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A study of morphometric characters of *Ammonia* species in the Chilika lagoon, Odisha, India

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Benthic foraminifera genus *Ammonia* is the most abundant taxon reported in the Chilika lagoon. The detailed identification of the *Ammonia* species of the Chilika lagoon has not been previously carried out. A 70 cm long core was collected from the lagoon's central part, characterized by a high abundance of *Ammonia* in the core's upper 10 cm. Morphometric analysis was carried out on 60 selected *Ammonia* specimens based on 26 external test characters in the spiral and umbilical side views. Pore geometry and density have also been considered as essential morpho parameters for distinguishing different species of *Ammonia*. The present study has reported four different *Ammonia* species, *i.e., Ammonia sobrina*, *Ammonia tepida*, *Ammonia parkinsoniana*, *Ammonia* cf *sobrina*, and two different morphotypes of *Ammonia parkinsoniana* and one morphotype of *Ammonia tepida* down the core. A distinct new pore pattern was examined in a few specimens of *A. parkinsoniana*. Different species of *Ammonia* have further morphofunctional adaption of the habitats; hence their detailed identification could also help interpret the Chilika lagoon's past environments. Stratigraphic distributions of different morphotypes of *Ammonia parkinsoniana* and *Ammonia tepida* in the up core suggest rapid environmental changes in the Chilika lagoon in the last few decades.

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INTRODUCTION

The first genus assigned to Foraminifera (Loeblich and Tappan, 1974) was *Ammonia* (Brunnich, 1772), and its type species, *Ammonia beccarii* was one of the first described foraminiferal species. Still, at first, it was described as *Nautilus beccarii* by Linne (1758). *Ammonia* prefers to live in sheltered, shallow marine, often slightly brackish, intertidal environments, deltaic environments, and stressed environments such as lagoon with high nutrients but mainly avoid high latitudes distribution (Walton and Sloan, 1990; Murray, 2006). Worldwide, *A. beccarii* is the most dominant species in the estuaries and lagoons such as Palar estuary, Pulicat lake and Chilika lagoon in East Coast of India, Ría de Vigo of Spain, and Santa Gilla lagoon of Italy (Diz and Francés, 2008; Frontalini *et al.,* 2009; Jayaraju *et al.,* 2007; Nagendra *et al.,* 2015). *Ammonia*'s high morphologic variability has led to difficulties in species-level identification, and hence taxonomy of *Ammonia* globally is not correctly described (Hayward *et al.,* 2004). Around 40 species and subspecies of *Ammonia* have been described worldwide (Ellis and Messina, 1940, and supplements) and these different subspecies of *Ammonia* prefer to live in different sub-environment. Hence, a detailed morphological distinction of *Ammonia* could help us in environment interpretation. Jorissen (1988) reported several different morphotypes of *Ammonia parkinsoniana* in the Adriatic Sea and described their environmental adaptation. Petersen *et al.* (2016) observed that different species of *Ammonia* have different pore patterns and densities. It is also evident that interspecies differences in pore parameters exist (Gooday and Alve, 2001). Molecular data have shown that certain pseudo cryptic species of *Ammonia* show apparent differences in porosity (Holzmann and Pawlowski, 1997; Hayward *et al.,* 2004). Hence pore geometry can also be used as a distinguishing morphological character of *Ammonia* species.

The Chilika Lagoon is a typical marginal marine environment. This environment is characterized by high nutrient input with longer residence times and represented by stress-tolerant taxa (Hallock, 2012). Few research studies have been carried out in the Chilika lagoon on the foraminiferal distribution (Rao *et al.,* 2000; Jaylakshmy and Rao., 2006; Sen and Bhadury, 2016; Barik *et al.,* 2019). *Ammonia beccarii* and *Ammonia tepida* are the dominant benthic foraminifera in the Chilika lagoon (Sen and Bhadury, 2016; Barik *et al.,* 2019). Based on a study over 21 months, Bhadury *et al*. (2015) reported *Ammonia* as the dominant form in the Chilika lagoon. They said eight different *Ammonia* species such as *Ammonia beccarii*, *Ammonia sobrina, Ammonia parkinsoniana, Ammonia tepida, Ammonia* sp.1, *Ammonia* sp.2, *Ammonia* sp.3, and *Ammonia pauciloculata.*

Sen and Bhadury, 2016, described three distinct morphotypes of *Ammonia* such as *Ammonia* T2, *Ammonia* T6, and *Ammonia* T10, T representing a molecular type of *Ammonia*. The study is based on a molecular approach as of Hayward *et al.* (2004). Barik *et al.* (2019) observed that *Ammonia* genus with six species such as *Ammonia beccarii*, *Ammonia tepida, Ammonia* sp.1, *Ammonia* sp.2, *Ammonia* sp.3, and *Ammonia pauciloculata* are found in the inner part of the lagoon. The study lacks detailed identification of *Ammonia* species. Mishra *et al.* (2019) documented three *Ammonia* species such as *Ammonia beccarii, Ammonia tepida*, *Ammonia convexa* in the lagoon's central sector but doesn't discuss the detailed morphological description of *Ammonia* species again.

Very few literature studies are available in this lagoon primarily based on molecular approach, but no survey of *Ammonia* species' morpho-characteristic features is recorded. Our study confirms that *Ammonia* is the most dominant taxa in the lagoon with several morphotypes of *Ammonia*. A detailed morphometric survey of the *Ammonia* species has been reported for the first time in this paper with pore geometry considered a distinct morpho parameter.

STUDY AREA

Chilika (19˚28'–19˚54' N; 85˚05'–85˚28' E), the largest brackish lagoon of Asia is situated along the Odisha coast on the east coast of India and connected to the Bay of Bengal. The Chilika Lagoon formed approximately 6000-8000 years ago due to sea-level changes (Gupta *et al.,* 2018). The freshwater influx into the lagoon mainly originated from the Mahanadi river's distributaries (Gupta *et al.,* 2018). The lagoon is bordered on its northern and eastern sides by a catchment region of 4406 km^2 which provides an additional freshwater inflow source (Panigrahi *et al.,* 2009). The northwestern monsoon which induces increased precipitation along the entire northeast coast of India also influences the Chilika lagoon. These can be evidenced by examining the lagoon's hydrological condition as the lagoon's watershed area increased from 704 km^2 in pre-monsoon (March to June) to 1020 km^2 during monsoon season (Gupta *et al.,* 2008). The lagoon has experienced a high level of salinity fluctuations due to seasonal flooding, siltation, choking of sea connecting mouth. The lagoon has been subjected to numerous openings and closings (Subrahmanyam *et al.,* 2006) and it is connected to the Bay of Bengal by three outlets of two were naturally formed in 2008 and 2012 (Sahu *et al.,* 2014) and another artificially created in September 2000 near Satapada (Sipakuda). Based on the lagoon's overall hydrologic condition, depth, and previously reported biological diversity; the lagoon can be subdivided into Southern, Central, Northern, and the Outer Channel (Sahu *et al.,* 2014). The central sector (370 km^2) lies between the northern and the southern sectors and is dotted with islands. The average water depth in the lagoon's central region is 2 to 3 m (Sen and Bhadury, 2016) and average salinity varies between 0.05–34.9 PSU (Patra *et al.,* 2010).

MATERIALS AND METHODS

A 70 cm (70 cm long PVC pipe and inner diameter 6 cm) long core has been collected from the lagoon's central part (Fig. 1). The core was sliced at every 1 cm interval after retrieval. The sliced samples were wet sieved through a 63µm sieve and oven-dried at 50°C in the laboratory. The sediment samples were split into the smallest portions with the help of a micro splitter, and 1gram of sediment was examined under NIKON SMZ 1000 stereo zoom microscope. All the specimens from split amounts of each sediment sample were counted in the size fraction of >63 µm. The abundance of *Ammonia* species was documented in the upper 10 cm of the core then the total foraminiferal number and the populations of *Ammonia* species, decreased. Hence a total of 60 specimens of *Ammonia* were selected from the upper 10 cm of the core for morphometric analysis. The 26 morpho parameters measurements of *Ammonia* specimens were carried out with Image Analysis software associated with Leica DM 2700 P optical microscope. The parameters are selected based on the study of Hayward *et al.* (2004). *Ammonia's* selected specimens were examined under Scanning Electron Microscope (Carl Zeiss Evo-18) at Jadavpur University, India for detailed identification and illustration. The foraminifera at the genus level was identified based on Loeblich and Tappan (1987), and species-level identification was made based on Jorissen (1988), Holzmann and Pawlowski (2000), Ghosh *et al.* (2014) and Barbieri and Vaiani (2018).

For pore pattern study a relatively flat surface was chosen on the last chamber of the species, and microphotographs were taken at 10000X magnifications. The number of pores (total number of pores counted in the measurement frame), pore density (the number of pores per surface area of measurement frame), porosity, and pore diameter (the mean surface diameter of all pores) were determined from image processing software ImageJ (Schneider *et al.,* 2012). The manual corrections were also done according to pore measurement techniques (Peterson *et al.,* 2016). The frame size with a surface area of 562 µm^2 [23.7 µm x 23.7 µm] has been taken as it is the best suitable frame for representation (Peterson *et al.,* 2016).

Both the bivariate and multivariate analysis was performed on various morphological characters of *Ammonia*. Q-mode cluster analysis (Unweighted pair-group using arithmetic averages of Bray-Curtis distance) was performed to generate a dendrogram classification of all the selected specimens. Data matrices were prepared for multivariate statistical analyses using measured morpho parameters of 60 specimens. The validity of clustering was examined by calculating the cophenetic correlation for each clustering output (highly reliable clustering schemes are indicated by values >0.7). Similarity percentages (SIMPER) analysis was performed with the Bray-Curtis distance on the standardized data matrices with samples grouped following the clustering outputs, to identify the contribution of different morpho parameters dissimilarity between clusters. Analysis of similarities (ANOSIM) provides a way to test statistical significance between other cluster groups. Computation of

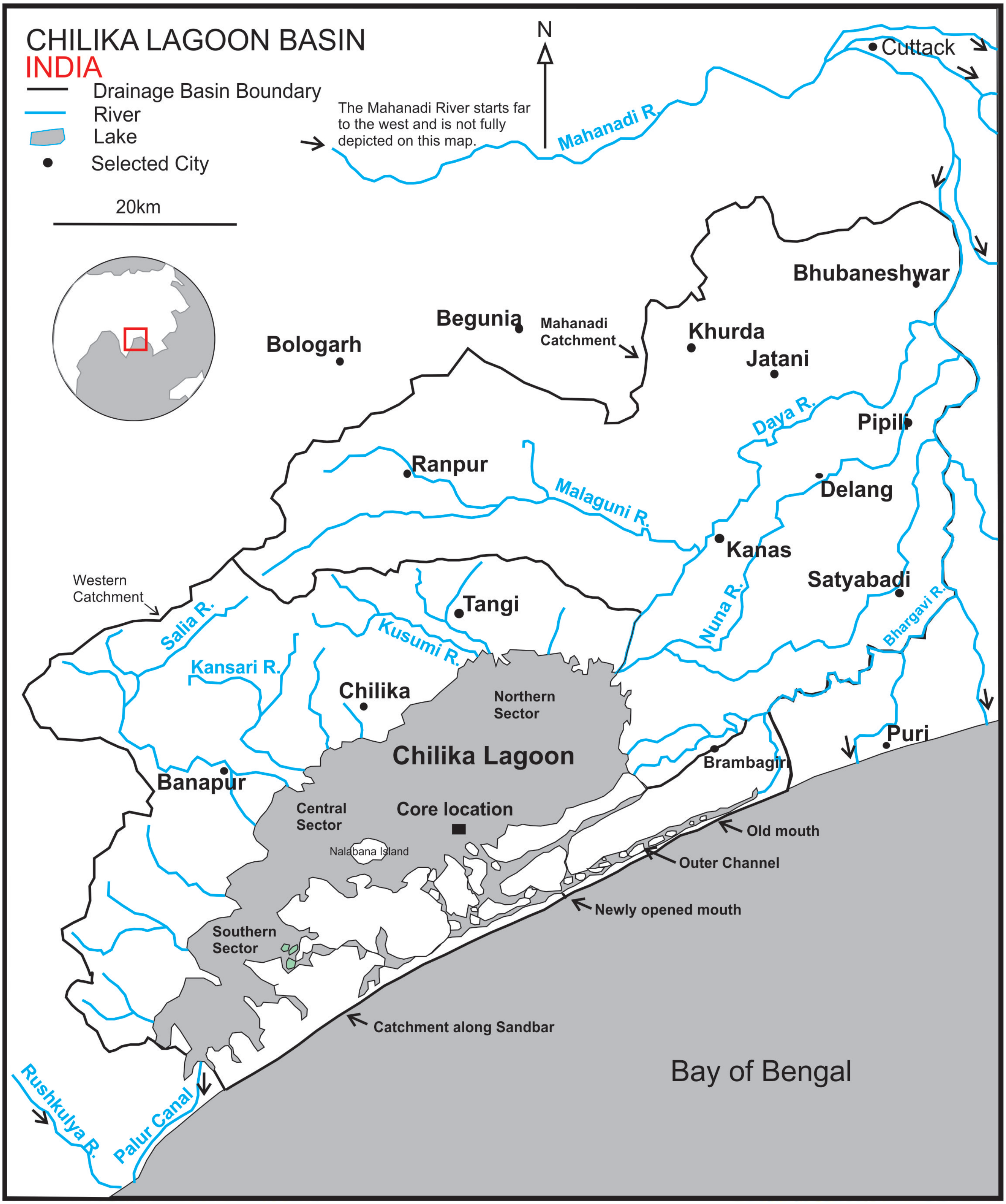
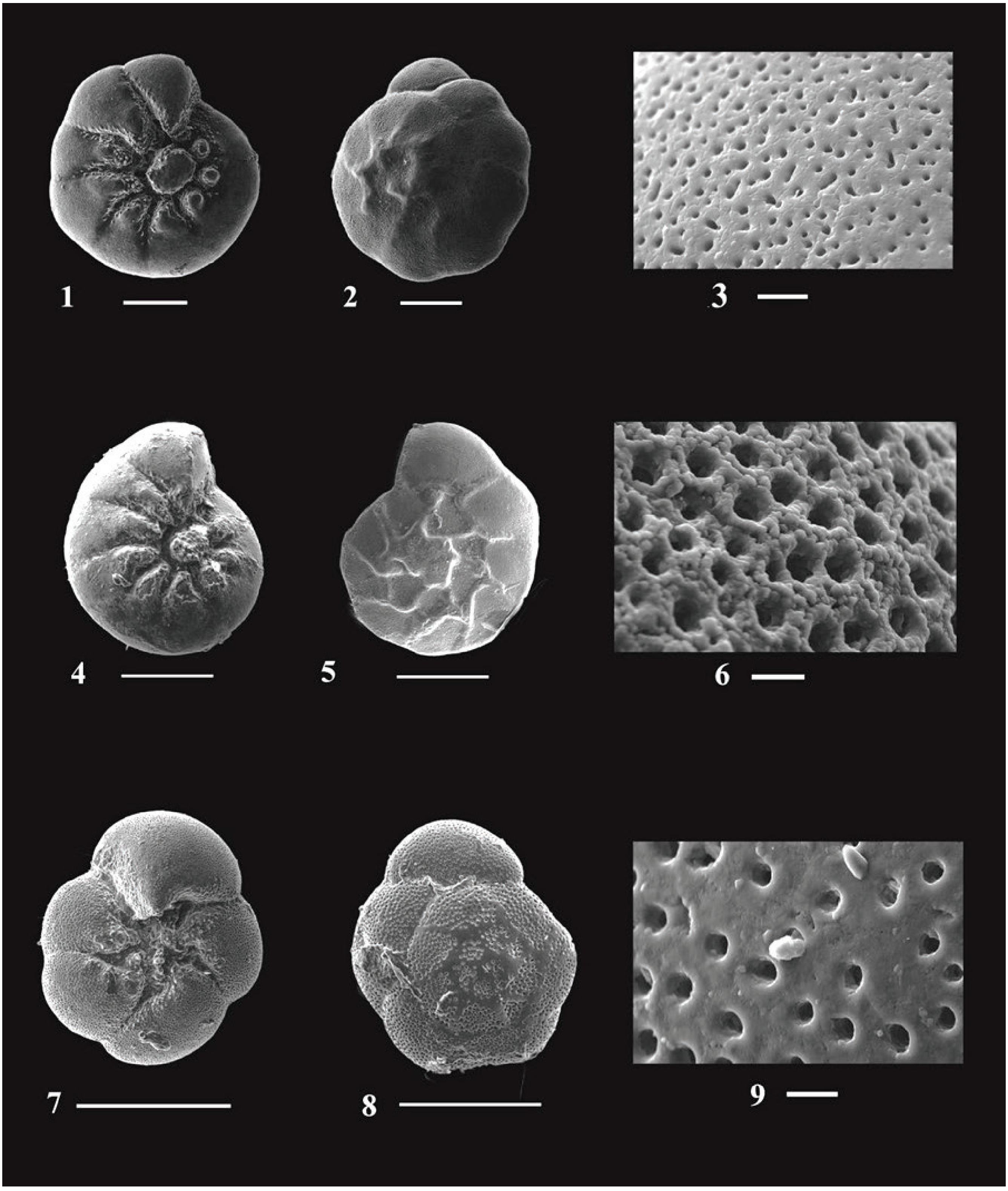


Fig. 1. Chilika lagoon basin, red circle, indicates the core collection location in the central sector of the lagoon.

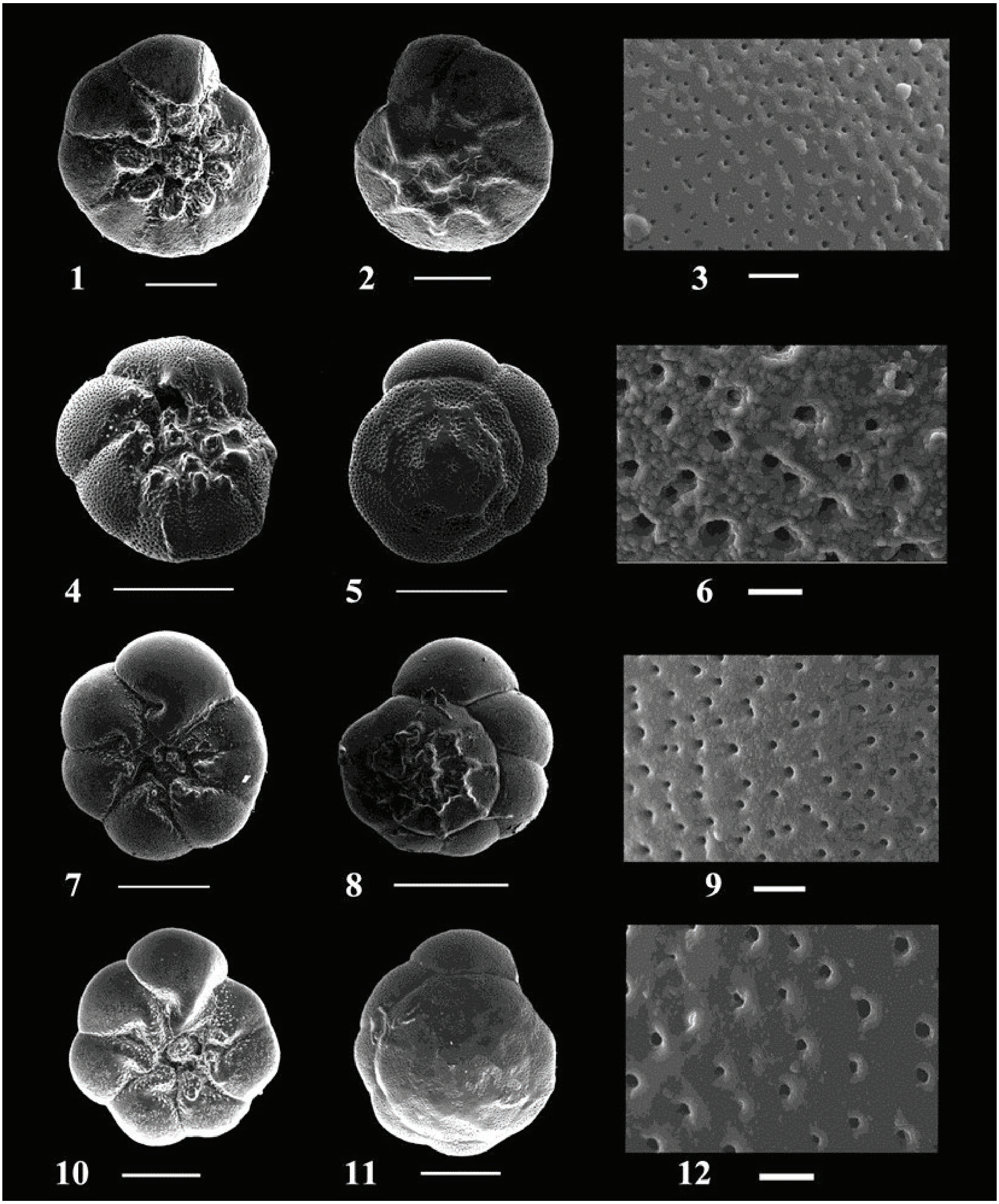
Plate I



EXPLANATION PLATE I

1. *Ammonia sobrina* (UV) 100 µm; 2. *Ammonia sobrina* (SV) 100 µm, 3; *Ammonia sobrina* (pores) 5 µm; 4. *Ammonia parkinsoniana* (UV) 100 µm; 5. *Ammonia parkinsoniana* (SV) 100 µm, 6. *Ammonia parkinsoniana* (pores) 5 µm; 7. *Ammonia tepida* (UV) 100 µm; 8 *Ammonia tepida* (SV) 100 µm; 9. *Ammonia tepida* (pores) 5 µm. (UV-Umbilical view, SV- Spiral view).

Plate II



EXPLANATION PLATE II

1. *Ammonia* cf *sobrina* (U.V.) 100 µm; 2. *Ammonia* cf *sobrina* (S.V.) 100 µm; 3. *Ammonia* cf *sobrina* (pores) 5 µm; 4. *Ammonia tepida* morphotype 1 (U.V.) 100 µm; 5. *Ammonia tepida* morphotype 1 (S.V.) 100 µm; 6. *Ammonia tepida* morphotype 1 (pores) 5 µm; 7. *Ammonia parkinsoniana* forma *tepida* morphotype 2 (U.V.) 100 µm; 8. *Ammonia parkinsoniana* forma *tepida* morphotype 2 (S.V.) 100 µm; 9. *Ammonia parkinsoniana* forma *tepida* morphotype 2 (pores) 5 µm; 10. *Ammonia parkinsoniana* forma *tepida* morphotype 4 (U.V.) 100 µm, 11. *Ammonia parkinsoniana* forma *tepida* morphotype 4 (S.V.) 100 µm; 12. *Ammonia parkinsoniana* forma *tepida* morphotype 4 (pores) 5 µm. (UV-Umbilical view, S.V.- Spiral view).

this cluster analysis, SIMPER analysis, and ANOSIM analysis was performed with the software PAST (PAleontological STatistic-ver.3; Hammer *et al.,* 2001).

RESULTS AND DISCUSSION

Selected specimens from the core sample were measured and morphologically coded for numerical analysis. All the measurements and codings were obtained from three views ~umbilical, spiral, and close-up view of the last chamber (n) on umbilical (for pore pattern study). In case the final chamber of the specimen is broken, the penultimate chamber (n-1) has been chosen. A total of 26 morphological characters were measured: a mixture of dichotomous, quantitative, ratios, and some qualitative assessments (Fig. 2.1, Fig. 2.2, Appendix 1; Table 1). A summary table of the main characteristics of different morphotypes of *Ammonia* identified from Chilika lagoon was given in Table 4. Pore patterns are also considered as morpho parameters and detailed pore patterns analysis of studied *Ammonia* specimens is provided in Appendix 1, Table 2.

Multivariate cluster analysis

The multivariate cluster analysis (Fig. 3) helps to group the specimens of *Ammonia* having similar characteristics. Cluster analysis of taxon 26 morphometric parameters shows seven clusters that indicate different forms of *Ammonia* are present in the Chilika lagoon. The ANOSIM analysis performed on raw data showed a higher dissimilarity between these clusters (R-value 0.8333). This dissimilarity between groups is also statistically significant (p-value 0.0001). SIMPER analysis shows that Greatest Spiral Diameter (GSD) plays the primary contributor (>35%) for the dissimilarity between these clusters. SIMPER on the raw data presents the average value of quantitative analysis in each set (Appendix 1, Table 3). Typical specimens representing each group of *Ammonia* and their pore patterns are illustrated on Plate I and Plate II.

Cluster 1: All the *Ammonia* specimens in cluster 1 are characterized by a big test (average value of test size is 346 µm), moderate proloculus (average 42.9 µm) to the test size. The number of chambers in the last whorl is high (9 to 10). The test periphery is smooth and entire. The spiral side of the test is much more convex than the umbilical side. The sutures are curved. The average radial sutural furrow length (RFL) is 55.9 µm. A large number of sutures are present, which are thickened and depressed on the umbilical side. The wall is calcareous, finely perforate radial in structure, irregular granules present on the umbilicus side, and a single large prominent boss present. The test has sharply pointed folia and thickened calcite deposit present on folia in the umbilical side. The spiral side raised thickened calcite developed over radial sutures of the last whorl and the central spiral area. The average angle value between radial and spiral sutures is 80.2˚, and folium angle value is 73.2˚ on average. The mean number of chambers in each whorl and mean diameter of the chamber is also very high (varies between 150-170 µm).

Pore patterns: This species of *Ammonia* consists of numerous small circular pores hence pore density is also high (0.0231Np/µm^2; Np represents the number of pores present in the measured frame), few large elliptical pores are also present. (Plate I, Fig. 3)

The specimens' morphological characteristics resemble *Ammonia sobrina* (Shupack, 1934), and hence this group of representatives is assigned to *Ammonia sobrina* (Plate I, Figs. 1, 2).

Cluster 2: The specimens in this cluster have a test size not as big as cluster 1 (test size average value is 271 µm) and proloculus value is also small (average 26.3 µm). These specimens consist of a single prominent boss, 7 to 9 chambers in the final whorl. The test outline of these specimens is smooth. The umbilicus is relatively closed, and the spiral side is more convex than the umbilical side. A thick calcite deposit was present on the folia and much-thickened calcite developed over radial sutures of the last whorl and the central spiral area. The average angle value between radial and spiral sutures decreases, but the folium angle value is similar to the previous cluster's specimens. The mean number of chambers in each whorl is high, and the chamber's mean diameter is varying between 100-110 µm.

Pore patterns: The pore patterns of these specimens are quite similar to *A. sobrina*. They also consist of small, numerous circular pores. The pore density is too high (0.0196 Np/µm^2; Np represents the number of pores present in the measured frame). (Plate II, Fig. 1).

This foraminifera show similar characteristics to *A.sobrina* but differ from its small test size, less chamber number, and very thick calcite deposits over radial sutures and central spiral area. Hence, it is considered a separate *Ammonia* species (*Ammonia* cf. *sobrina*) (Plate II, Figs. 2,3).

Cluster 3: The specimen's test size varies from 250-290 µm, and its average value is 242 µm, and proloculus size is relatively small for test size (average 23.9 µm). The test has 8-9 chambers present, circular in outline, rounded periphery, medium thickened calcite deposit present on folia in the umbilical side. On the spiral side, very thick calcite developed over the last whorl's radial sutures and the central spiral area. The reticulate pattern of calcite riblets was observed over the central spiral area. The angle between radial and spiral sutures is very high (average 94.3˚) though folium angle value remains the same. The mean number of chambers in each whorl and the chamber's mean diameter are also very high (100-110 µm).

Pore patterns: They are characterized by distinct pore structure, egg holder-like pore geometry with prominent calcitic rims along the boundary of the circular pores (Plate I, Fig. 6).

This group is assigned to *Ammonia parkinsoniana* as it has similarities with the *Ammonia parkinsoniana* reported by Jorissen (1988) and Barbieri and Vaiani (2018) (Plate I, Figs. 4, 5).

Cluster 4: The specimens are characterized by medium-sized test (190 -220 µm, avg 208 µm), 6- 8 chambers in the

Table 1. Values of the analysed variables for each specimen.

Serial No.	Gsd (µm)	Prol (µm)	Chwh1	Chwh2	Sw (µm)	Fol (degree)	Rad (degree)	Du/Gsd	Rfl/W	Maxbos	Maxbos/Gsd	Ch/Wh	Lc/Wc	(Gsd-Prol)/Wh
1	195.37	21.87	7	8	107.92	76.59	65.91	0.19	0.31	11.2	0.06	7.5	1.92	86.75
2	267.21	26.4	7	9	106.16	50.88	54.15	0.25	0.44	54.27	0.20	8	2.03	120.41
3	213.91	32.96	6	8	80.52	56.22	68.71	0.36	0.42	61.49	0.29	6	1.16	90.48
4	167.91	9.42	8	6	54.53	52.48	94.22	0.20	0.44	20.6	0.12	8.5	1.22	79.25
5	287.28	28.84	6	7	117.95	77.11	44.62	0.30	0.46	60.76	0.21	6.5	2.72	129.22
6	274.43	15.74	7	9	79.39	70.34	89.53	0.27	0.29	46.09	0.17	8	1.55	129.35
7	233.81	21.17	5	7	176.18	63.46	77.37	0.2	0.30	0	0	5.33	1.7	70.88
8	357.91	40.61	9	10	93.06	75.75	101.27	0.21	0.52	49.58	0.14	9.5	2.69	170.65
9	263.35	21.08	8	8	122.36	82.92	81.09	0.30	0.26	0	0.00	8	1.66	121.14
10	182.06	9.79	9	8	58.65	98.67	68.41	0.11	0.12	0	0.00	7.5	2.27	86.14
11	172.01	10.78	8	8	66.42	74.8	81.39	0.12	0.17	0	0.00	9	2.95	80.62
12	173.95	27.85	7	7	84.09	60.25	66.96	0.12	0.10	8.62	0.05	7	3.01	73.05
13	289.65	29.48	6	7	119.95	72.17	45.63	0.19	0.36	0	0.00	6.5	2.81	130.09
14	288.05	28.84	6	7	120.59	69.75	46.57	0.22	0.34	53.26	0.18	6.5	2.54	129.61
15	176.74	23.49	7	8	112.58	66.53	77.97	0.12	0.00	0	0.00	8	2.29	76.63
16	244.62	16.4	6	9	47.88	77.62	78.607	0.33	0.00	68.02	0.28	7.5	1.66	114.11
17	232.92	19.71	5	8	42.19	95.44	87.63	0.25	0.36	50.97	0.22	5.5	1.46	106.61
18	222.89	26.73	7	7	63.08	66.3	82.41	0.22	0.43	13.69	0.06	6.5	1.58	98.08
19	185.15	15.44	7	6	43.68	65.6	106.81	0.28	0.35	0	0.00	5.67	2.88	56.57
20	223.05	19.52	6	6	89.23	73.64	92.57	0.25	0.70	0	0.00	8		101.77
21	225.76	26.09	0	8	65.36	85.28	81.67	0.34	0.43	47.49	0.21	4	1.14	99.84
22	240	37.82	7	8	73.85	84.62	111.94	0.30	0.37	54.39	0.23	6	1.92	101.09
23	176.03	9.26	5	7	44.32	71.38	66.66	0.14	0.48	8.49	0.05	5.5	2.74	83.39
24	159.73	8.24	6	7	63.62	70.14	71.24	0.04	0.16	0	0.00	6.5	1.27	75.75
25	216.3	22.19	6	6	55.71	101.32	97.11	0.20	0.19	0	0.00	6	1.06	97.06
26	332.99	39.41	8	10	109.72	85.71	68.36	0.26	0.44	74.45	0.22	7.67	1.09	150.53
27	227.71	17.53	8	7	91.22	90.75	151.34	0.17	0.64	0	0.00	7	1.67	105.09
28	248.67	27.11	8	9	97.69	59	114.25	0.32	0.21	73.64	0.30	7.33	3.42	73.85
29	340.79	53.85	7	10	157.1	57.13	74.91	0.29	0.24	59.78	0.18	8.5	1.63	128.47
30	196.72	16.68	8	8	62.29	61.5	87.21	0.37	0.23	57.04	0.34	9	1.51	75.02
31	152.52	26.19	6	5	68.44	95.91	74.52	0.13	0.38	0	0.00	5.5	1.83	63.17
32	197.23	13.6	5	6	84.85	75.01	62.31	0.22	0.49	10.17	0.05	5	1.83	91.82
33	214.05	10.51	6	8	83.27	80.89	89.25	0.31	0.26	46.98	0.22	7	1.13	101.77
34	270.54	26.38	5	7	112.55	83.28	59.04	0.22	0.22	37.19	0.14	6	1.98	122.08
35	179.7	15.66	8	7	60.81	59.18	97.86	0.10	0.00	0	0.00	7.5	6.32	82.02
36	193.25	15.86	5	8	61.92	75.67	86.12	0.40	0.27	59.38	0.36	6.5	1.46	73.70
37	151.27	9.79	6	6	43.31	85.37	95.89	0.14	0.27	0	0.00	6	1.55	70.74
38	189.2	22.11	8	7	84.46	86.03	92.55	0.22	0.19	11.54	0.06	7.5	1.31	83.55
39	211.78	9.21	6	8	81.5	87.18	86.54	0.31	0.22	47.55	0.22	7	1.16	101.29
40	268.2	21.1	8	8	136.38	79.15	37.87	0.20	0.21	44.09	0.16	7	2.26	123.55
41	179.26	25.94	8	8	68.24	58.23	69.96	0.06	0.00	0	0.00	9	1.14	76.66
42	244.94	18.42	6	9	47.58	69.82	78.9	0.34	0.42	69.74	0.28	7.5	1.22	113.26
43	148.03	15.77	6	6	40.31	63.39	92.98	0.17	0.46	0	0.00	6	3.07	66.13
44	190.46	23.21	8	7	86.64	57.19	92.15	0.26	0.19	34.105	0.18	7.5	1.31	83.63
45	219.7	23.37	7	9	60.8	66.47	80.14	0.27	0.49	46.37	0.21	6.5	1.65	98.17
46	222.61	16.35	8	7	89.19	78.3	90.57	0.08	0.23	0	0.00	7	1.72	103.13
47	204.69	20.34	7	8	68.45	73.36	92.93	0.28	0.25	35.4	0.17	7.5	1.42	92.18
48	206.62	15.44	8	7	98.51	94.53	81.51	0.16	0.47	0	0.00	7	1.40	95.59
49	212.97	32.96	6	6	81.59	69.17	125.81	0.26	0.33	25.86	0.12	6	1.51	90.01
50	208.26	22.43	7	8	70.54	85.13	104.39	0.26	0.37	23.6	0.11	7.5	1.48	92.92
51	180.71	22.28	9	7	73.92	100.76	79.67	0.22	0.38	7.42	0.04	8	2.61	79.22
52	251.06	29.97	9	9	87.31	66.15	63.59	0.27	0.19	49.26	0.20	9.5	1.40	110.55
53	156.28	23.95	7	6	48.96	66.61	89.13	0.11	0.31	0	0.00	8.33	1.58	44.11
54	156.43	22.59	7	6	46.69	51.58	85.31	0.11	0.00	0	0.00	10	1.54	66.92
55	200.4	18.99	5	8	74.99	62.67	65.8	0.28	0.20	32.09	0.16	6.5	1.38	90.71
56	227.77	32.23	8	6	64.95	70.09	65.12	0.23	0.49	18.52	0.08	9	1.61	97.77
57	254.77	35.02	9	9	96.12	84.64	66.81	0.27	0.00	49.79	0.20	7	1.43	73.25
58	330.91	37.82	9	9	78.46	65.8	70.71	0.28	0.33	66.86	0.20	7.33	1.29	167.70
59	253.21	23.42	8	7	70.87	79.47	69.53	0.22	0.39	0	0.00	7.5	1.66	102.40
60	366.86	42.98	10	9	117.63	81.55	85.69	0.21	0.32	28.84	0.08	9.5	1.53	161.94

last whorl, lobate outline, inflated chambers with a prominent boss in the umbilicus. Curved sutures are present. The mean radial sutural furrow length is 43.1 µm. A weak thick calcite deposit was observed on the folia, over radial sutures of the last whorl and the central spiral area. The angle between radial and spiral sutures is relatively high. The mean number of chambers in each whorl is medium, and the chamber's mean diameter is low (90-100 µm).

Pore patterns: They are characterized by lower pore density (0.0071 Np/µm^2; Np represents the number of pores present in the measured frame) as they have very few circular smaller pores. (Plate II, Fig. 12)

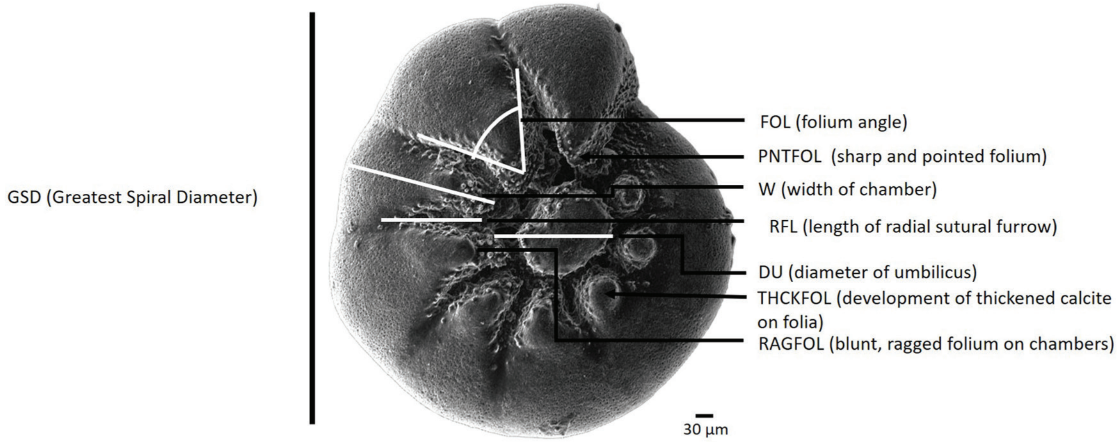


Fig.2.1. Scanning electron microscopic image of the umbilical side of *Ammonia* specimen showing the characters used in morphometric analysis.

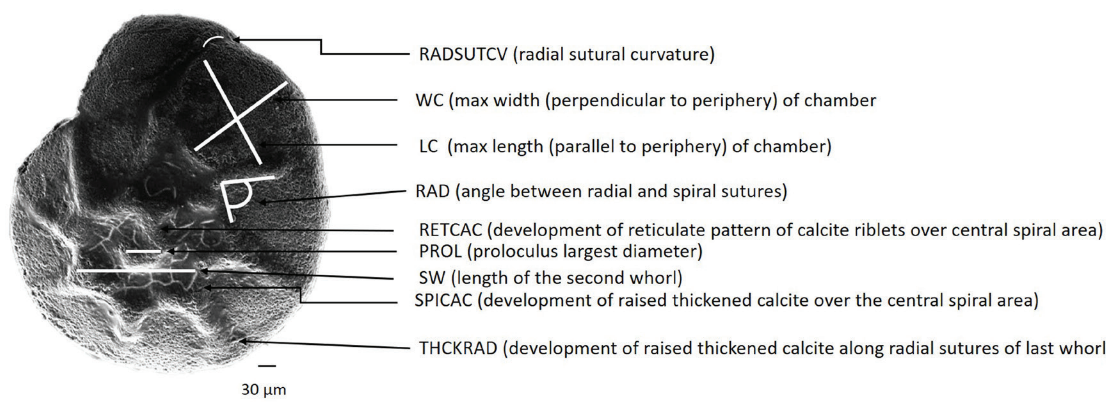


Fig. 2.2. Scanning electron microscopic image of the spiral side of *Ammonia* specimen showing the characters used in morphometric analysis.

These specimens have morphological similarities with *A. parkinsoniana* forma *tepida* morphotype 4 of Jorissen 1988, pl.10, fig 2-3. The pore patterns of our studied samples are also similar to the morphotype of *A. parkinsoniana* (Jorissen, 1988, pl. 10, fig. 2d). Still, they have some calcitic deposits on the spiral side of the test. The morphological parameters are quite similar to *A. tepida* but differ with the presence of the prominent boss. Surface ornamentation is not an essential morpho parameter for distinguishing *Ammonia*'s different morphotypes (Hayward *et al*., 2004). Hence, we considered it a morphotype of *A. parkinsoniana* as described by Jorissen, 1988 (Plate II, Figs. 10, 11).

Cluster 5: In this cluster, *Ammonia* individuals are moderate-sized (200- 250 µm, average 225 µm) with medium-sized proloculus (average 21.7 µm), 6-7 chambers in the last whorl. The test outline is lobate, with enormously inflated chambers and a broadly rounded periphery. The umbilicus is depressed with no prominent boss but with few calcite deposits. No thick calcite deposit is present on folia but thick raised calcite presents over the central spiral area, and a reticulate pattern of calcite riblets occurs over the central spiral area. The average radial sutural furrow length is 4.03 µm. The average value of the angle between radial and spiral suture and folium angle is 91.3˚ and 81.8˚ respectively.

Pore pattern: This morphotype of *Ammonia* has smaller circular pores. It has a moderate pore density (0.0107/ Np/ µm^2; Np represents the number of pores present in the measured frame). (Plate II, Fig. 9).

The form has similarity to the *Ammonia parkinsoniana* forma *tepida*, morphotype 2 of Jorissen 1988, pl.7, fig. 2-4. The pore patterns are also quite similar to the morphotypes described by Jorissen, 1988, pl. 8, fig. 6. But, thick raised sutures in the spiral area are also a distinctive feature. Again, here the surface ornamentation was not considered as separate morpho parameters. Hence, it is a morphotype of *Ammonia parkinsoniana* (Jorissen, 1988). (Plate II, Figs. 7, 8).

Cluster 6: The specimens have a relatively small test size (average test size is 178 µm) with a short proloculus size (average 18.5 µm), 7-8 chambers present in the last whorl. The chambers in the final whorl are rounded, periphery

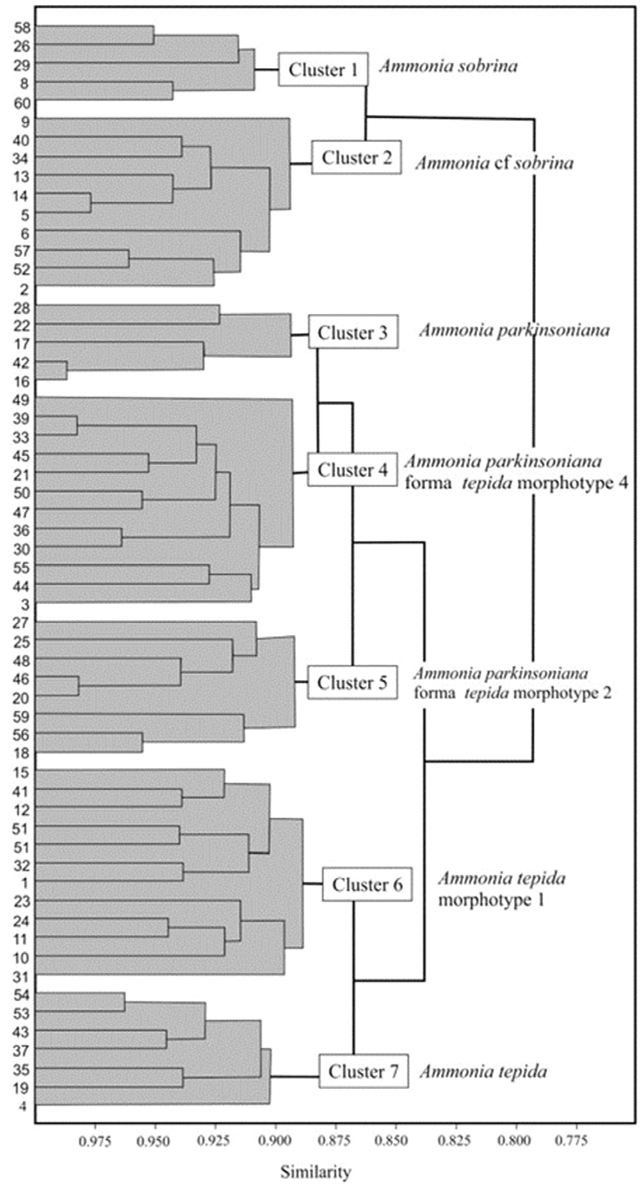


Fig. 3. Multivariate cluster analysis based on the 26 measured morphoparameters of *Ammonia* specimens.

Table 2. Pore patterns measurement.

	Total no of pores (Np)	*Total surface area covered by pores (µm)²*	*mean surface pore area (µm)²*	*Porosity*	*Pore density (Np/(µm)²*
Ammonia sobrina	13	49.212	3.786	8.76	0.0231
Ammonia cf sobrina	11	16.379	1.489	2.916	0.0196
Ammonia parkinsoniana	5	47.352	9.47	8.429	0.0089
Ammonia tepida	3	26.254	5.251	4.673	0.0053
***Ammonia tepida* morphotype 1**	2	31.784	15.892	5.658	0.0036
***Ammonia parkinsoniana forma tepida,* morphotype 2**	6	31.997	5.333	5.696	0.0107
***Ammonia parkinsoniana forma tepida* morphotype 4**	4	20.024	10.012	3.564	0.0071

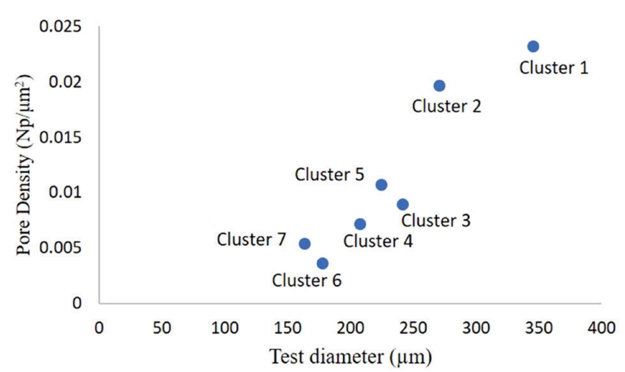


Fig. 4. Scatter plot of test diameter and pore density of *Ammonia* specimens.

slightly lobate in outline and progressively becomes lobate at the end of the last whorl; umbilicus without a central knob. Some specimens show calcite deposits present in the umbilicus region. The radial sutural furrow length is 4.79 µm on average. Thickened calcite deposit present in the folia of some representatives and over the central spiral area. The mean diameter of the chamber is low (varies between 75 to 85 µm).

Pore pattern: These species have large circular pores. Their pore patterns are quite similar to *Ammonia tepida* of Cluster 7 (Plate II, Fig. 6).

This group of species differs from *Ammonia tepida* mainly due to its high chamber number. Therefore, it is considered a separate morphotype of *A. tepida* (*Ammonia tepida* morphotype 1) (Plate II, Figs. 4,5).

Cluster 7: The specimens in this cluster have a small test size (average test size is 169 µm) to other groups and very small proloculus (15-35 µm, average 16.1 µm). The outline is lobate, with enormously inflated chambers and a broadly rounded periphery and 5 to 6 chambers in the last whorl. The umbilicus is depressed without any boss. No thickened calcite deposit is present in the folia and also over radial sutures of the last whorl. Radial sutural furrow length is minimal (avg value is 2.94 µm). The folium angle is also low concerning the other present morphotypes of *Ammonia* (average value is 63.5°) Weak calcite deposit present over the central spiral area. The mean number of chambers in each whorl and mean diameter of the chamber varies from 50- 80 µm.

Pore patterns: These specimens have lower pore density (0.0053 Np represents the number of pores present in the measured frame) as large circular pores characterize it. (Plate I, Fig. 9)

This cluster represents *Ammonia tepida* as it has similarities to species of *Ammonia tepida* reported earlier by

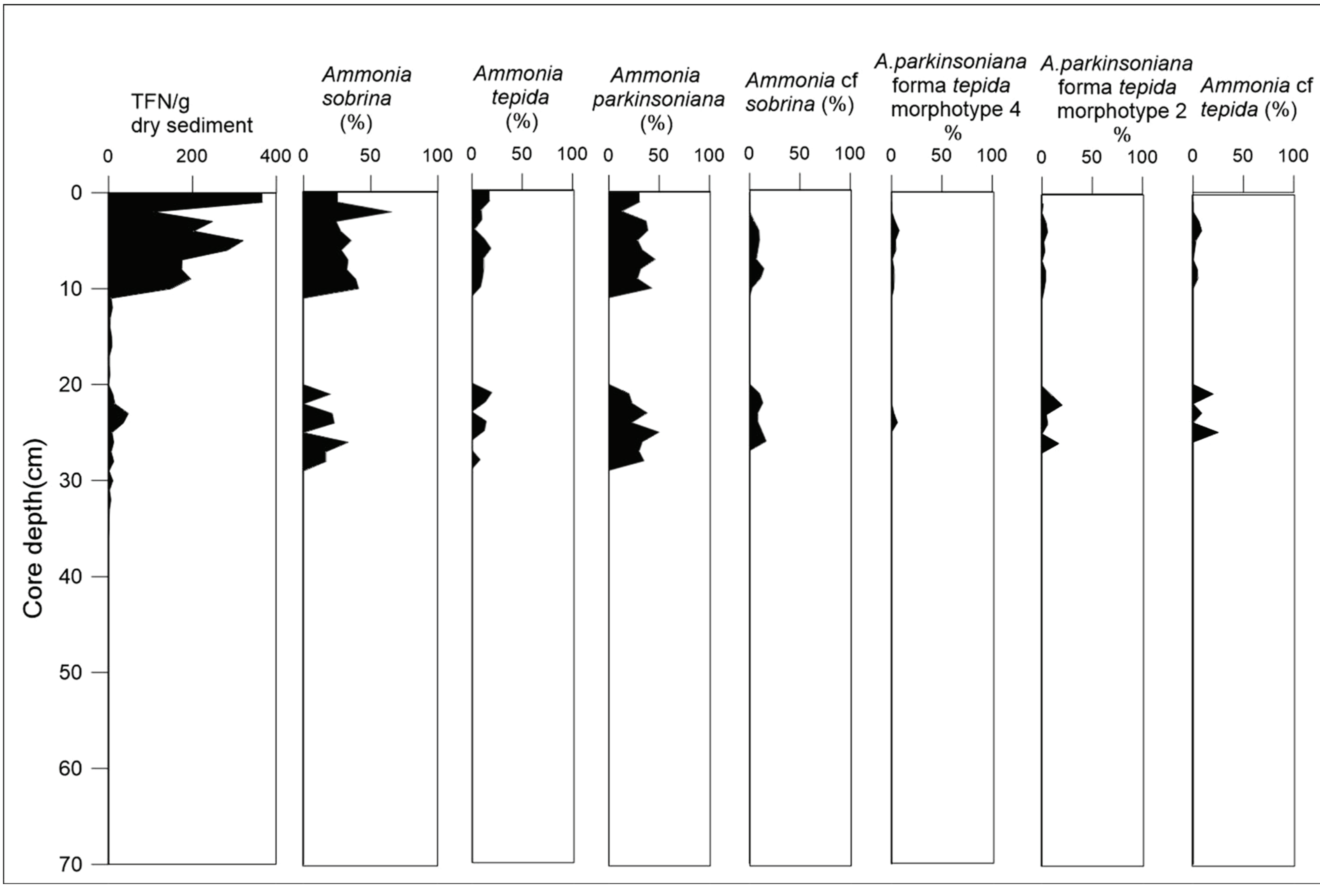


Fig. 5. Vertical distribution of different morphotypes of *Ammonia.*

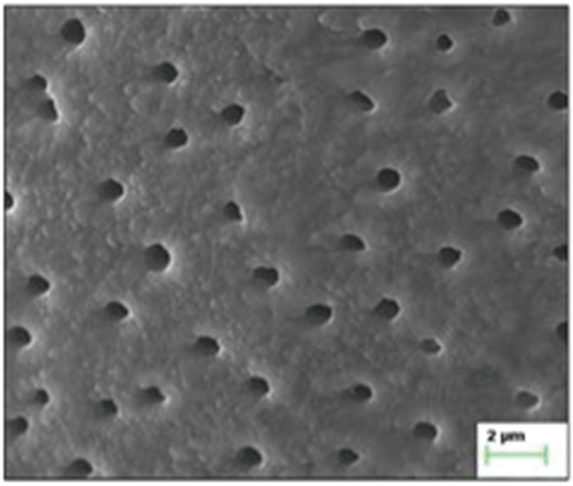
Table 3. Results of the SIMPER analysis performed on the essential measured morphoparameters, showing the relative contribution of each parameter to the determination of clusters and the average composition of each cluster.

Measured Morpho parameters	Av. dissim	Contr %	Cumulative %	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5	Cluster 6	Cluster 7
Test diameter (µm)	6.118	36.15	36.15	346	271	242	208	225	178	164
Radial sutural furrow length (µm)	3.029	17.9	54.04	55.9	39.5	63.4	43.1	4.03	4.79	2.94
Length of second whorl (µm)	2.946	17.4	71.44	111	110	61.8	73.2	77.8	76.5	48.3
Rad (degree)	2.113	12.48	83.93	80.2	58.9	94.3	88.4	91.3	73.1	94.6
Folium angle(degree)	1.421	8.397	92.32	73.2	73.6	77.3	71.7	81.8	77.9	63.5
Prol (µm)	1.014	5.994	98.32	42.9	26.3	23.9	21.1	21.7	18.5	16.1

Jorissen (1988) and Barbieri and Vaiani (2018) (Plate I, Figs. 7, 8).

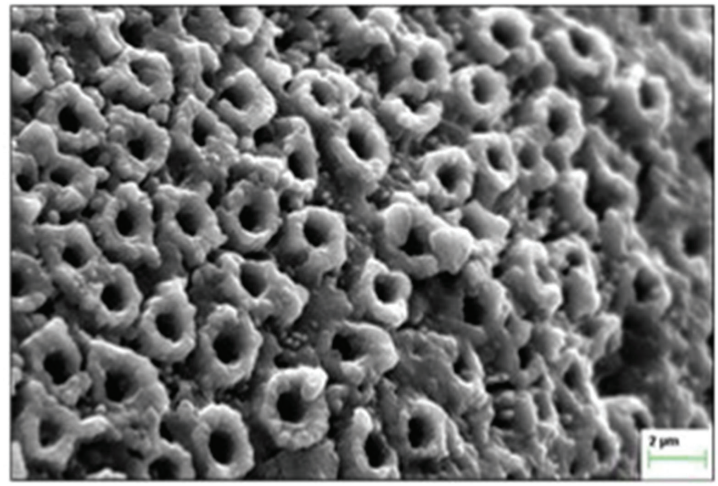
Relationship between pore density and test diameter of different morphotypes of *Ammonia*

In the studied core, the bivariate plot (Fig. 4) between pore density and test diameter of these seven clusters shows that these two variables have a positive correlation. *A. sobrina* (Cluster 1) and *Ammonia* cf. *sobrina* (Cluster 2) both have high test diameters and high pore density. *Ammonia parkinsoniana* (Cluster 3) and its morphotype *Ammonia parkinsoniana* forma *tepida*, morphotype 2 (Cluster 5), and *A. parkinsoniana* forma *tepida* morphotype 4 (Cluster 4) have a quite average test diameter and low pore density. *Ammonia tepida* (Cluster 7) and its morphotype (Cluster 6) have low pore density as they contain few large pores and low-test diameter. Cluster 4 and Cluster 5 are morphotypes of *Ammonia parkinsoniana* (Cluster 3), and Cluster 6 is the morphotype of *Ammonia tepida* (Cluster 7) based on the similarity coefficient of 0.875 (Fig. 3). Laboratory cultures of *Ammonia beccarii* show an increase in pore size under low-oxygen conditions (Moodley and Hess, 1992). Low pore density (increase in pore size) in morphotype of *A. tepida* and moderate pore density in different morphotypes of *A. parkinsoniana* and their occurrence in a particular part of the core can be related to environmental changes in the lagoon during certain phases.



(A)

Fig. 6 (A). Circular pore patterns of *Ammonia* sp.



(B)

Fig. 6 (B). Peculiar egg holder type pore patterns in *Ammonia parkinsoniana.*

Distribution of different morphotypes of *Ammonia* along with the core

The vertical distribution plot (Fig. 5) of the core shows that total foraminiferal abundance is high in the core's upper 10 cm. Total foraminiferal number per 1 gram of dry sediment varies between 107 and 367 in the core's upper 10 cm. Then foraminiferal abundance decreases rapidly (TFN/g varies between 0 and 9). Between 20 and 10 cm intervals, the foraminiferal count is low ranging between 10-47. In the interval from 32-20 cm of the core, foraminifera's sporadic occurrence was noticed at a few depths. The most dominant genus in the core is *Ammonia* (> 90%) with a low occurrence of agglutinated species such as *Miliammina fusca* and *Trochammina advena* and calcareous *Cribroelphidium* species(< 10%). The main foraminiferal taxa are *A. sobrina* and *A. parkinsoniana*. Other morphotypes of *A. parkinsoniana* and *A. tepida* are present in the core in low percentage. The presence of morphotypes of *A. parkinsoniana* and *A. tepida* is noticed in the upper 10 cm of the core, but it is not in a significant number (< 10%). The percentage of different morphotypes increases between 20 cm to 32 cm (varies between 10 to 20%). The literature review suggested the physicochemical parameters of Chilika Lagoon have gone through several changes due to threats from both natural and anthropogenic pressures over the past 80-85 years (Patra *et al.,* 2010). Foraminiferal species attempt to adapt to the changing environment by changing their external morpho parameters (Weinkauf *et al.,* 2016). Before 2000 A.D. the lagoon had experienced several environmental problems such as siltation, sea mouth choking and salinity decrease that led to the scarce presence of foraminifera. After artificial dredging in 2000 A.D., the ecological condition of the lagoon improved and it is well supported by the gradual increase of the foraminiferal number and species diversity. Hence, the presence of different morphotypes of *Ammonia* in the core can be related to the lagoon's rapid environmental changes during the last few decades (Dasgupta *et al.,* 2020; under review).

Table 4. Summary table of important diagnostic features of different morphotypes of *Ammonia*.

Different morphotypes of *Ammonia*	Diagnostic features
Ammonia sobrina Shupack, 1934	Large test and large proloculus, 9-10 chambers in the last whorl, a single large boss, numerous small circular pores present in the test
Ammonia cf. *sobrina*	Large test and small proloculus, 7-9 chambers in the last whorl, a single large boss, numerous small circular pores present in the test
Ammonia parkinsoniana d'Orbigny,1839	Relatively small test and small proloculus, 8-9 chambers in the last whorl, a small prominent single boss present, numerous small circular pores present in the test
Ammonia tepida (Cushman, 1926)	Small test and very small proloculus, 5-6 chambers in the last whorl, Depressed umbilicus with no boss present, large circular pores present
Ammonia tepida morphotype 1	Small test and small proloculus, 7-8 chambers in the last whorl, Depressed umbilicus without a central knob but calcite deposit present in the umbilicus region, large circular pores present
Ammonia parkinsoniana forma *tepida*, morphotype 2	Medium size test and medium-size proloculus, 6-7 chambers in the last whorl, Depressed umbilicus with no boss present, few calcite deposits present in the umbilicus region, smaller circular pores present
Ammonia parkinsoniana forma *tepida*, morphotype 4	Medium size test and medium-size proloculus, 6-8 chambers in the last whorl, a prominent boss present in the umbilicus region, very few circular smaller pores present

New pore pattern of *Ammonia* species

The pores are formed on benthic foraminifera test at the early stage of their chamber formation (Banner and Williams, 1973; Berthold, 1976; Hemleben *et al.,* 1977; Spero, 1988). Most of the calcareous foraminifera have pores both in the umbilical and spiral sides. They use their pores on the test for gas exchange. Many laboratory studies correlate the pores on benthic foraminifera test (mainly the pore size and pore density) and ambient bottom water oxygen concentrations (Glock *et al.,* 2011; Kuhnt *et al.,* 2013). *Ammonia* also contains pores in the entire test. The pores pattern in *Ammonia* of Chilika lagoon shows that different species have different pore pattern types. Most of *Ammonia* species present in the Chilika lagoon are characterized by a circular pore (Fig. 6A). The pore size differs, but their shape remains the same though few elliptical pores can also be present just like the pore patterns of *Ammonia* sp. 1 observed by Holzmann (2000). But in the lagoon completely new pore pattern was observed in *A. parkinsoniana* (in both live and dead specimens). In the last and penultimate chamber, egg holder-like pore geometry with prominent calcitic rims was noticed (Fig. 6B). It has been observed that these calcitic rims are cloudy white, but not transparent. So, it is not the calcite made by foraminifera. Calcite veneer of the topmost layer of the test is dissolved, and further overgrown calcite crystals occurred around the pores of this species. Hottinger (2006) described a similar feature called egg holder as an internal pore structure in a larger benthic foraminifer *Heterostegina depressa*. Egg holders are considered to be the pores enlarged to a cup with polygonal rims with or without spinose projections at the junction with the adjacent pores. But in our study, we reported this calcitic structure as an external feature, not an internal feature described by Hottinger (2006). This type of pore pattern is the first time documented in any smaller benthic foraminifera. The reason behind the development of calcitic rim along the pores is still a matter of study.

CONCLUSIONS

The cluster analysis of morphometric parameters of the *Ammonia* documents four different species- *Ammonia sobrina*, *Ammonia* cf. *sobrina*, *Ammonia parkinsoniana*, and *Ammonia tepida*. Two distinct morphotypes of *Ammonia parkinsoniana* and one morphotype of *Ammonia tepida* are also recorded in the Chilika lagoon. Different morphotypes of *A. parkinsoniana* exist in the upper 10 cm and from 20-32 cm of the core. The study of pore geometry of these seven different *Ammonia* types shows that *A. sobrina* and *Ammonia* cf *sobrina* have high pore density. These two species are characterized by two different shapes of pores ~ one circular and another elliptical. *A. parkinsoniana* has a typical pore pattern similar to egg holder types as a thick calcite deposit present along the pores' boundary. *A. tepida* and its morphotypes have fewer larger circular pores with low pore density. The detailed study of *Ammonia* species' pore pattern along the core and its relation to change in environmental conditions of the lagoon is the future scope of this study.

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