forms such as *Ephyra*, *Planula*, *Alima (Squilla)*, *Bipinnaria*, *Ophiopluteus*, *Cercaria*, *Rhabditiform* and *Doliolaria* are

observed along Zone (1), indicating passive tidal ingress/salinity cues. Similar studies carried out along the prominent estuaries of the east coast of India reported 17 to 21 larval forms with higher larval density during the post and pre-monsoon season. Moreover, their abundances are attributed to the organic matter, catchment area, and stable hydrological parameters post-rainy season (Santhanam and Perumal 2003; Baliarsingh *et al.*, 2014). Such results are well-matched with the present study observations.

With reference to the percentage composition of the larval distribution, *Copepod nauplii* (15.30%), *Egg & fish larvae* (10.52%), *Shrimp zoea* (8.20%), *Barnacle nauplii* (8.30%) and *Bivalve veliger* (6.9%) were found to be dominant during the study period. The following larvae have prolonged larval duration and maximum larval forms were benthic in habitat. *Copepod nauplii* were numerically dominant throughout the studied period, which is attributed to continuous breeding and higher reproductive capacity and subsequently, adaptation to variable environmental conditions (Ramaiah and Nair, 1997; An *et al.*, 2012; Baliarsingh *et al.*, 2014). Dominate larval forms recorded during monsoon season (*Copepod nauplii*, *Egg & fish larvae*, *Bivalve veliger*, and *Gastropod veliger*) can be attributed to their euryhaline adaptations; subsequently, the occurrence of *Egg & fish* larval coincides with the breeding pattern induced by the season (Santhanam and

Perumal, 2003). A study on the barnacle larval population along the well-mixed estuarine gradient reported higher ingress during the post-monsoon period at Mondovi and Zuari estuarine complexes (Grinson *et al.*, 2013), which represents geographical and hydrological similarities with the Kali estuary. Larvae follow the tidally driven migration pattern with higher abundance during the flood tide at both stations. Zone (1) mainly comprised marine habited larval diversity, which indicates intrusion from the adjacent sea, wherein larval compositions along Zone (2) represent marine-brackish with horizontal dispersion to their preferred habitat. During the study, crustacean larval ingress was higher during the lunar phase which demonstrates nocturnal tidal upward movement similar to the study of Srichandan *et al.* (2021).

The ordination of larval and environmental relationship by Redundancy Data analysis (RDA) showed that the increasing salinity influences the dispersal of larvae and other species due to incoming tidal contributes (Mohan and Rao, 1972; Vineetha *et al.*, 2015; Srichandan *et al.*, 2021) whereas, the lunar-tidal cycle is related to the spawning period due to the intensity of moonlight and plays an important role in structuring dispersion of larval forms (Pankhurst and Porter, 2003; Oliveira *et al.*, 2009; Takemura *et al.*, 2010). Crucially, the low diversity and abundance during the monsoon season are due to changes in the coastal

current followed by monsoonal rainfall and river runoff (Rakesh *et al.*, 2008).

Hence, the present study surmises that the larval distributions are typically complemented by the tidal, lunar phases and seasonally fluctuating environmental parameters that shape the estuarine larval complex.

Kali estuary is a critically vulnerable coastal area (CVCA) and its effective ecosystem management and biodiversity accounts are valuable. The estuary supports traditional fishing activity throughout the year and the inner reach of Zone (1) and the meeting point of Zone (2) is considered a bivalve bed. Subsequently, Zone (2) is known as the shrimp fishing basket sustaining the associated fishermen's livelihood. The present study generates the baseline database on larval diversity and distributions, which are critical components of the estuarine food web. Hence, considering the Kali estuary a delicate larval pool and protecting its sensitive habitat is crucial in juvenile management and conservation.

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