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Fragilariopsis sp. bloom causes yellowish-brown waters off Alappuzha, south-central Kerala coast, India, during the mud bank-upwelling phase

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Mud banks (Chakara) of Kerala are calm coastal waters that form in several isolated stretches along the coast usually during the southwest monsoon (SWM) period (June–September). They are characterized by the damping of incident waves, generating localized calm sea environment conducive for fishing activities, while the high monsoon waves create hostile environment in the rest of the region. Here, we present the scientific basis of the yellowish-brown discoloration of water column that occurs off Alappuzha, Kerala annually during the peak and late SWM associated with coastal upwelling-mud bank event. The discoloured waters that occur off Alappuzha associated with these events are locally known as 'pola vellam', which is nothing but diatom blooms. In 2014, pola vellam was actually caused by the bloom of *Fragilariopsis* (= *Fragilaria)* sp.; hereafter *Fragilariopsis*, which was

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widespread in the study region, even beyond the mud bank domain. This bloom feature is attributable to the nutrient enrichment associated with intense coastal upwelling that was dominant over a larger spatial extent in the study domain, including the mud bank. FlowCAM-based plankton data strongly support the above view, as the abundance, biovolume and biomass of *Fragilariopsis* had similar temporal trend both in the mud bank and non-mud bank regions. The general ecology and importance of *Fragilariopsis* bloom in the study domain, from the point of view of commercial fisheries, is also elaborated in this communication.

Keywords: Bloom, coastal upwelling, *Fragilariopsis* sp., mud bank, fisheries.

MUD bank (Chakara) of Kerala is a unique coastal oceanographic feature that occurs in several isolated parts along the Kerala coast, between Alappuzha (south) and Ponnani (north), usually during the southwest monsoon (SWM; June–September). In the mud bank, significant damping of the incident waves results in a calm sea environment conducive for fishing while the high monsoon waves create hostile environment in the rest of the region[1–6]. Relatively high concentration of suspended sediments in the water column derived from a few metres thick, fluid, muddy layer close to the sea bottom is responsible for wave damping and calm sea conditions in the mud bank[1,5,6]. Several hypotheses are in place to explain mud bank formation along the Kerala coast[7]; however, a foolproof, scientifically convincing explanation is yet to evolve. Among the several mud banks that form along the Kerala coast, the one that forms off Alappuzha has attracted special scientific and societal attention due to its consistent occurrence every year and also the rich fishery associated with it[1–4]. Indeed, large stocks of planktivorous fishes (sardine, mackerel and anchovies) and shrimps are being landed on the adjacent shore every year associated with the mud bank off Alappuzha during SWM[2–4].

Another significant and rather widespread oceanographic process that occurs along the southwest coast of India, having profound biophysical impact in the region, is the coastal upwelling operating during SWM[8–13]. The physico-chemical signatures of coastal upwelling include the surfacing of cool, nutrient-rich, low-oxygenated (hypoxic to anoxic) waters towards the coast; these eventually induce significantly high plankton biomass and production in the continental shelf region[8–13]. World over, the coastal upwelling areas possess immense socio-economic significance as they represent regions of high fish availability[14,15]. It is pertinent here to consider that the mud banks of the Kerala coast during SWM actually coexist with coastal upwelling[2]. Therefore, it was not clear earlier whether the enhanced plankton production and high fish landings observed every year associated with the mud bank event was contributed by the mud bank itself or by coastal upwelling, which has a demonstrated biophysical impact over a large spatial extent[2]. This enigma was investigated in an extensive fisheries study in the Alappuzha coastal upwelling-mud bank region spanning several years, which brought to light the fact that the entire area off Alappuzha, predominantly outside the mud bank region, harbours large quantities of fish[3,4]. These studies deciphered the enigma and presented a lucid interpretation that the high fish catch landing that is apparently associated with Alappuzha mud bank is the biological manifestation of coastal upwelling[3,4]. As proposed originally by Banse[8,9], the surfacing of low-oxygenated subsurface water associated with coastal upwelling tends to concentrate the fishes towards surface waters, and these high fish concentrations in the surface waters are being exploited efficiently by fishermen by operating their fishing vessels through the mud bank[7]. This implies that the high fishery landing in the adjacent shore of the mud bank is coastal upwelling-driven and not mud bank-generated[3–5]. In short, the mud bank, being a calm area with smooth sea surface, acts as launching/landing place and safe anchorage for fishing vessels to exploit the coastal upwelling-induced high fish stock available in the surface waters of the mud bank and its vicinity[3–6]. All the above-stated points strongly indicate that the high fish landing associated with Alappuzha mud bank, is no longer a puzzle. Nonetheless, it is a fact that high fish landing occurs on the shore concomitant with the Alappuzha mud bank event, even though its major role is to function as a launching/landing place and safe anchorage for fisher folks (Figure 1).

In 2014, CSIR-National Institute of Oceanography launched a multidisciplinary field programme off Alappuzha (AMPS – Alappuzha mud bank process studies), mainly to track the causes of mud bank formation and sustenance during SWM (Figure 2 *a*). Under AMPS, field sampling in three locations (M1–M3) was carried out from April (pre-monsoon) to September (late-SWM), which included 15 weekly samplings (22 April to 2 August) and three biweekly samplings (16 August to 20 September). Three locations off Alappuzha were sampled in

Figure 1. Typical view of mud bank as a launching/landing place and safe anchorage for fishing vessels.

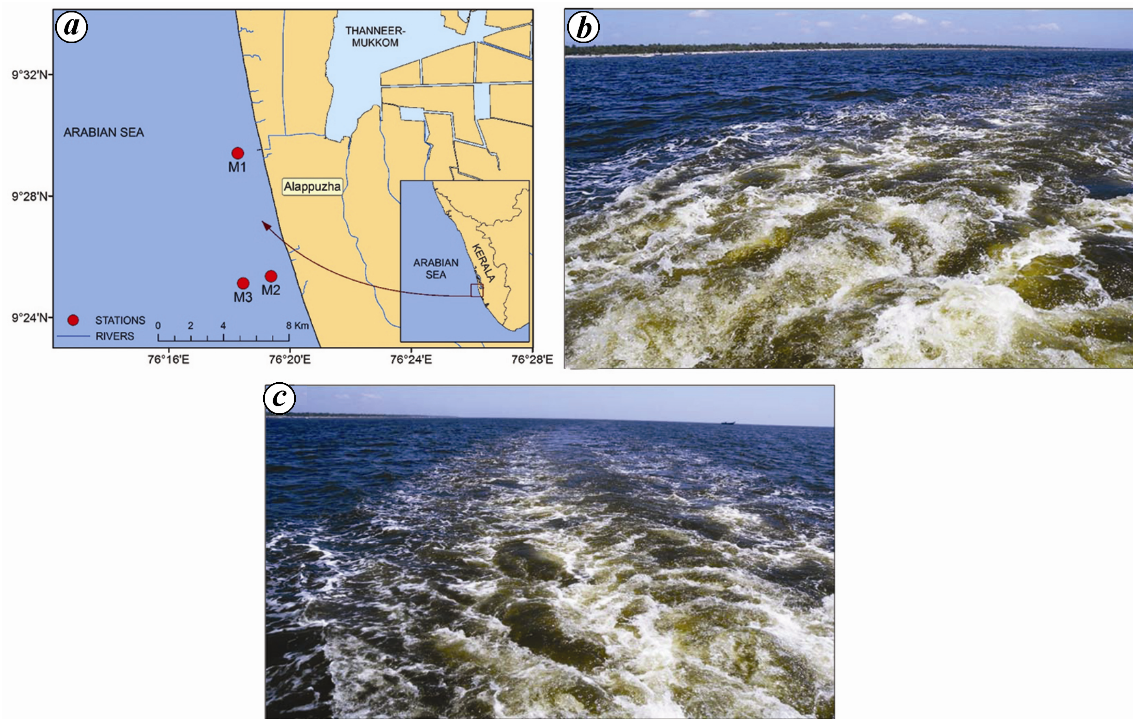

Figure 2. ***a***, Study area and sampling locations; ***b***, ***c***, trails of the boat between locations M1 and M2 that depict yellowish-brown discoloration of water column due to proliferation of *Fragilariopsis* sp., especially in the subsurface waters.

this programme that included one location in the region where mud bank forms during SWM (M2). Two other locations (M1 and M3) were considered as reference points of the mud bank location. M1 and M2 were located at 8 m depth contour, while M3 was at 13 m depth contour. The alongshore distance between M1 and M2 was 8 km, whereas the cross-shore distance between M2 and M3 was 3 km. Typically, the mud bank off Alappuzha forms during the onset of SWM (June), characterized by relatively high suspended sediments and damping of waves in a roughly semi-circular shape[1]. During the present sampling period also, mud bank formed around location M2 by the early SWM (June), signified by visually discernible damping of incident waves and prevalence of calm sea environment in a semi-circular shape.

During the peak and late SWM (July and August), when mud bank was prevalent at location M2, a peculiar discolouration of water was noticed in the entire study area. This discolouration was not only found in all three sampling locations, but also the entire sailing route between the three locations (8 km alongshore between M1 and M2 and 3 km cross-shore between M2 and M3). Importantly, it was noticed that the discolouration was more prominent in the trail of the sampling boat, indicating its high concentrations in the subsurface waters (Figure 2 *b* and *c*). Several sightings of such discoloured water during the course of our time-series sampling during the peak and late SWM period motivated us to examine the history of incidence of such discoloured waters in the study area. According to native fisher folks, such discolouration of the water is a characteristic feature off Alappuzha during the peak and late SWM period, for which the local terminology in use is ‘pola vellam/kadal kara’. Here we present the scientific basis of pola vellam in the near-shore waters off Alappuzha during the peak and late SWM of 2014, and describe the hydrographical setting that favoured such an enigmatic feature, with their possible linkages with the high fishery resources in the study domain.

The data collected using the following methods have been utilized for this study. Temporal variation in the vertical distribution of temperature and salinity in all three locations sampled was measured with a conductivity temperature depth (CTD) profiler (Seabird Electronics, USA). Surface (0.5 m) and subsurface (5 m in M1 and M2, and 8 m in M3) water samples collected using Niskin samplers were analysed for nitrate and dissolved oxygen concentrations[16]. Turbidity of the surface and subsurface water was analysed using a turbidity meter (Eutech TN 100). The temporal change in surface and subsurface chlorophyll *a* concentration was estimated using a Trilogy fluorometer (Turner designs, USA)[17], with water samples collected using a Niskin sampler (Hydrobios, Germany). Water samples from the surface and subsurface were collected from all three locations and the plankton components present in the water column were qualified using a FlowCAM (Fluid Imaging Technologies, USA), which is an advanced semi-automated equipment that combines the principles of both microscopy and flow cytometry[5,18–20]. FlowCAM is an efficient

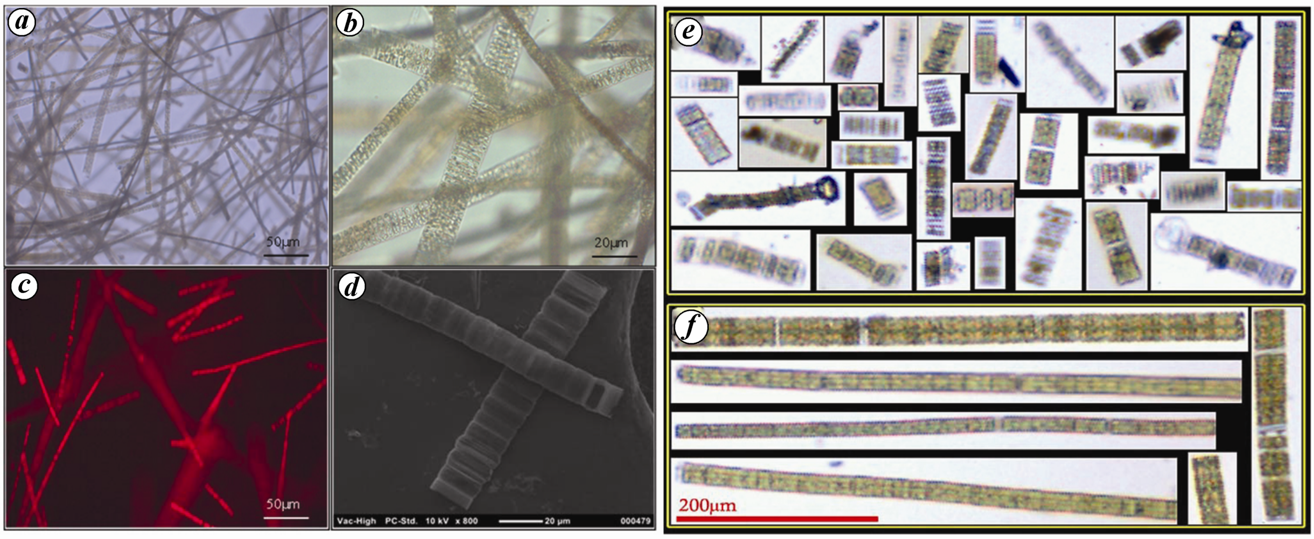

Figure 3. Photomicrographs of *Fragilariopsis* sp. Bloom. (***a*** and ***b***) light microscopy, (***c***) fluorescence microscopy and (***d***) scanning electron microscopy. (***e***, ***f***) FlowCAM images of *Fragilariopsis* sp. during (***e***) pre-monsoon/early southwest monsoon and (***f***) peak/late-southwest monsoon periods. Scale bars of (***e***) and (***f***) are the same; 300 µm FOV flow cell and 4X objective of FlowCAM were used to capture these images.

tool to estimate the biovolume of plankton components present in the water samples, in addition to the usual qualitative and numerical abundance parameters[5,18–20]. Details of all the procedures adopted for the FlowCAM analyses under AMPS have been presented recently[5]. Unpreserved samples were brought to the laboratory in black polythene bottle in cool condition within ~2 h of collection and analysed immediately using FlowCAM. The volume of the samples analysed was chosen on the basis of the nature and abundance of the phytoplankton cells present in the samples during different seasons. The FlowCAM analyses was based on 1000 ml of water samples during the pre/early SWM, whereas only 200 ml of water samples was analysed during rest of the peak and late SWM. Prior to the analyses, water samples were prefiltered using a 300 µm bolting silk and then concentrated with a 20 µm bolting silk to 20 ml volume. This was done by concentrating the prefiltered samples by siphoning through a PVC tube till 20 ml volume remained inside the bottle. One end of the siphoning tube, which was immersed in the sample, was attached with a 20 µm bolting silk for retaining all the particles >20 µm size at the bottom of the sample container. Analysis of each concentrated sample (20 ml) through FlowCAM took around 20 min to complete the imaging of all particles present in the sample. The quantification of phytoplankton using FlowCAM was based on a combination of 300 µm field-of-view (FOV) flow cell and 4X objective. Area based diameter (ABD) algorithm of FlowCAM was used as it measures the biomass of complex biological particles with more accuracy than a traditional microscope[21]. In ABD algorithm of FlowCAM, diameter is measured by the number of grey-scale pixels of the binary image of particles, which is then automatically converted to a circle with the same number of pixels[21]. Subsequently, from the pixel volumes of the image, total biovolume of the individual gets generated, which is more accurate in cases of organisms with extended and protruded body parts[21]. The water samples during each sighting of the discoloured water were collected and analysed with a combination of light (Olympus IX 51), fluorescence (Olympus BX 53) and scanning electron (Neoscope, Nikon) microscopes for obtaining relevant morphological and physical characteristics of the plankton components. Every time during the sighting of the discoloured water, 5–10 Indian oil sardine (*Sardinella longiceps*) specimens were collected from the fish landings on the adjacent shore, which was irrespective of the sampling locations, and the gut contents were inspected qualitatively under an inverted microscope (Olympus IX 51) following standard procedure[22]. This was done to understand whether there is a direct linkage between high landings of oil sardine and discoloured water in the mud bank region during the peak and late SWM period.

Figure 3 presents the results of microscopic analyses of the discoloured water. It is clear that the causative organism of the discoloured water in the study area during peak and late SWM is *Fragilariopsis*, a long, chain-forming, ribbon-like diatom considered to be significant in sustaining high oil sardine fishery along the southwest coast of India[23]. The oceanographic process that favoured the blooming of *Fragilariopsis* in the study domain is evident in the temporal evolution of water column temperature in all three study locations (Figure 4). The most noteworthy feature is the significant drop in temperature in the entire water column (cool waters) during the onset of SWM (June), which indicates intense upwelling occurring in the study region. This upwelling signature was well reflected

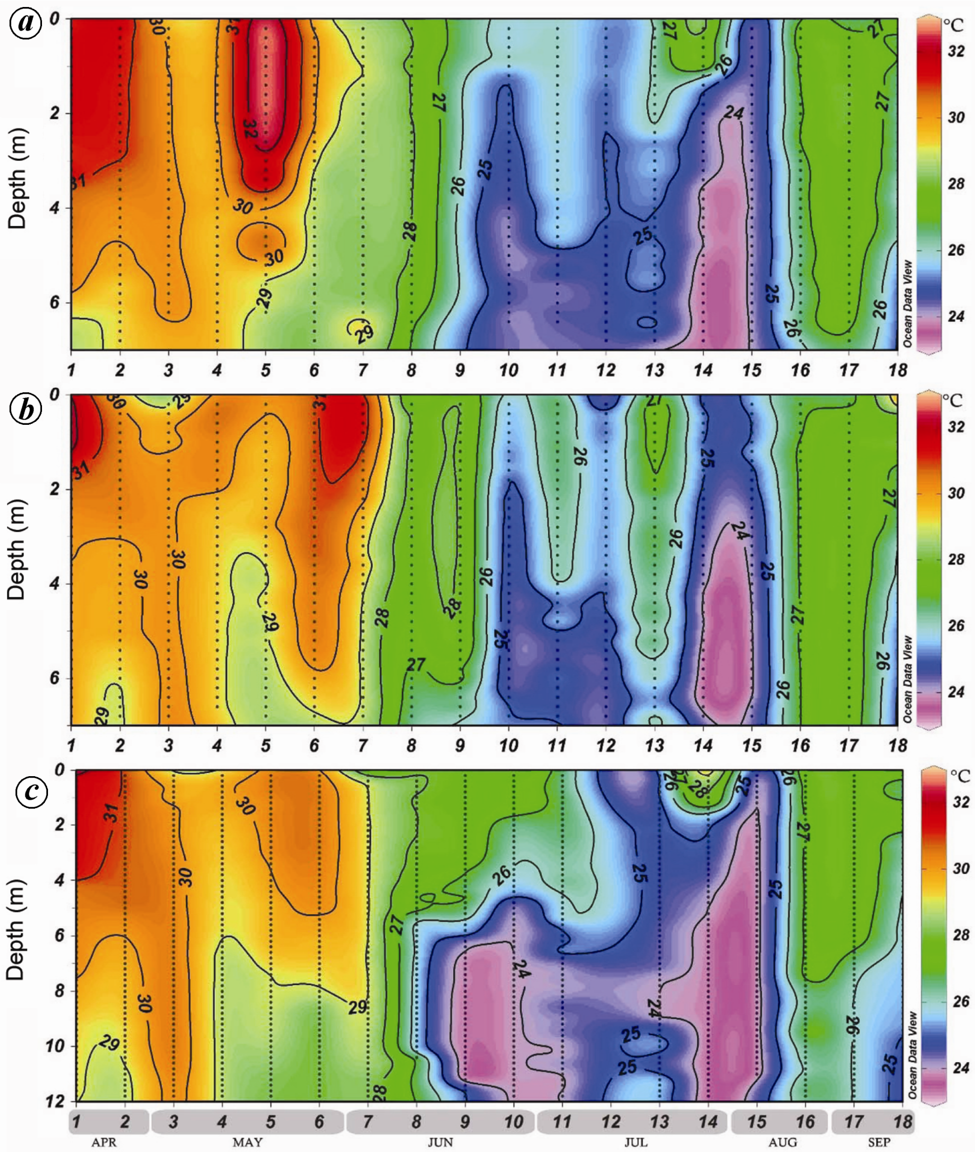

Figure 4. Temporal variation in the vertical distribution of temperature. Intense coastal upwelling is reflected in all three locations studied, i.e. (***a***) M1, (***b***) M2 and (***c***) M3, as a significant drop in water column temperature from June onwards.

in the concentration of dissolved oxygen and nitrate in all three locations (Table 1). Dissolved oxygen showed a large drop, especially in the subsurface waters during SWM, whereas nitrate concentration noticeably increased in the entire water column during the period. High salinity (>30) prevailed in the area throughout the study period, with a relatively low salinity period during late SWM period associated with increased rainfall, land runoff and river influx. There was a general increase in the water column turbidity in all the locations by the onset of the SWM, which was more pronounced in the bottom waters. In the surface waters, relatively more turbidity was found at M2 during SWM due to the prevailing mud bank in the region. Based on the turbidity and its influence on the native plankton community, it can be concluded that the turbidity level in the mud bank is well below the critical level to inhibit plankton growth and sustenance[6]. Similarly, results of the size, composition, abundance and diversity aspects of plankton considered under AMPS, based on FlowCAM data, have recently been published[5]. Importantly, as evident in Table 1, there is a noticeable increase in phytoplankton biomass (chlorophyll *a*) during the three different phases of SWM compared to the pre-monsoon. A significant percentage of this high chlorophyll *a* stock especially during the peak and late SWM, was contributed by the large, ribbon-like chains of *Fragilariopsis* dominant during these periods.

Table 1 presents the temporal variations in abundance, biovolume and biomass of *Fragilariopsis* based on FlowCam measurements. These data reveal some

Table 1. Physico-chemical and biological parameters in three locations (M1–M3) in the study area. Mud bank prevailed in location M2 during the SWM period

Station	Parameters	Depth	Pre-monsoon (April–May)	Early-SWM (June)	Peak-SWM (July)	Late-SWM (August)
M1	Temperature (°C)	S	30.77 ± 1.21	27.45 ± 1.50	26.34 ± 1.30	26.46 ± 1.43
		SS	29.46 ± 0.75	26.82 ± 2.02	24.46 ± 0.65	25.75 ± 1.38
	Salinity	S	34.26 ± 0.64	34.71 ± 0.32	34.15 ± 0.43	32.80 ± 2.45
		SS	34.71 ± 0.42	35.01 ± 0.41	34.48 ± 0.28	34.40 ± 0.84
	Dissolved oxygen (μM)	S	190.48 ± 21.34	172.45 ± 68.90	178.65 ± 37.74	192.18 ± 20.90
		SS	153.64 ± 26.74	66.58 ± 51.53	32.12 ± 25.55	76.19 ± 58.83
	Turbidity (NTU)	S	1.66 ± 0.42	3.67 ± 0.62	5.76 ± 0.72	2.45 ± 1.06
		SS	2.42 ± 0.16	6.65 ± 2.24	11.59 ± 0.71	6.27 ± 0.86
	Nitrate (μM)	S	0.66 ± 0.94	1.18 ± 1.02	3.41 ± 2.13	1.39 ± 0.72
		SS	1.61 ± 1.76	3.65 ± 2.69	6.04 ± 3.27	3.49 ± 2.81
	Chlorophyll *a* (mg m^{-3})	S	1.77 ± 0.85	5.03 ± 2.62	9.3 ± 2.42	3.19 ± 1.84
		SS	1.76 ± 1.59	2.42 ± 1.04	5.20 ± 1.10	3.3 ± 0.49
	Fragilariopsis sp. abundance (ind.l^{-1})	S	66 ± 63	126 ± 79	966 ± 650	470 ± 156
	Fragilariopsis sp. biovolume (mm^3 l^{-1})	S	0.01 ± 0.01	0.03 ± 0.03	0.27 ± 0.12	0.23 ± 0.05
	Fragilariopsis sp. biomass (mgC l^{-1})	S	0.01 ± 0.01	0.02 ± 0.02	0.23 ± 0.12	0.20 ± 0.04
M2	Temperature (°C)	S	30.37 ± 1.11	27.58 ± 1.60	25.84 ± 1.43	27.06 ± 1.61
		SS	29.45 ± 0.58	26.88 ± 1.81	24.80 ± 0.95	25.89 ± 1.70
	Salinity	S	34.67 ± 0.23	33.85 ± 0.75	33.93 ± 0.89	32.93 ± 3.52
		SS	34.71 ± 0.53	34.63 ± 0.69	34.57 ± 0.30	34.10 ± 1.91
	Dissolved oxygen (μM)	S	175.06 ± 24.79	147.8 ± 0.52	199.7 ± 51.15	221.55 ± 34.39
		SS	128.34 ± 40.31	69.55 ± 7.54	34.41 ± 30.10	119.72 ± 76.25
	Turbidity (NTU)	S	1.72 ± 0.26	5.41 ± 0.76	6.74 ± 1.91	3.63 ± 1.96
		SS	3.15 ± 0.50	8.20 ± 2.60	12.85 ± 0.60	5.96 ± 1.72
	Nitrate (μM)	S	0.70 ± 0.69	4.21 ± 3.01	3.25 ± 3.19	1.15 ± 0.97
		SS	1.94 ± 1.02	5.60 ± 4.30	5.87 ± 4.40	3.91 ± 1.86
	Chlorophyll *a* (mg m^{-3})	S	2.66 ± 1.82	4.51 ± 2.92	10.72 ± 7.78	4.98 ± 4.51
		SS	2.09 ± 2.19	1.66 ± 0.68	5.70 ± 1.27	3.86 ± 1.83
	Fragilariopsis sp. abundance (ind.l^{-1})	S	84 ± 70	323 ± 120	1599 ± 930	280 ± 35
	Fragilariopsis sp. biovolume (mm^3 l^{-1})	S	0.02 ± 0.01	0.05 ± 0.02	0.39 ± 0.21	0.92 ± 1.2
	Fragilariopsis sp. biomass (mgC l^{-1})	S	0.01 ± 0.01	0.04 ± 0.02	0.35 ± 0.25	0.77 ± 1.3
M3	Temperature (°C)	S	29.98 ± 1.36	27.52 ± 0.65	26.92 ± 2.08	26.72 ± 1.57
		SS	28.98 ± 0.76	24.33 ± 0.86	24.32 ± 0.79	25.26 ± 1.17
	Salinity	S	34.34 ± 0.65	34.29 ± 0.35	33.97 ± 0.49	32.91 ± 1.08
		SS	34.90 ± 0.40	35.02 ± 0.49	34.70 ± 0.11	35.30 ± 0.28
	Dissolved oxygen (μM)	S	181.22 ± 9.65	147 ± 12.30	225.53 ± 60.55	248.07 ± 73.72
		SS	146.80 ± 30.43	16 ± 11.13	29.94 ± 19.25	41.72 ± 25.23
	Turbidity (NTU)	S	0.61 ± 0.24	1.69 ± 0.72	3.72 ± 1.92	1.37 ± 1.31
		SS	1.71 ± 0.78	10.18 ± 0.30	9.28 ± 0.71	6.24 ± 0.74
	Nitrate (μM)	S	0.37 ± 0.47	0.54 ± 0.42	1.72 ± 1.53	1.32 ± 1.13
		SS	2.28 ± 1.41	11.26 ± 5.44	11.11 ± 2.07	11.35 ± 8.51
	Chlorophyll *a* (mg m^{-3})	S	0.66 ± 0.20	7.38 ± 6.42	10.05 ± 7.63	3.97 ± 2.95
		SS	0.63 ± 0.36	2.13 ± 1.49	6.03 ± 3.01	2.82 ± 2.38
	Fragilariopsis sp. abundance (ind.l^{-1})	S	16 ± 20	98 ± 141	1245 ± 534	360 ± 65
	Fragilariopsis sp. biovolume (mm^3 l^{-1})	S	0.01 ± 0.01	0.04 ± 0.05	0.31 ± 0.17	0.26 ± 0.01
	Fragilariopsis sp. biomass (mgC l^{-1})	S	0.01 ± 0.01	0.03 ± 0.1	0.27 ± 0.15	0.25 ± 0.02

Mean ± standard deviation values are presented. SWM, Southwest monsoon; S, Surface (0.5 m) and SS, Subsurface (5 m in M1 and M2 and 8 m in M3). ind.l^{-1}, Individuals per litre.

important ecological aspects of *Fragilariopsis* in the study domain during different hydrographical settings, as was the case during the pre-monsoon and different phases of SWM. During the pre-monsoon, when the water column was warmer and the nutrient concentration was relatively low, the abundance, biovolume, and biomass of *Fragilariopsis* was also low compared to different phases of SWM. The most striking ecological feature of *Fragilariopsis* observed in the present study is the increase in its biovolume and biomass during the peak and late SWM, which was more pronounced than its increase in abundance during these periods. As mentioned above, the significantly high biovolume and biomass of *Fragilariopsis* observed during peak and late SWM was due to the high abundance of ribbon-like long chains, favoured by cool and nutrient-enriched upwelled waters. The FlowCam images representing the overall size of *Fragilariopsis* during the pre-monsoon/early SWM and peak/late SWM are presented in Figure 3 *e*, which depicts the above aspect. During the pre-monsoon/early SWM, *Fragilariopsis* chains were composed of 2–5 fragments, while during the peak/late SWM, they appeared to be composed of tens of fragments. The long chain-forming character of diatoms under favourable growth conditions in cultured and natural populations has been noticed earlier as well, even though the factors determining the chain length or number of cells per chain in a given species have not been clearly explained[24]. Also, from the preliminary experimental data we have generated along with AMPS sampling, the optimum solar radiation requirements of *Fragilariopsis* blooms along the southwest coast of India is quite low (30–70 μE m^{-2} s^{-1}). This could be the reason why in the present study blooms were found in less-illuminated subsurface waters[5]. Further experimental studies are, however, necessary on the above aspects.

A synthesis of the historical records of *Fragilariopsis* bloom along the southwest coast of India is presented below in relation to the present observations. Nair and Subrahmanyan[23] reported extensive blooms of *Fragilariopsis* off Calicut (Kozhikode) and presented the view that the success of the Indian oil sardine fishery is directly linked with such blooms that form every year. Subrahmanyan[25] has discussed the importance of *Fragilariopsis* in connection with the small pelagic fishes along the southwest coast of India. Devassy[26] also recorded blooms of *Fragilariopsis* along the southwest coast of India and related them with successful oil sardine fishery. Even though the above historical studies described the bloomed diatom, here we do not consider the species-level identity of the bloomed diatom in the study region. This seems to be essential considering the standard phytoplankton identification manual of Tomas[27], which describes *F. oceanica* as a cold-water species. Therefore, more careful and focused studies are required regarding the bloom-forming diatom *Fragilariopsis*, its species-level identity and linkage with monsoon fishery along the southwest coast of India. Nonetheless, the present fish gut content data also underline that *Fragilariopsis* forms a major diet content of Indian oil sardine in the study domain (Figure 5). However, due to the limited data, it is currently premature to draw general conclusions regarding the linkage of *Fragilariopsis* stock and oil sardine fishery success along the southwest coast of India. We suggest that future research on this aspect should consider the fact that *Fragilariopsis* bloom usually occurs close to the southwest Indian coastline, as in the case of the present study and also in historical times[23,26]. Therefore, any future research in this line should focus on near-shore waters along the southwest coast of India. A focused multidisciplinary study considering the above aspects could be useful in resolving the current uncertainty associated with the occurrence of *Fragilariopsis* blooms along the southwest coast of India, and its proposed linkage with the oil sardine fishery in the region.

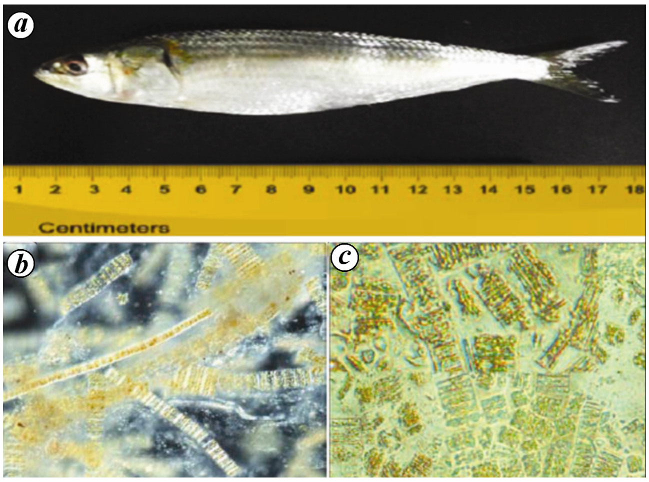

Figure 5. ***a***, Indian oil sardine (*Sardinella longiceps*) specimen from the mud bank region; ***b***, ***c***, *Fragilariopsis* sp. in their gut content.

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AdaBoost-based long short-term memory ensemble learning approach for financial time series forecasting

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A hybrid ensemble learning approach is proposed for financial time series forecasting combining AdaBoost algorithm and long short-term memory (LSTM) network. First, LSTM predictor is trained using the training samples obtained by AdaBoost algorithm. Then, AdaBoost algorithm is applied to obtain the ensemble weights of each LSTM predictor. The forecasting results of all the LSTM predictors are combined using ensemble weights to generate our final results. Four major daily exchange rate datasets and two stock market index datasets are selected for model evaluation and model comparison. The empirical study demonstrates that the proposed AdaBoost-LSTM ensemble learning approach outperform other single forecasting models and other ensemble learning approach in terms of both level forecasting accuracy and directional forecasting accuracy. This suggests that the AdaBoost-LSTM ensemble learning approach is a highly promising for financial time rates forecasting.

Keywords: AdaBoost algorithm, ensemble learning, financial time series forecasting, long short-term memory network.

GLOBAL financial markets function in a complex and dynamic manner as high noisy data volatility is routine. Many factors impact the financial market, such as economic conditions, political events, and even traders' expectations. Hence, financial time series forecasting is usually regarded as one of the most challenging tasks among time series forecasting due to the high degrees of nonlinearity and irregularity. How to accurately forecast stock and exchange rate movement is still an open question with respect to the economic and social organization of modern society.

Many common econometric and statistical models have been applied to financial time series forecasting, such as linear regression models, autoregressive integrated moving average (ARIMA) models[1,2], co-integration models[3,4], generalized autoregressive conditional heteroscedasticity (GARCH) models[1,5], vector auto-regression (VAR) models[6,7] and error correction models (ECM)[4].

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